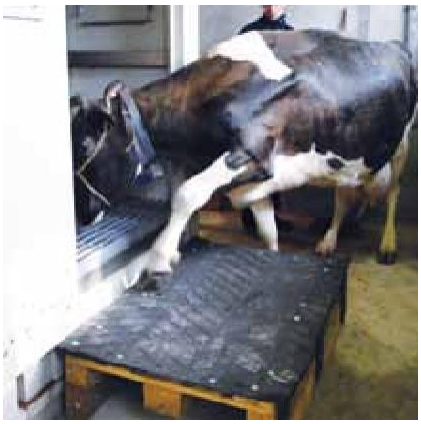
Technical Manual on Respiration Chamber Designs



September 2018

Editors

The original manual (Version February 2014) has been edited by Cesar Pinares and Gary Waghorn. The manual (Version September 2018) has been updated with the addition of Chapter eight by Sinead Leahy and Kate Parlane.

Acknowledgements

This manual has been commissioned by the New Zealand Government to support the goals and objectives of the Global Research Alliance on Agricultural Greenhouse Gases, but its contents rely heavily on the contributions from individual scientists in Alliance member countries. The participation of these scientists and their institutions is gratefully acknowledged and warm thanks are extended for their contribution to this document.

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Introduction

Anthropogenic emissions of methane (CH_4) are estimated to be responsible for about 30% of the warming caused by increased greenhouse gas concentrations in the atmosphere. Enteric emissions of CH_4 from farmed ruminants, that is, those arising from the fermentation of feed in the digestive tract is the single most important source of anthropogenic CH_4 emissions.

Measuring enteric CH_4 emissions is challenging as emissions arise from a large number of point sources (that is, individual animals), and these point source emissions vary considerably in space and time and are influenced by multiple variables. Respiration chambers constitute the most accurate and precise means of measuring emissions of CH_4 and other gases (CO_2 and H_2) arising from enteric fermentation. Historically, measurements of methane emissions have been a component of animal energy metabolism studies during which indirect calorimetry methods have been used to estimate heat production based on exchange of gases (CH_4 and CO_2 production, and O_2 consumption) and urinary N excretion. Open circuit respiration chambers have been the technique of choice for such studies.

Over the last decade, prompted by the renewed interest in finding methods to reduce the quantity of CH_4 emitted by ruminant animals, new respiration chambers have been commissioned in several institutes around the world. These new models are generally much simpler in design and structure to those used previously, yet by taking advantage of the rapid advances in electronics and materials they are both accurate and animal-welfare friendly.

This ***Technical Manual of Respiration Chamber Designs*** was identified as a priority project by the Livestock Research Group of the Global Research Alliance at its first meeting in 2010. The New Zealand government requested proposals in December 2010 for the compilation of a comprehensive technical manual of modern CH_4 respiration chambers, which would cover design, performance and operation of existing chambers but without attempting to evaluate them against any pre-determined performance criteria. The project was co-ordinated by AgResearch, a Crown Research Institute in New Zealand, who invited scientists from around the world to supply details on the design and operation of the different types of existing, newly developed respiration chambers. A standard template was designed and sent to each participant to complete, which requested details on the design and operation of the individual respiration chamber systems. These individual submissions were then collated to produce the manual.

The chambers presented in this book are:

1 **The New Zealand Ruminant Methane Measurement Centre, AgResearch, Palmerston North** was opened in February 2011. It is a purpose-designed facility to house 24 respiration chambers for sheep and four respiration chambers for cattle. The facility allows continuous measurements of methane emissions under highly controlled conditions. The new facility enables the easy flow of animals from the acclimatisation stage to the measurement phase which reduces labour requirements and experimental costs. The state-of-the-art building is fully air conditioned to deliver fresh air to the respiration chambers at a temperature and relative humidity which maximises animal comfort; it has a back-up power supply in case of a power cut, and is continuously monitored in case of emergency.

2 **The Cattle Respiration Facility, Armidale, New South Wales, Australia** is a new, purpose-designed facility to house 10 cattle chambers. The chambers are located inside a 48m x 24m concrete floored shed fitted with 36 individual cattle pens. The shed is insulated though not heated, and is well ventilated by roof vents and windows. Immediately outside the shed are 4 “Ruddweigh” self-feeders with data recorders for measurement of feed intake.

3 **The Institute for Agriculture and Fisheries Research (ILVO) Ruminant Respiration Facility, in Melle, Belgium** is a large facility housing six airtight cattle chambers. The chambers are monitored by one system, which performs dedicated gas sample conditioning, gas analysis, data logging and animal monitoring. This facility has been designed for rapid and efficient feeding, milking, cleaning and animal entrance and exit, so gas emissions are monitored for more than 95% of the time under normal operational conditions.

4 **The Aarhus University Cattle Respiration Facility, Denmark** was built in 1984 with the main purpose of intensive cattle studies. Four respiration chambers were built for milking cows but calves larger than 250 kg can also be measured in the chambers. The chambers are used for measuring methane, but also emissions of carbon dioxide, hydrogen and the consumption of oxygen. The chambers are constructed of steel and polycarbonate.

5 **The Sheep Methane Facility at Aberystwyth University** (UK) houses four open circuit respiration chambers that are suitable for sheep and goats. The chambers are used to measure methane only and recovery of methane through the chamber is quantified over a 24-hour period. Details of a replica system built at **Consejo Superior Investigacion Cientifica (CSIC, Spain)** with only slight modifications are also included in this book.

6 **The Metabolic Centre of the University of Zurich and ETH Zurich** was still under construction during the writing of this manual (2011). In its first stage it will be situated at a temporary location and will consist of two large chambers for cattle, two medium chambers for sheep and goats and two small chambers for smaller animals (piglets etc., not discussed in detail in this manual). It is planned to enlarge the metabolic centre to four chambers of each size.

7. **The Large Animal Respiration Facilities at the Leibniz Institute for Farm Animal Biology (FBN), Dummerstorf, Germany** consists of 4 respiration chambers for cattle or sheep, 4 chambers for pigs and 6 chambers for mice. All chambers as well as the gas analyser and data acquisition system are located in a dedicated facility. The chambers are used to measure methane emissions, but continuous monitoring of carbon dioxide production and oxygen consumption together with feed intake is also possible.

8. **The Small Ruminant Chambers of Universiti Putra Malaysia, Malaysia** were **c**onstructed in 2016 and consist of 5 open circuit respiration chambers for small ruminants (goats and sheep). All chambers are located in a dedicated room. The chambers are used to measure methane emissions. (Added September 2018)

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DairyNZ, New Zealand

February 2014 Updated September 2018

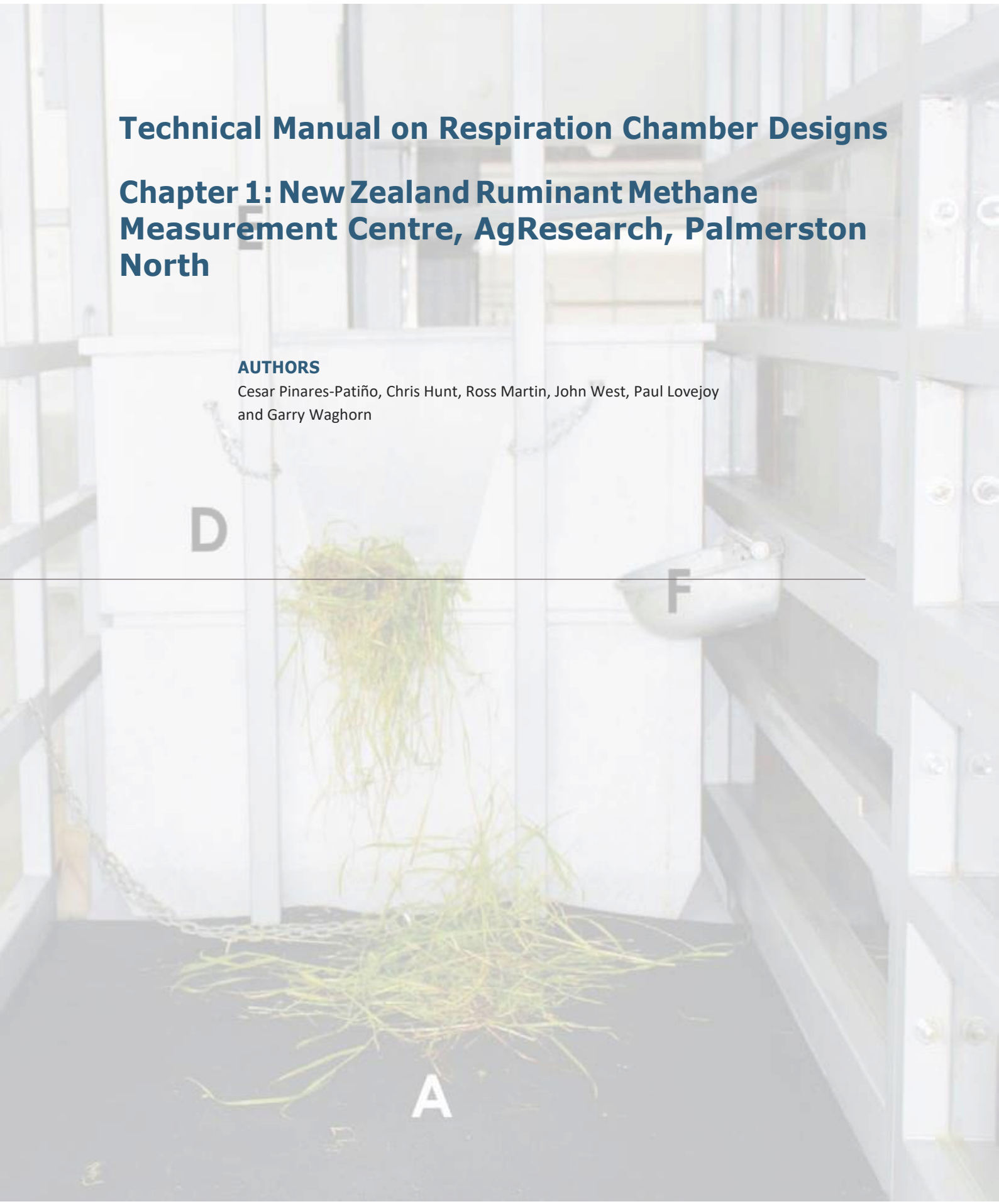
List of Abbreviations

cm	centimeter
d	day
DM	dry matter
g	gram
h	hour
i.d.	inside diameter
k	rate constant
kg	kilogram
kPa	kilopascal
kW	kilowatt
L	litre
m	metre
mm	millimetre
min	minute
mbar	millibar
ml	millilitre
o.d.	outside diameter
pp	polypropylene
petg	terapthtalateglycol (clear polymer)
pfa	perfluoroalkoxy (tubing)
ppm	parts per million (volume/volume)
pvc	polyvinylchloride
stp, STP	standard temperature and pressure
µM	micromolar (10-6)

Technical Manual on Respiration Chamber Designs

Chapter 1: New Zealand Ruminant Methane Measurement Centre, AgResearch, Palmerston North

AUTHORS
Cesar Pinares-Patiño, Chris Hunt, Ross Martin, John West, Paul Lovejoy and Garry Waghorn



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Chapter 1: New Zealand Ruminant Methane Measurement Centre, AgResearch, Palmerston North

1.1 Summary

The AgResearch animal respiration facility comprises 24 airtight chambers for sheep and four for cattle. The sheep and cattle chambers are housed in adjacent but separated rooms within a dedicated facility. The rooms occupied by the sheep and cattle facilities are 32.3 m×5.8 m and 11.8 m×7.7 m, respectively. Each room has an independent ventilation (8 m^3/min) and air conditioning system, which maintains a slightly positive pressure inside the building, while providing a relatively constant temperature (15−25 °C) and relative humidity (37−45%). The sheep chambers (1.8 m^3) are constructed of aluminium (frame and floor) with clear polycarbonate walls, whereas the cattle chambers (15.8 m^3) are made from steel (structure and floor) with clear polycarbonate walls. The chambers operate at a slight negative pressure, with a set air flow of 300 and 1500 L/min for sheep and cattle, respectively. The chambers are deployed side by side along the length of the building.

For purposes of management, the sheep chambers are grouped into three independent systems, each of eight chambers, whereas the cattle chambers are all within a single system. Each system (eight sheep or four cattle) have dedicated gas sample conditioning, gas analysis, data logging and animal welfare monitoring. Within each sheep system, the chambers are grouped in two sets of 4, each sharing an air circulation system and flow adjustment manifold. Within each system, gas samples from all chambers (eight or four) and the ambient are continuously sampled at about 2.5 L/min and a gas switching system delivers a sample stream to the gas analyser over a period of about 30−60 sec, based on CH_4 concentrations stability. The gas sample delivered to the analyser is dried and concentrations of CH_4, H_2, CO_2 and O_2 are measured using a multigas analyser. The gas analyser is calibrated every morning, whereas gas recovery from each chamber is tested routinely and animal welfare is a priority.

The facility has been designed to achieve rapid and efficient feeding, cleaning and exchange of animals, so gaseous exchanges are monitored for more than 95% of the time under normal operation.

1.2 Location of the facility

The physical address of the facility is:

AgResearch Limited
Grasslands Research Centre
Tennent Drive
Palmerston North 4442, New Zealand

Mailing address:

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Private Bag 11008
Palmerston North 4442, New Zealand

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The sheep and cattle respiration chambers are located at the New Zealand Ruminant Methane Measurement Centre (NZRMMC), a purpose-built facility. The facility is part of a research complex, comprising about 10 research institutions, with about 800 personnel. The research complex is located close to the Massey University campus (1 km, 9000 students) and Palmerston North city (four km, 70,000 people). The building housing the chambers is adjacent to grazed paddocks. There are no major industrial sites within 2 km of the facility.

The building housing the chambers was completed in 2011 and is of a concrete and steel construction built to New Zealand standards, which include structural design able to withstand mild earthquakes, and also insulation under floor, walls and ceiling. Electric heating and cooling facilities have been installed in order to regulate relative humidity (via a 2.4 kW condenser). Control of relative humidity in the air supplied into the building is necessary to prevent condensation within the chambers, and is usually maintained at about 40% in the inlet air stream. The building can be maintained within ± 2°C, between about 15 and 25 °C, and the usual working temperature is about 20°C. Air circulation is maintained at all times, with an exchange at 3-4 minute intervals via ceiling vents located about 3 m apart. Air pressure is not directly controlled, but positive pressure (relative to atmospheric pressure) is warranted and prevents leakage of contaminated air into the building from the surrounding areas.

The purpose-built facility houses all 24 sheep chambers in one room (Plate 1), and four cow chambers in another room. A separate room houses instrumentation for sampling, instrument calibration, measurement and data handling that is common for both the sheep and cattle systems. This room is also air-conditioned. Feed preparation and cleaning facilities are nearby (in the same building) as are animal pens, yards and equipment required for handling livestock. The 24 sheep chambers are deployed side by side along the length of the sheep room (32.3 m). Likewise the four cattle chambers are side by side along the length of the cattle room (11.8 m). Sheep access from the adjacent metabolism area is through four large sliding doors, whereas cattle access from acclimatisation area in a covered yard is through an external race.

The 24 sheep respiration chambers are subdivided into three independent system groups, each of eight chambers (Plate 2). This is for purposes of chamber ventilation control, gas sampling, gas analysis, data management and animal welfare monitoring. Each system has a dedicated sample conditioning and gas analyser. The four cattle chambers are independently managed from the sheep systems, the four chambers constituting a single system. Nevertheless, both the sheep and cattle systems operate under the same principles, being the chambers' structure and size, and ventilation rates the only differences.

The following description refers to sheep chambers in a system (Plate 2). The cattle chambers are described at the end of this chapter.

Plate 1: The sheep respiration facility comprises 24 chambers housed in a dedicated building.

Plate 2: The sheep facility comprises three independent systems. Each system integrates eight respiration chambers. The diagram shows a single system with eight chambers and the configuration of pipes for gas flow, manifold for adjusting flow, sampling lines, data acquisition, sample conditioning, etc.

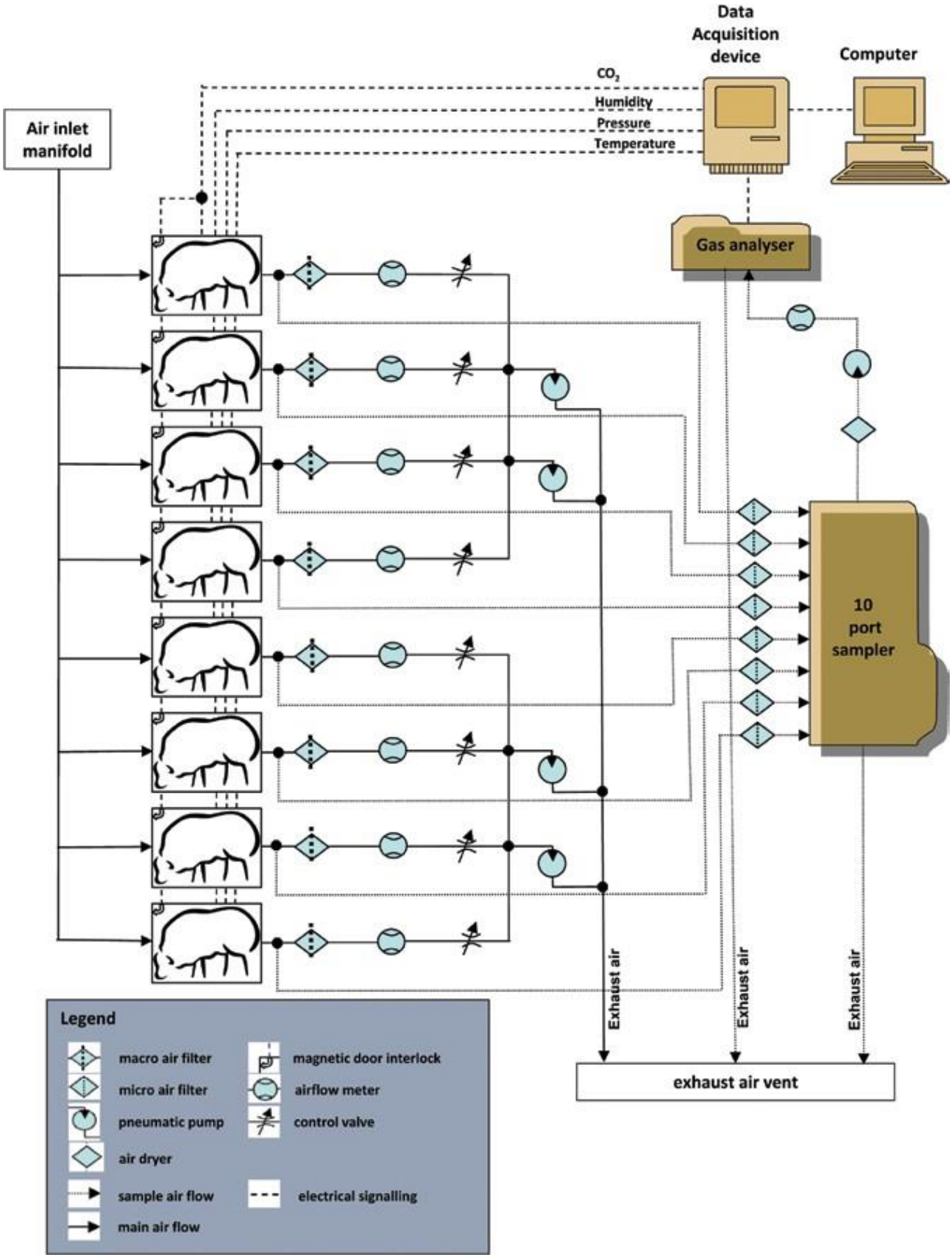


Plate 3: Structure of the sheep chamber, showing the locations of doors, air inlet/ outlet, fans and sensors.

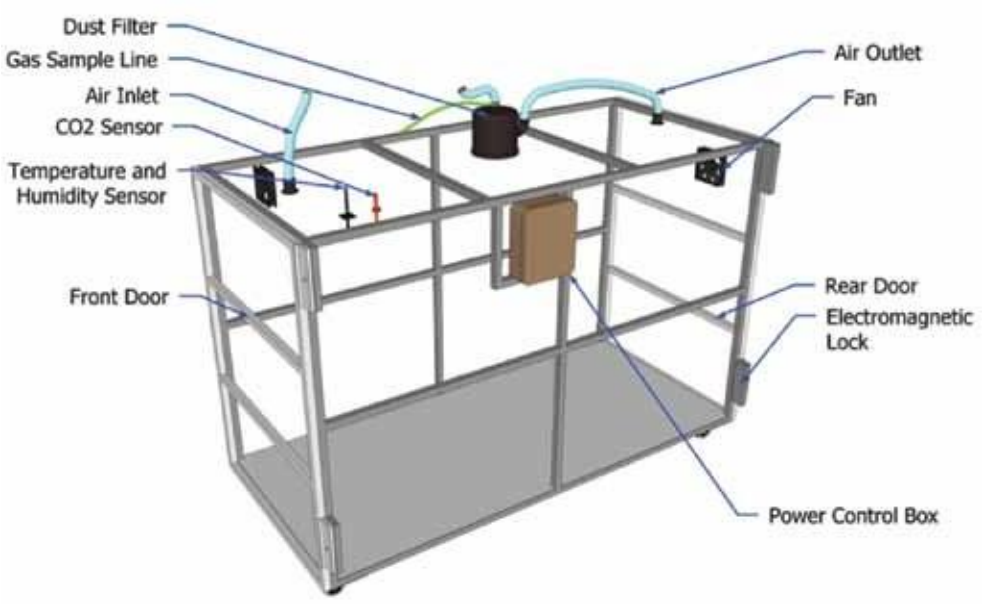


Plate 4: A modified metabolic crate used to hold the sheep in the respiration chamber.

Plate 5: A ramp is used to wheel the sheep crate into the respiration chamber.

1.3 Description of the sheep chambers structure

The respiration chambers were designed to enable accurate measurements of gaseous exchanges and provide a comfortable and safe environment for the animals. The design was derived from existing chambers in Australia and Spain and can accommodate other animal species such as goats or pigs in addition to sheep. Each chamber provides sufficient room for the animal, but enables rapid air exchange (10 air changes per h).

The volume of each chamber is 1.84 m^3, with outside dimensions (mm) of 1800 long, 850 wide, 1200 high (Plate 3). The chamber has front and rear doors covering the whole of each end, and closing onto the frame surface, with a hollow rubber strip adhered to the door where it contacts the frame, to create an air-tight seal. The frame was made of 25×25×3 mm square section aluminum tube, welded with smooth joints and the floor is 3 mm aluminum sheet welded throughout to the frame. The walls and roof are 6 mm clear UV stabilised polycarbonate sheets, each fixed to the aluminum frame using non-acetic cure silicone sealant and rubber seated TEK screws to form an air tight seal. Silicone sealant ensures no leakage around the sides and aluminum floor.

Each access door is held shut by two electromagnetic locks (Maglogs, GL650, Pivotal Solutions Ltd, Auckland, New Zealand), which are opened from the computer at feeding and cleaning times and when animals are exchanged. The rear door enables a crate with the animal in it to be wheeled into the chamber, and also is used for twice a daily exchange of faeces and urine trays. The front door is used primarily for feeding and water replacement. Both the front and rear doors also open in the event of power failure and a rise in CO_2 concentrations above a set concentration threshold (5000 ppm CO_2).

The chambers have four 100 mm diameter castor wheels, with rear ones swivelling so they can be moved easily. The front castors are lockable, whereas the two located at the back are free. In addition, the chambers have four leveling feet so the castors can be raised off the floor for added stability.

1.4 Sheep holding, feeding and cleaning

The sheep are placed in modified metabolism crates (Plate 4) that are wheeled up a ramp into the chamber (Plate 5). This is so to facilitate a rapid exchange of animals to maximise chamber operation time, and also to reduce the physical aspects of the job. In addition, a system has been developed for rapid cleaning of faeces and urine (by exchange of collection trays), again to minimise the time chambers are opened each day. Cleaning and feeding sheep

Plate 6: The mesh faecal collection tray fits on top of the stainless steel urine collection tray and both slide underneath the modified metabolism crate. Exchange of trays during cleaning is facilitated by a use of a trolley and waste bin.

in 24 chambers take 2 technicians 15 minutes. When exchanging animals is required, all the work is completed in 30 min.

The modified metabolic crates used to hold sheep have polycarbonate side panels with large (100 mm) holes to facilitate air circulation, and there are trays mounted below for faeces and urine separation and collection (Plate 6). The back of the crates are fitted with a mesh to avoid air pockets and at the same time to prevent faeces and urine falling outside the collection trays.

The clear polycarbonate sides of the crates and chambers allow sheep to see each other so that they acclimatise to the system almost immediately, indicated by either eating, ruminating or lying. The crates have provisions for holding a feed bin and a drinking water bucket (Plate 4), both easily accessible for the animal. The standard crates are suitable for sheep up to about 55 kg body weight. Large sheep should be shorn (not more than 3 cm wool cover) to ensure they have some movement, alternatively slightly bigger crates are used. Usually animals are kept in chamber for two days. Coefficient of correlation of methane emissions between day 1 and day 2 is high ($r > 0.90$).

The daily procedure enables >23 h of gas measurements to be made, even when sheep are exchanged. The normal procedure is to open (from the computer) the front and rear doors to remove any feed residues and clean the (removable) feed bin. New feed is added, and water supply replenished (usually 2−3 L, as required). At the rear, the faeces and urine collection trays are removed and replaced with clean trays. Doors are closed and the chamber is allowed to equilibrate. Feeding is usually twice daily, with refusals removed at each feeding. Feeding is usually at 0830 and 1630 h.

At the end of the second day of measurements, the crates holding the sheep are wheeled out of the chambers and new group of sheep in crates are wheeled in. The sheep are allowed 3 to 5 days to acclimate to the crates in the metabolism area (Plate 7), so they are easily wheeled into the chambers.

1.5 Chamber airflow piping and measurement (sheep)

Air flow through each sheep chamber is typically 300 L/min. Air is ducted through the top of each chamber, entering (inlet) the front and exiting (outlet) from the rear, with two internal fans (120 mm; 12 volt) mixing air within the chamber (Plate 3). All the chambers within a system share a common inlet source (located at the ceiling of the building) (Plate 8), which is monitored for gas concentrations at hourly intervals. A flexible 38 mm o.d. EOLO hose (Hose & Coupling Distributors, Wellington, NZ) with spiral reinforcing and a smooth interior surface is used for piping air circulation throughout the chamber systems. Each chamber is fitted with a sensor, close to the outlet (Vaisala Humicap® HMT100 (Vaisala Oyj, Helsinki, Finland), to monitor changes in relative humidity and temperature (Plate 9). In addition, a Vaisala PTB 110 barometric pressure sensor is fitted at the chamber air flow measurement device. Data on these environmental parameters enable air flows to be adjusted to dry standard temperature and pressure (STP) conditions. The sensors operate within their prescribed range, and there is no need for adjustment.

The air pumping system comprises two pumps (SCL-K03-MS, FPZ, Concorezzo (MB), Italy) enabling a continuous draw of air through the chambers. The air pump is located at the end of the air circulation system and exhaust is removed out of the building. Thus, throughout the system (chambers and ducting) a negative pressure is maintained and leakage out of the system is avoided.

Plate 7: The sheep are maintained in the metabolism area in sets of 24 animals, ready to be wheeled into the respiration chambers every second day.

Plate 8: A common air inlet (located near to the building ceiling) for the eight chambers within a system. Background air measurements are based on samples taken at this point. Temperature and relative humidity are also monitored at this point.

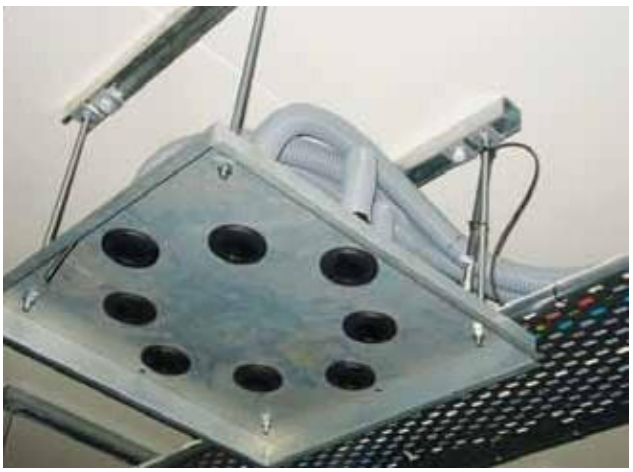


Plate 9: Top of the chamber showing the ducting for air to leaving the chamber via outlet **(A)**, before it is filtered **(B)** and sampled **(D)** to determine gas concentrations. The chamber is fitted with a temperature and humidity sensor **(F)** as well as a CO_2 concentration monitoring sensor **(C)**.

The air handling capacity of each pump is about 1230 L/min, but regulation of air flow is based on two sets of four chambers (Plate 10) and there is a common air flow manifold and pumping system for each set. The pumping system for each set of four chambers involves two pumps connected in parallel (Plates 2, 10).

Air flowing out of each chamber is first piped to a F198 dust filter (PVR srl, Valmadrera, Lecco, Italy) and then to a wet flow measurement unit (1 for each chamber). The outlets of the air flow measurement units are piped to a common manifold, which is used for adjustment of air flows for each chamber (Plate 10). The manifold has eight valves: four serving as inlets to receive air from each of the four chambers, whereas the other four are interconnected outlets, of which two are relief valves open to the building environment and the other two valves connect the airflow to the inlets of each the two pumps. The outlets from the pumps are exhausted outside the building (a common exhaust for two pumps) (Plates 2 and 10). Two pumps for each set of four chambers enable adjustable air flow in the range 100−400 L/min. The air flows are maintained at a constant flow and there is little variation between trials, associated with animal size (displacement).

Air flow (wet) from each chamber is measured using the principles of differential pressure within a Venturi tube, which was made by welding stainless steel tubes of 40 and 20 mm o.d. (Plate 10). The 40 mm tube has a length of 80 cm, whereas the 20 mm tube is of 40 cm. Within each tube section air pressures are measured at two different points using Vaisala PTB 100 sensors. Six mm polyethylene tubing connects the pressure sensors to the Venturi tube sections. The connectors are fitted into the stainless steel tube, but they do not extend into the Venturi, so turbulence and anomalous readings are avoided. The air flow is set to a fixed rate and, calibrated using 6−12 h serial measurements with a diaphragm gas meter (AL425, Elster American Meter Company, Essen, Germany).

1.6 Sampling, sample conditioning and analysis (sheep or cattle)

Outlet gas from each chamber is sampled continuously (2.5 L/min) immediately after the dust filter into a multiport gas switching unit (S.W. & W.S. Burrage, Ashford, Kent, UK) through 6 mm nylon tubing, with an in-line 7 µm filter.

The multiport unit switches the samples (eight chamber samples + 1 background for sheep or four chamber samples + background for cattle) at variable times (within 30−60 seconds), depending on the stability of the gas concentrations determined by the gas analyser. The gas analyser (Plate 11) determines gas concentrations at 5 sec intervals, and purpose-built software enables the gas switching system to change samples once the concentrations of CH_4 (the target gas for our purposes) from the last three readings stabilises with a variation less than 1 ppm. Sample concentrations usually stabilise within 20−45 sec, but a minimum of 30 sec and a maximum of 60 sec are allowed, whereas the background air concentration is measured hourly over a 60 sec period. The cycling time to measure gas concentrations from the 9 (sheep) or 5 (cattle) sample streams is completed within a 5-min or 3-min period, respectively.

Sample gas is delivered to the analyser by a means of a diaphragm pump (N89KNE, KNF Neuberger Inc, Freiburg, Germany) at 2.0 L/min. Before entering the analyser the sample is dried using a heated drier MDH-110-96F-4 and an unheated drier MD-110-24P-4 (both from Perma Pure, New Jersey, USA) connected in series (Plate 12). These driers have Naflon membrane elements and utilise dry air in counter flow configuration, at double the sample

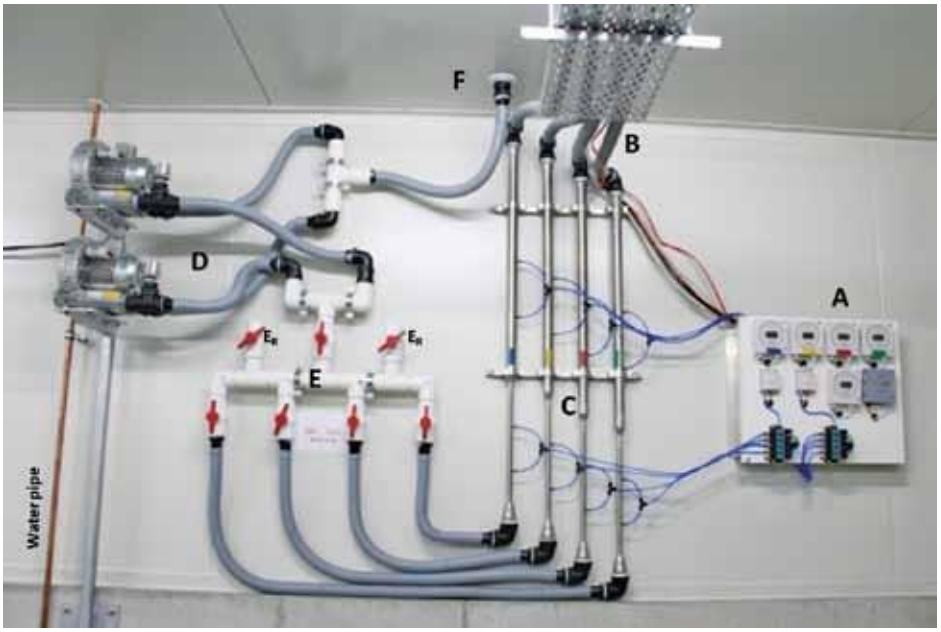


Plate 10: Chamber environment control panel **(A)**, assembly of chamber outlet pipes **(B)** with the stainless steel Venturi flow meters **(C, showing pressure outlets)**, manifold for adjustment of flows **(E, showing relief valves, ER)**, parallel assembly of pumps **(D)** and common exhaust **(F, to outside the building).** All these features are shared by four chambers within a system. Two of these sets constitute one system. The water pipe is not part of the system.

Plate 11: A dedicated gas switching unit **(middle)** and a gas analyser **(bottom)** are allocated to a system of eight chambers (sheep). A diaphragm pump (close to the gas switching unit) supplies the sample to the gas analyser, but the sample is split into two streams using a sample conditioning **(top)**, one for CH_4, CO_2 and H_2 and another for O_2.

Plate 12: Sample drying system. Sample is delivered (~2 L/min) to the analyser using a micro pump **(F)**, but is first dried using a heated drier **(D)** and then a non-heated drier **(E)**. Removal of moisture from the sample requires dry air to be circulated in counter flow (4 L/min) to sample. Dry air is generated by a self-regenerator drier **(C)**, but to avoid condensation, a refrigeration unit (B) is placed between the pump **(A)** and the dry air generator **(C)**.

flow rate. The dry air is provided by a heatless self-regenerative air drier (Nexus Analytical Pty Ltd, Australia). Room air supply to the dry air generator is via a Thomas 617CD22-194C pump (Thomas, Sheboygan, WI, USA). However, to avoid condensation inside the tubing supplying air from the pump, the air is cooled to 4°C using a thermoelectric refrigeration unit (XC3000A, Tropicool, Christchurch, New Zealand).

External to the gas analyser, the dried sample is divided into two sample streams: 1.5 L/min for CO_2, CH_4 and H_2 measurement and 0.2 L/min for the O_2 measurement, with the remaining air released. Before entering the analyser, the sample streams are filtered (0.5 µm).

Gas concentrations in dried air are measured using a Servomex 4900 gas analyser (Servomex Group Ltd., East Sussex, UK). Methane and CO_2 are measured using the infrared technology, whereas O_2 is measured using a paramagnetic cell. In addition, the gas analyser is fitted with an electrochemical H_2 detector (7HYT Citicel, City Technology Ltd., Portsmouth, Hampshire, UK). The detection ranges for CH_4, CO_2, O_2 and H_2 are 0–200 ppm, 0–2500 ppm, 0–25%, and 0 to 500 ppm respectively, with corresponding accuracies of 0.2 ppm, 25 ppm, 0.05% and 5 ppm.

The CH_4 analyser is calibrated every morning using zero gas (N_2, 99.99%) and an alpha standard containing a mix of gases: CH_4, 200±3 ppm; H_2, 100±2 ppm; CO_2, 2000±20 ppm; O_2, 21.0±0.1%, in N_2 as carrier. The calibration gases are supplied by BOC Limited (Auckland, New Zealand).

1.7 Gas recovery test (sheep and cattle)

Gas recovery tests of the respiration chamber system are independently monitored by the National Institute of Water and Atmospheric Research (NIWA, Wellington, NZ) by mass flow metering of ultra-pure CH_4 and H_2 (separately). The gas metering is set to achieve a concentration of about 100 ppm of CH_4 and 20 ppm of H_2. The metering of these gases is carried out separately, that is, no gas mix is used. It is carried out over a 1 hr period, time enough to stabilise the gas concentrations and at the same time to have an accurate ventilation rate measurement. Gas recovery tests are carried out at variable intervals depending on whether the system has suffered sizeable alterations (for example, replacement of hoses and pumps etc.), but it usually is done at about 3 month intervals. These tests revealed that the system is quite stable with mean recovery rates of 98.2 ± 0.60 and 100.5 ± 4.01 for CH_4 and H_2, respectively for the sheep chambers, while the cattle chambers have recoveries of CH_4 and H_2 of the order of 101 and 102%, respectively.

1.8 Emissions calculation

Calculation of enteric emissions of CH_4 (and other gases) is based on accurate measurements of the chamber wet ventilation rate (Wet VR), the net concentration of gas in dry sample (that is, above the background concentration), and the percentage of gas recovery in the entire system.

The wet ventilation rate (Wet VR) has to be adjusted to dry standard temperature and pressure ventilation rate (Dry STP VR). For a given point of measurement, the instantaneous emission of CH_4 is calculated using the formula:

$$CH_4 \text{ emission (L/min)} = (\text{Dry STP VR} \times ([CH_4 \text{ ppm}]/1000000)) / \text{gas recovery rate}$$

For example, if the Dry STP VR was 290 L/min (for sheep) and the net CH_4 concentration in the sample was 50 ppm, with a gas recovery rate of 98%, the instantaneous CH_4 emission will be 0.0153 L/min. Note that the 1000000 factor converts ppm to litres.

For any chamber within a system, data for wet ventilation rate (Wet VR), gas concentrations and environmental conditions are available at 5 sec intervals over a 30−60 sec period and it is assumed that for all these variables their mean values for the last three values are representative of the 4−5 min cycle period (the case of the sheep system).

The data files are saved on daily basis (00:00 h to 23:59 h), but animal measurements usually start at about 08:30 h when animals are brought in, fed and chambers closed. Chambers are also opened to exchange excreta trays and provide feed and water at 16:30 h on day 1 and 0800 and 16.30 h on day 2. Data are collated for 24 h periods starting when chambers are first closed 08:30 h and ending after 48 hours (08:30 h). Missing data when the chambers were open are estimated by interpolation based on the 10 values (~50 minutes) immediately before the chambers were opened. With animals fed twice daily, emissions before chambers are opened for feeding are fairly stable and lower than the daily means.

The calculation of dry STP ventilation rate (Dry STP VR) requires data for relative humidity (%), temperature (°C) and pressure (hPa) specific for each chamber.

Dry STP ventilation rate (L/min) =

[(Air pressure × Dry gas VR) / (Chamber T + 273.15)] × 273.15/1013.25, where pressure is in hPa, Dry gas VR is in L/min, Chamber T is the chamber temperature in °C.

Dry gas VR (L/min) *= Wet VR × [(100 – VMR)/100],* where Wet VR is the ventilation rate recorded from the flow meters (L/min), VMR is the Volume Mixing Ratio of moisture (%).

Volume mixing ratio (VMR) (%) *= 100 × PWP/air pressure,* where PWP is the partial water pressure (hPa), and the air pressure in hPa.

Partial water pressure (hPa) *= (6.1117675 + 0.4439 T + 0.014305 T^2 + 0.000265 T^3 + 0.00000302 T^4 + 0.0000000204 T^5 + 0.00000000006388 T^6) × RH/100*

The partial water pressure (hPa) is obtained using the Wexler equation, where T is chamber temperature (°C) and RH is the chamber relative humidity (%).

Once instantaneous emissions of CH_4 (L/min) are calculated for each interval of time for each chamber, and the missing values (when the chambers remained open) have been estimated, the daily emission (for a 24 h period) from a particular animal housed in a given chamber is calculated by time integration (area under the curve).

Daily emissions can be converted from L/day to g/day using the conversion: 1 g CH_4 = 1.3962 H_4.

1.9 Animal welfare and operators' safety

Animal welfare

All animal experimentation must conform to good ethical considerations. The design of these chambers is based on both animal and operator safety.

The system ensures that all environmental conditions are within the thermoneutral zone for the animals, and there is minimal exposure to stress or risk. The system is monitored for temperature, air flow, relative humidity and gas concentrations, and alarms will activate when abnormal conditions are detected (Plate 13). Should the conditions fall outside the following values, the doors open automatically and operators are called: temperature (15–24 °C), air flow (250–310 L/min for sheep; 1400–1600 L/min cattle), relative humidity (40–80%) and CO_2 (800–5000 ppm), among other indicators.

Because the chambers are air-tight and operate under a slight negative pressure, the most critical animal safety risk is the lack of ventilation throughout the system. Consequently, power failure or malfunctioning of air pumps could cause suffocation if the air flow ceased. Four safety measures have been incorporated to overcome this risk:

1. The doors of the chambers are held in place by powered solenoids and in the event of power failure they automatically open allowing fresh air to enter the chamber.
2. The vacuum pumps (two) responsible for continuous air flow are arranged in parallel to maintain air circulation even one pump fails.
3. Since four (sheep) or two (cattle) chambers are piped into a single air pumping system, any failure will result in increased CO_2 concentrations in all chambers sharing the pumping system. One sheep chamber in each set of four is fitted with a CO_2 sensor (GMP222, Vaisala Oyj, Helsinki, Finland), which is linked to the controls of power supply to all of the door solenoids and if the CO_2 concentration reaches 5000 ppm all doors of the eight chambers within a system will open. In the case of cattle chambers, each of them is fitted with a CO_2 sensor.

Plate 13: Computer (and power up system) with alarms set for abnormal environment conditions.

4. The respiratory chamber system is connected to the AgResearch computer network which enables remote monitoring. Alarms are in place for abnormal CO_2 and CH_4 concentrations, temperature, relative humidity and airflow, in which case, emails and cellular telephone messaging alert personnel. An elevated CH_4 level (> 300 ppm) may also indicate failure of the pump system and serves as a contingency in the event of a CO_2 sensor failure. Operator response is within 5 minutes when on site or 15 minutes after hours.

Operator safety

Operator safety is also important and the use of aluminum has ensured the chambers and crates are light. Although the use of aluminum for crate construction is more expensive than steel, the absence of rusting and easy cleaning compensates through lower labour costs. The ramp for wheeling sheep in and out of chambers has worked very well, as has the removable faeces and urine trays. The sieve that separated faeces from urine (Plate 6) speeds cleaning, and the trolley to hold the dirty trays removed from the chambers also reduces effort required for routine cleaning and avoids unnecessary opening of the building doors for removal of trays during cleaning. Operators' safety in the cattle chamber is warranted by allowing an area for the operator separated from the cattle area.

1.10 Weaknesses of the system (sheep and cattle)

The main weakness of the system is the inability for uniform environmental conditions to be set in all the chambers. The differences in animal size, defecation and urination events, and animal activity imply that environmental conditions inside the chambers are not only different between chambers, but conditions change within a day. The control of the temperature and relative humidity is for the building incoming air. Fitting an air conditioning system for each single chamber would be onerous and probably inefficient due to the lack of insulation in the chambers.

Occasionally delays in delivery of messages to cell phones have been experienced but this problem is outside of our control. Animal welfare was never compromised because in case of serious risk (for example, power failure) the chamber doors opened automatically.

The absence of automated feeding and feed weighing devices within the chambers limits the type of research that can be undertaken. Developments in electronics now allow the implementing of these facilities without compromising air tightness, efficiency of air mixing and response time of the system, but they still are onerous.

1.11 Description of components and equipment suppliers

Chambers structure (sheep)

- Chamber frame is 25x25x3mm aluminum tube
- Chamber floor is 3 mm aluminum sheet, with 8 mm corner sections to support
- Castors (100 mm; fixed and swivel); Wheels & Castors, Glenfield, Auckland, NZ
- Sides, doors and roof are 6mm UV Stable Polycarbonate sheet; Graley Plastics Supplies, Petone, Lower Hutt, NZ
- Fasteners; TEK screws 10-6x25 with neoprene washers; Ullrich Aluminum, Manukau City, NZ
- Silicone sealant (Bostik Industrial Grade Neutral Cure clear); Bunnings, Palmerston North, NZ
- Fans 12v DC 120mmx120mmx25mm; Globelink Limited, Palmerston North, NZ

- Switch Mode power Supplies; PSU1B 13.8 v dc 1.2amp; B&M, Palmerston North, NZ
- Electromagnetic locks (Mag lock 650-LC 1.2 amp); Pivotal Solutions Ltd, Auckland, NZ

Air circulation and pumps (sheep)

- Piping, 38 mm Eolo tubing; Hose & Coupling Distributors, Naenae, Wellington, NZ Fittings and connectors for 38 mm hoses, Hansen Products (NZ) Limited, Whangarei, NZ
- F198 dust filter (PVR srl, Valmadrera, Lecco, Italy); distributed by HIVAC Ltd, Silverdale, Auckland, NZ
- Air pumps (SCL-K03-MS, FPZ, Concorezzo, MB, Italy), distributed by HIVAC Ltd, Silverdale, Auckland, NZ
- Test gas meter to calibrate Venture flow meters, AL425, Elster American Meter Company, Essen, Germany

Sensors, analyser, sampling and sample conditioning (sheep or cattle)

- Environment sensors: Temperature and relative humidity (Vaisala Humicap® HMT100), pressure (PTB 110), and CO_2 concentration (GMP222), all from Vaisala Oyj (Helsinki, Finland), distributed by Vaisala Pty Ltd, Hawthorn, Melbourne, Australia
- Nylon tubing 6 mm for sampling; Norgren, Palmerston North, New Zealand
- Gas switching unit: 10 channel unit; S.W. & W.S. Burrage, Ashford, Kent, UK
- Unit for splitting sample into two streams: Applied Instruments Group (2007) Ltd, South Auckland, Auckland, NZ
- Gas analyser, Multigas 4900 gas analyser (Servomex Group Ltd., East Sussex, UK), distributed by Applied Instruments Group (2007) Ltd, South Auckland, NZ
- Micro-diaphragm pump for sample delivery to analyser: N89KNE (KNF Neuberger Inc, Freiburg, Germany), distributed by HIVAC Ltd, Silverdale, Auckland, NZ
- Heatless Nexus HLD dry air generator; Nexus Analytical Pty Ltd, Engadine, NSW, Australia
- Pump for fresh air supply to the cooler: Thomas 617CD22-194C, distributed by Nexus Analytical Pty Ltd, Engadine, NSW, Australia
- Heated and non-heated sample gas drier using Naflon membrane from Perma Pure (NJ, USA), distributed by Nexus Analytical Pty Ltd, Engadine, NSW, Australia
- Air cooler (XC3000A, Tropicool) to supply cool air to the dry gas generator; Tropicool, Christchurch, NZ

Data acquisition and logging (sheep or cattle)

- Standard PC, tower case (full height PCI slots to accommodate relay cards)
- Relay output card: DASP 52032 PCI-16 channel relay output card: Jenlogix Ltd, Auckland, NZ
- Data acquisition: Picolog 1216, 16 channel, 12 bit resolution Data acquisition module; distributed by Metermaster Ltd, Auckland, NZ
- Control system software, custom software written in Visual Basic 6.0, Microsoft, USA
- Backup Power Supply: 650VA UPS (Cat No. MP-5204); JayCar, Palmerston North, NZ

1.12 Costing of the sheep facility for a complete system of eight respiration chambers and the ancillary equipment required

ITEMS	NZ$	US$
LABOUR FOR		
Design of the system	2,000	1,600
Building of chambers	35,000	28,000
Piping air circulation and sample lines	2,000	1,600
Wiring, data acquisition, software development	15,000	12,000
Monitoring and commissioning	10,000	8,000
Tests	3,000	2,400
MATERIALS		
Building materials	30,000	24000
Pipes, cables, etc	4,000	3200
EQUIPMENT		
Minor assets (sensors, pumps, etc)	20,000	16,000
Gas analyser	40,000	32,000
Sample drying system	5,000	4,000
Gas switching system	6,000	4,800
Calibration gases	2,500	2,000
Computer and data acquisition system	5,000	4,000
TOTAL COST[1]		143,600

1.13 AgResearch cattle respiration chambers

The cattle respiration facility involves four chambers, housed in a room (11.8 m×7.7 m) adjacent to the sheep respiration facility. The air conditioning system for this room is similar to that for the sheep system, but independently controlled. The chambers were constructed from mild steel rectangular hollow tube sections, and covered in clear 6 mm thick UV stable polycarbonate sheet. The net volume of each chamber is 15.8 m^3 and 4.0 m long, 2.0 m wide and 2.2 m high. Due to their size and building access, it was necessary to construct the chamber in two halves. Then the two halves were bolted together inside the facility using a full bead of silicone sealant ensuring a stable airtight seal.

The main frame of the chamber uses 70 ´ 50 ´ 5 mm steel tube for its superstructure and 50 ´ 50 ´ 5 mm steel tube for the sub-frames, with animal/operator safety dividing partitions fabricated from 30 ´ 30 ´ 3 mm steel tube. The floor of the chamber is 6 mm thick mild steel plate welded to the main frame and silicone-sealed. The clear polycarbonate covering walls, roof and doors of the chamber are fixed to the metal frame using neutral-cure silicone sealant and rubber-seated TEK screws at 100 mm centres to ensure air-tightness. The chamber has one front and two rear access doors fitted with rubber seals. The front door (115 cm wide ´ 215 cm high) is located at the left side and closed by two electro-magnetic locks. In the event of either a power failure or a CO_2 build-up to predetermined levels, the door locks de-energise, and the doors spring open allowing a fresh air supply to reach the animals. This door is also used for animal feeding.

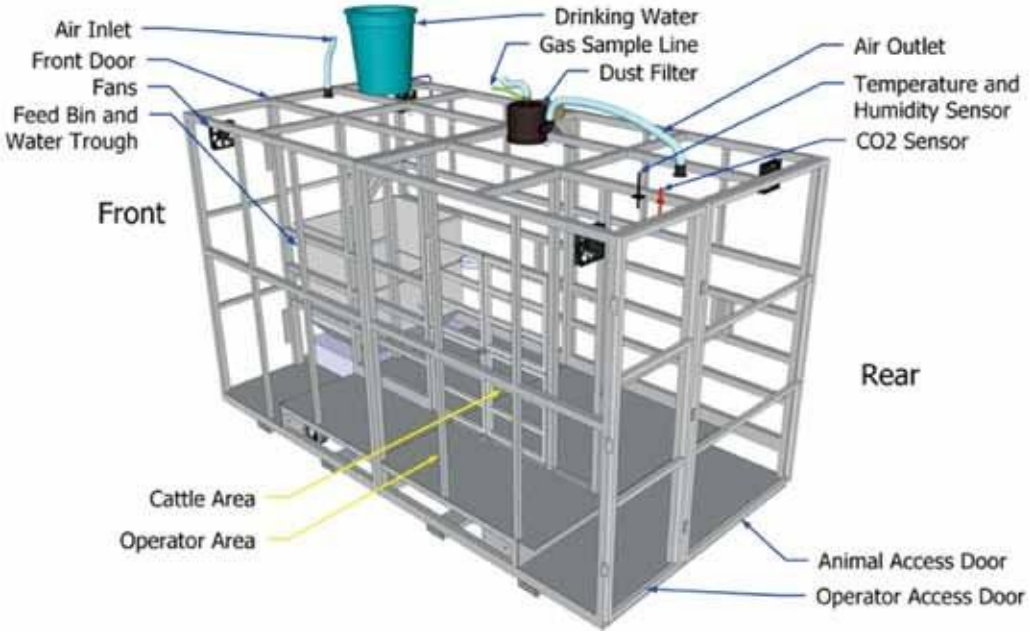
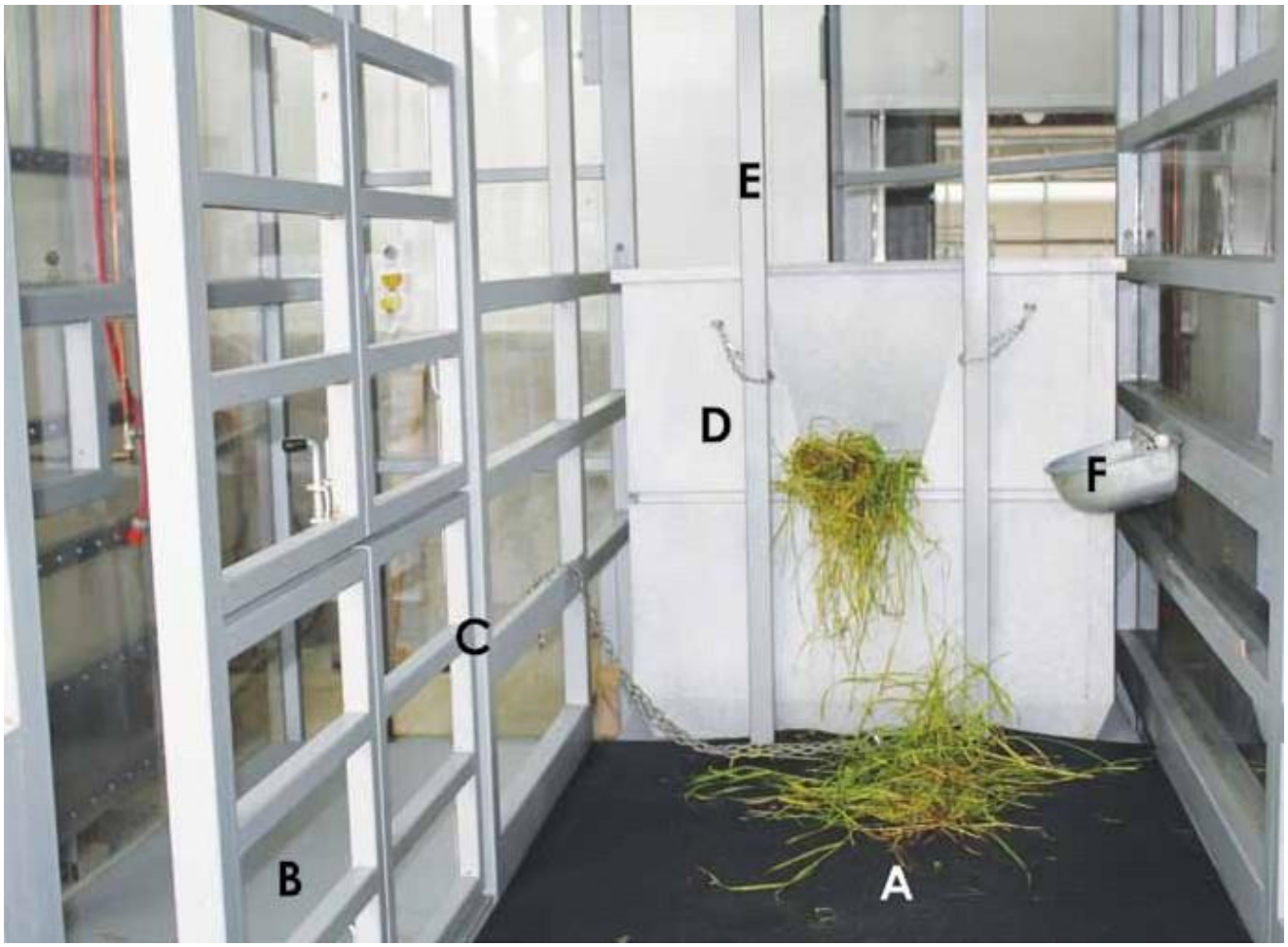


Plate 14: Design of the cattle chamber showing its structural frame and ancillary equipment. The frame is covered with clear walls and roof.

Plate 15: Internal view of the chamber showing the cattle area covered in a rubber mat **(A),** the operator area **(B)**, the protection frame **(C)**, feed bin **(D)**, the bars to avoid cattle passing throughout the front door (E) and the water trough (**F**).



The two rear doors are hinged on either side of the chamber (Plate 14). The main rear door (115 cm wide ´ 215 cm high) located at the right side is used for animal access, whereas the smaller door (90 cm wide ´ 215 cm high) located at the left side is used for operator access. These doors are secured with three large clamping wing nuts ensuring that an air-tight seal is achieved by the door against the main chamber frame.

The main floor is the whole width of the chamber and is 20cm higher than recesses in the front (80 cm deep) and rear (65 cm deep). The chamber comprises two sections (A and B; Plate 15). Section A (117 cm wide and 250 cm long) is the cattle holding area and is covered in rubber mats. Section B (90 cm wide and 250 cm long) is the operator safety area and is separated from section A by an internal metal frame with 'easy-access' openings for rumen sampling. The low floor at the front is used for placement of the feeding bin, whereas the low floor at the rear is where the excreta collection bins are placed. Drinking water is provided by a water trough located at the right internal wall of the chamber. For reasons of safety and requirements for measuring water intake, the water supply is piped from a 50 L covered bucket located on top of the chamber.

A removable head bail is built into the chamber to prevent cattle from turning around. It allows easy movement of the animal during feeding, drinking, resting and lying down. However, for well acclimatised animals, the head bail is removed and instead a neck collar is used to tie the animal down to the floor of the chamber. The front of the cattle housing area has two perpendicular metal bars to prevent the animals passing through the front, but these bars do not impede access to the feeding bin. The rear of the cattle area is designed so that a metal bar can be placed behind the animal, 70 cm above the floor, to prevent the animal stepping into the excreta collection bin. The chamber has provisions for adjusting the water trough and head bail to suit different size animals (for example, calves and adults). The whole chamber is leveled using four adjusting screws that can be retracted so the chamber is able to be maneuvered on large braked castors. The animals access and exit the chamber throughout the rear door.

The chamber is fitted with a fresh air inlet in the front section of the ceiling and an air outlet in the rear ceiling. All the piping for air circulation is through flexible polyurethane hoses (51 mm o.d.). The air inlet is piped from an air intake in the ceiling of the building. Incoming fresh air and the respiratory gases are mixed by four fans located at the top corners of the chamber. The outlet hose is connected to a 51 mm air filter (F300, P.V.R. srl, Valmadrera, Lecco, Italy) and then to a wet gas flow measurement system. The gas flow measurement system is based on the differential pressure principles, similar to that for the sheep chambers, except that the large diameter (50 mm o.d.) and small diameter (30 mm o.d.) stainless steel tubes are 100 and 50 cm in length, respectively. The gas flow system is calibrated using a quantometer (Qa100-80-016, Elster/Amco, Malnz-Kastel, Germany). The outlet from the gas flow measurement system is connected with polyurethane hoses to a manifold and then to a vacuum pump system using side-channel pumps (SV 7.190/1-01, Gebr. Becker Gmbh & Co., Wuppertal, Germany). Two pumps connected in parallel, draws air through two chambers at continuous but adjustable flow rates (1000–2000 L/min). Air (with respiratory gases) exiting the air pump is exhausted outside the building. Relative humidity and temperature sensors are installed in the chamber, as with the sheep chambers for standardisation of the gas flow. Air pressure is measured at the gas flow measurement system.

Gas sampling, sample conditioning and analyses are conducted as for the sheep system. An exception is that gas analysis cycles for the cattle system are shorter than for sheep due to the smaller number of cattle chambers (four versus eight for sheep). Similarly, all the safety measures found in the sheep system are installed in the cattle respiration facility.

The performance of the system is evaluated using the gas recovery protocol as for the sheep chambers.

The cost of a facility comprising a four-chamber system for cattle is about NZ$250,000 (US$200,000), with labour (excluding design, software development and commissioning), materials and equipment comprising 15, 55 and 30% of it, respectively.

Technical Manualon Respiration Chamber Designs

Chapter 2: Cattle Respiration Facility, Armidale, New South Wales, Australia

AUTHORS
Roger Hegarty, Simon Bird and Reg Woodgate

Contents

Chapter 2: Cattle Respiration Facility, Armidale, New South Wales, Australia

2.1 Summary

Ten open circuit respiration chambers have been constructed, initially for use in quantifying cattle methane production but will in future be used for energetics research. The chambers are of 20m^3 internal volume with an air flow rate of 1.6 m^3/min and consist of enclosed pens (1.8m x 3m) within a polycarbonate shell (3.6m x 2.4m x 2.4m). The chambers have no inbuilt floor but seal in a water trench recessed into the floor and are raised by pneumatics to allow hosing out of waste. Chamber air flow is measured by individual mass-flow meters and a continuous subsample of gas is drawn from immediately before each flow meter. Moisture is removed by a cold trap and a multiplexer used to direct dried sample air from each chamber and the ambient air into the analyser in turn. Methane, oxygen and carbon dioxide concentrations are measured over 10s after a 40s purge time by a Servomex analyser. Air flow and gas concentration data in the sampled air are loaded directly into a daily workbook with separate Excel spreadsheets for each chamber to allow gas production every 9 min to be determined. Methane recovery through chambers is measured by injection of a known dose of methane and integration of the peak area using first order kinetics.

2.2 Location of the facility

The physical address of the facility is:

University of New England
Trevenna Road, Armidale
NSW 2351, Australia

Mailing address:

Dept. Animal Science
University of New England
Armidale
NSW 2351, Australia

Contact persons:

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Email: hutton.oddy@industry.nsw.gov.au

The chambers are located in a new animal house facility in Armidale in northern New South Wales at 1000m altitude (Plate 1). This is on the campus of the University of New England and adjacent to existing animal house facilities for sheep studies (respiration chambers, metabolism cages, floor pens). The chambers are located inside a 48 m x 24 m concrete floored shed fitted with 36 individual cattle pens. The shed is insulated though not heated, and is well ventilated by roof vents and windows. Immediately outside the shed are four "Ruddweigh" self-feeders with data recorders for measurement of net feed intake. These feeders are formed on concrete bases but the surrounding pens are finished with compressed road-base (gravel) and a slope to ensure drainage. Cattle will be able to walk from the new facility to adjoining research farms by laneways in the near future. The "Tullimba" research feedlot (2000 head capacity) is 45 min drive away and trucks bringing cattle to the calorimeter facility can unload at the shed entrance, then cattle can be treated and weighed as required. A crush is located in the shed to facilitate weighing and taking of rumen, faecal and blood samples.

2.3 Description of the chambers structure

The ten chambers are arranged in two rows of five, with chamber doors opening onto a common aisle that runs between the 2 rows (Plate 2). The chambers are constructed of 75 mm hot dipped galvanised tubular steel modular frames to give length, width and heights of approximately 3.6 m, 2.4 and 2.4 m respectively, and an approximate volume of 20 m^3. The panels consist of roof, 2 sides, rear wall and front door with polycarbonate (3 mm or 6 mm) sheeting fitted to the inside of each panel. Joins between panels were sealed with silicone and panels bolted together on-site. The front door of the chamber is full width and opens into the aisle to allow cattle entry. Water is plumbed in for a drinker and a 120 L feed bin is provided on the front gate. The feed bin can only be removed by opening the chamber door.

Each chamber has made be raised or lowered into a matching 100 mm wide rebate into concrete floor which can be filled with water to provide a water seal (Plate 3.) and prevent gas loss when the chamber is lowered. Raising or lowering of chambers is achieved by 4 pneumatic rams, fitted one to each corner of the chamber and these are connected to a common compressed air line. The floor itself is painted with a 2 pack epoxy to prevent CO_2 absorption.

Bolted to the floor inside each chamber and 300mm in from the chamber frame, is a pen (3 m x 1.8 m) constructed of cattle panels which are bolted to the floor and for which a full width gate is positioned directly behind the front door of the chamber itself (Plate 4). The gate has a spring loaded catch for operator safety when cattle are being introduced. The permanent pen allows cattle to be enclosed without risk of damage to the polycarbonate chamber.

2.4 Animal holding, feeding and cleaning

Animals to be measured for methane production are adapted to diet for at least 14 d in the self-feeders outside of the shed. The quantity of feed loaded into the feed hopper each day is selected to provide the average intake required relative to maintenance for the total number of animals in the pen (max = 10). The feed intake data from these individual animals can be monitored electronically enabling any animals with variable or extreme intakes to be identified. In a typical measurement cycle, cattle are adapted to diet for 14 d on self-feeders in groups, then cattle are individually housed and fed a fixed ration for 2 d prior to entering the chambers for a further 2 d. A 2 day cleaning cycle is used throughout the shed. After 48 h in chambers, cattle are removed and sampled as required, then returned to the outside pen.

Plate 1: New Centre for Large-Animal Science Studies facility at UNE, Armidale.

Plate 2: Two rows of five chambers showing aisle between rows, overhead duct and droppers bringing fresh air into each chamber.

Plate 3: Pneumatic rams fitted one to each corner of the chamber, which lift the entire chamber from the water filled recess used to seal it. Lifting the chamber is initiated by a button on the chamber or automatically if the power fails or oxygen concentration decreases below 18%.

All chambers can be raised 20 cm above floor level (by compressed air pneumatics), to allow manure and urine from the chambers to be hosed across the shed into side trenches covered by grates. The next group of cattle that have been in individual pens are then moved into the chambers and the individual pens hosed clean in similar manner. A 2 m x 1 m rubber matt is located in the centre of each pen and chamber and the floor of the shed is sealed with a 2-part epoxy paint containing gravel aggregate to ensure cattle get adequate grip on the floor.

2.5 Chamber airflow piping and measurement

The respiration chamber air flow is reliant upon negative pressure in the system, achieved by high pressure fans placed at the exhaust of the system. A common stream of ambient air is drawn through a 30 cm circular galvanised iron duct from the outside of the shed (on eastern end) running above the mid line of the two rows of chambers. This duct is suspended from the roofing purlins of the shed and 10 x 150 mm diameter outlet droppers are positioned to provide the air intake above each chamber (Plate 2). From the duct to the chamber, a flexible (100 mm diameter x 2.5 m) steel reinforced flexible hose is used to connect to the top of the chamber, immediately above the feed bin and chamber entrance.

Inside the chamber, air is mixed by an oscillating fan (Plate 3) mounted on the roof at the rear of the chamber. On the roof behind the fan, a 100 mm outlet connected to the same type of flexible reinforced hose takes air from the chamber up 3 m to a 100 mm diameter PVC pipe running from directly above the chamber to directly above the analysis room. The length of PVC pipe varies from 22 m to 27 m reflecting the distance from that chamber to the analysis room. Above the analysis room, all PVC pipes are reduced to 50 mm (Plate 5) to thread directly onto a flow control manifold (Plate 6) composed of 10 mass flow meters fitted before individual gate valves which can regulate flow. Flow meters are from Fluid Components International, (Model ST75V). A sampling port to take 6 mm sample hose is tapped into each flow manifold immediately above the flow meter and a matching sample hose is connected to the air intake duct to provide a sample of ambient (incoming) air, giving 11 sample lines in total.

The common exhaust from the sample manifold is a 150 mm diameter PVC pipe that is connected by PVC pipe to the high flow fans that draw air through the systems. These are 2 x Aerovent HPE400 3-phase fans placed in parallel. One fan is fitted with a TECO Speecon 7200 inverter to provide variable speed control. The outflow from the two fans is combined and exhausted through the building roof.

2.6 Sampling, sample conditioning and analysis

Each of the 11 sample lines are connected to its own continuous flow pump drawing approximately 1 L of sample/min. These pumps are mounted above the refrigeration cabinet and pump (push) sampled air down through stainless steel coils contained in a refrigerator at 1–3°C (Plate 7). At the bottom of each cooling coil is a water trap to remove condensed water. The exit of each water trap is fitted with a non-return valve.

The 11 streams of dry air are pushed (using approx 3 m of 6 mm tubing) to the inlet ports of a sample multiplexer (Plate 8). In the multiplexer are two manifolds fitted with vacuum solenoids and in the closed position, the gas streams are vented from the multiplexer to waste. In this way, fresh air from each chamber is always passing through the multiplexer so there is only approximately 1 m of dead space to be purged between multiplexer and analyser for each analysis.

Plate 4: Inside of chamber (before protective film was removed from polycarbonate) showing internal pen that protects the chambers from damage by cattle. Also 40cm fan for internal air circulation. The chin closure system on the gate (visible) has since been replaced by a spring-loaded catch for operator safety.

Plate 5: Air ducting system from the 10 chambers is reduced from 10 cm o.d. to 5 cm o.d. at the point of entry in to the analysis laboratory and manifold. Exhaust is drawn to the high pressure fans by the 15 cm o.d. PVC pipe to rear.

Plate 6: Flow measurement manifold featuring 10 mass flow meters with digital output and gate valves to control flow.

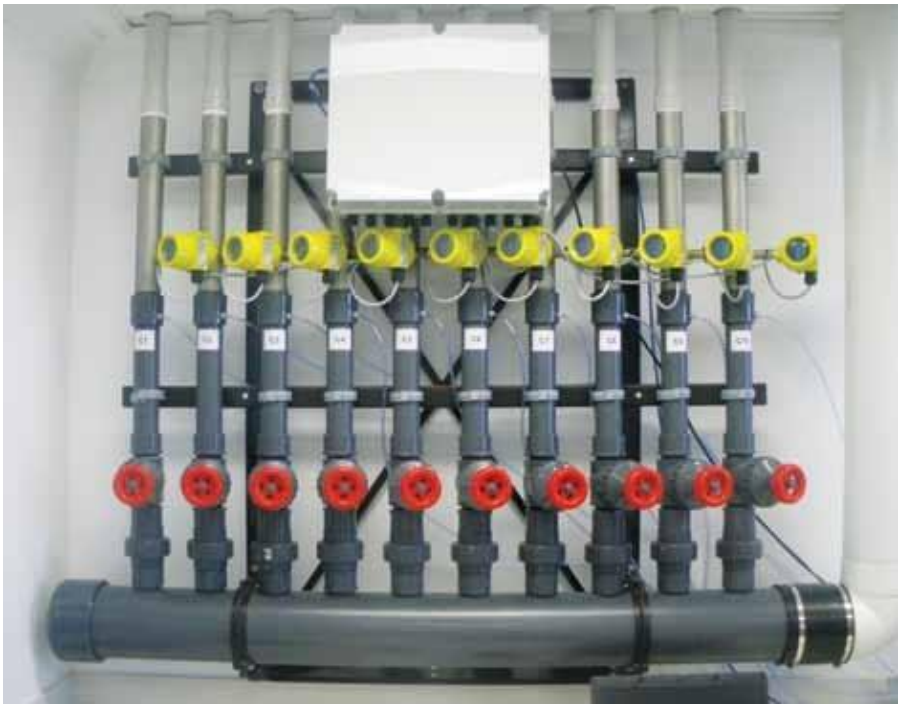


Plate 7: Sample drying system. Sample lines (10 sample + ambient) are drawn from the flow control manifold (Plate 6 above) at 1L/min by pumps mounted above the refrigerator (at top), which push the air through the refrigerated cooling coils which have autodrains of condensed moisture at the base.

The multiplexer is operated by a touch screen that allows any or all chambers to be included or excluded from sampling and to set the duration of sample purge and sample measurement. When the solenoid for a chamber (or the ambient line) is open, dried air from that sample line is directed to the sample outlet of the multiplexer (instead of the common vent) and 1 m of tubing connects the sample outlet to a pump (Vacuubrand ME1) which then draws 6 L/min from the flow manifold, through the 1 L/min pump, down through the refrigerated drier, through the multiplexer and pushes it into the flow control module. In so doing, this pump generates a pressure of 2 kPa that is tightly regulated in the flow control module (control valve is SMC, model KLF IBCA412A60000) after which 2 L/min is directed to a an outlet to connect to the CO_2 and CH_4 sensors in the analyser (in series), and 200 ml/min is directed to an outlet which will connect to the analyser's O_2 sensor.

The analyser itself is a Servomex model 4100C1 fitted with infrared detectors for methane (GFx1210. 500 ppm) and carbon dioxide (IR1520, 1% CO_2), and a paramagnetic sensor for oxygen (PM1158). These are spanned to apply the 4-20 mA signal between a low standard (0 ppm CH_4, 0 ppm CO_2, 16% O_2) and a high standard (98 ppm CH_4, 1010 ppm CO_2, 20.9% O_2). Calibration of the analyser is done daily for each gas using the mentioned standards, with the calibration gases being dispensed at 10 psi from the cylinder into the flow control module

2.7 Gas recovery test

Recovery of methane through the chamber system is quantified following a single injection of methane as follows. With the high pressure fans drawing air through the set of chambers, the air intake and exhaust pipes of a single chamber are temporarily sealed but the internal fan is left running. This circulates air within the chamber but no air is able to escape the chamber. A fixed volume of methane (1200 ml at ambient conditions) is injected near the internal mixing fan via a portal from outside and allowed to mix within the chamber for two minutes. After this time, the intake and exhaust pipes are opened to allow flow of air into and out of the chamber. The exhaust gas stream is sampled as occurs during routine operation, but sampling is every 50 seconds for approximately 20 minutes. Sampling time and methane concentration are recorded and the sampling time adjusted for the lag time between air leaving the chamber and arriving to the analyser (approximately 22 seconds). The methane concentration follows a logarithmic decline ($[CH_4]t = ae^{-kt)}$ so a linear regression can be fitted between the natural log of methane concentration verses the time of sample ($r^2>0.99$).

From this fit, three assessments can be made:

RECOVERY OF ADDED METHANE. From the regression, the total quantity of methane estimated to be present at the moment of methane injection can be calculated by multiplying area under the curve x the flow rate. This quantity is then expressed as a percentage of the known dose of methane injected. A failure to achieve acceptable recovery (98-102% of added CH_4) can occur due to errors in either methane measurement or flow rate measurement and these can be differentially diagnosed as below:

METHANE CONCENTRATION. By taking the antilog of the intercept from the linear regression, the concentration of methane in the chamber at the moment the exhaust and intake pipes were opened is estimated. This can be compared directly with the 'expected' methane concentration at this time, which is derived by dividing the methane injected (1200 ml) by the volume of the chamber (20,000 L). If the two do not match, then there is (a) a loss of methane from the sealed chamber or (b) some error in methane measurement as there is little error in the methane dose or the volume of the chamber which is based on measurement of its internal dimensions.

FLOW RATE. The linear regression provides a 'k' value or rate constant describing the decline in methane concentration, measured in terms of chamber volumes/minute. The theoretical k value (measured flow/min as shown on the mass flow meters divided by chamber volume) is calculated with adjustment for temperature and pressure (Data Harvest Group Ltd.) to a k vale for the ambient conditions of the day. If the theoretical and regression-derived k values differ, it identifies an error in the flow measured. This error could arise from leaks between the sample line and the flow meters (unlikely) or leaks into the sample line after the mass flow meter which allows air to dilute the sample. These leaks could be anywhere in the refrigeration drying unit, the multiplexer, flow control module or analyser plumbing. Sequential leak tests with nitrogen will be required to locate the leak.

2.8 Emissions calculation

Because the system relies on mass flow meters rather than dry gas meters, there is minimal data processing required. The 11 Excel spreadsheets capture sequential data (date, time, flow meter reading, and concentrations of three gases) in rows of a spreadsheet. An average gas concentration for each gas at each time is calculated. The concentration of gases in the ambient air stream at the start of that sampling cycle is removed. The values for net production/concentration of each gas over the period of study can simply be averaged and multiplied by the air flow over that time (final flow meter reading – starting flow meter reading). The output from this is gas production (or consumption) at normal temperature and conditions (0°C, 1.0 atmosphere pressure) that can then be converted to STP and so weight of methane produced.

Feed intake and DM content are recorded daily to enable emission to be reported as g methane and g methane/kg DM intake.

2.9 Animal welfare and operators' safety

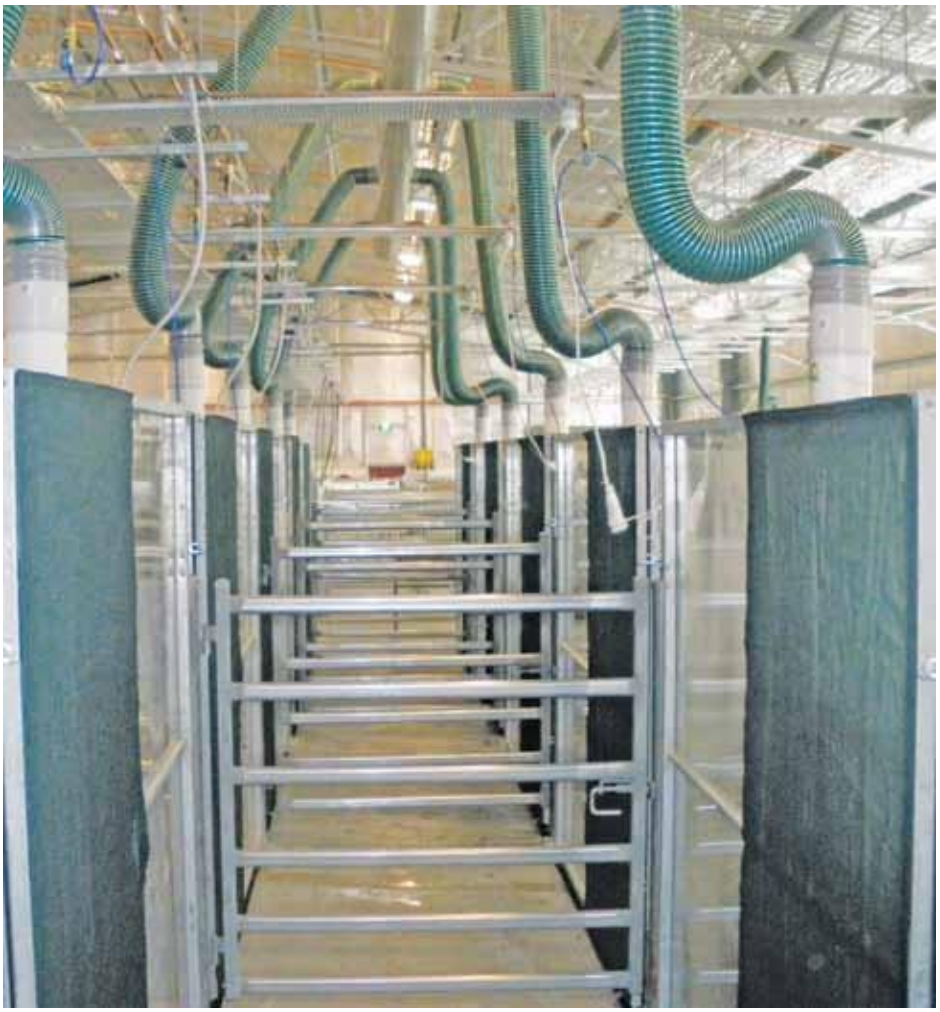
Operator safety

The chambers are designed to allow cattle to be walked into them, with safety of the operator being paramount. Two handlers are always present to move cattle. This is achieved by:

- A panel walkway linking the cattle crush/handling facility to the aisle between the two rows of chambers, and a separate return walkway back to the crush area (Plate 2).
- For getting animals into chambers, the chamber door (framed polycarbonate) and pen gate to one of the furthermost chambers is opened so the animal can only enter the chamber or return the way it has come. A hook on a cord is attached to the open pen gate so that the gate can be pulled shut from further up the aisle way. As the animal proceeds down the aisle, a series of gates with snap-lock fasteners can be shut behind it by the operator who follows on foot, preventing the animal's return (Plate 9).
- Once the animal is in the pen, the nearest aisle gate is shut and the chamber gate pulled closed by the hook and cord so that the spring-loaded chamber gate latch secures the animal in the chamber pen.
- The polycarbonate chamber door is then closed using two industrial snap latches.
- The door and gate on the opposite chamber are then opened and that chamber filled in like manner.

Plate 8: Gas handling and analysis system. The 11 dried sample streams are continuously pushed (1 L/min) through the gas multiplexer (bottom) which features a touchpad allowing the desired number of chambers to be selected for sampling, as well as their purge time and measurement time. Solenoid valves within the multiplexer allow one gas stream to be open for analysis, which involves sucking that sample (at 6 L/min) through the flow control module (middle box) which regulates pressure and creates differential flow to the 3 detectors. The individual sample flows are then directed to each detector in the Servomex 4100 analyser.

Plate 9: Aisle between chambers shown with all gates shut. These gates are all open to start and the door to the most distant chamber is opened. As cattle walk down the aisle, the gates shown can be progressively closed to ensure the animal does not come back to harm the operator following it down to the chamber. The individual sample flows are then directed to each detector in the Servomex 4100 analyser.



Animal welfare

The primary risk is asphyxiation in the event that air flow through the chambers stops (power blackout or fan fault), and this risk has been minimised by:

- Use of two high pressure air fans in parallel, so that failure of one fan will not stop air flow.
- Automatic lift of the chambers 20 cm above floor by pneumatic rams (as for cleaning) in the event of power failure or oxygen concentration falling below 18% in the chamber.

2.10 Weaknesses of the system

The system is new and has much scope for improvement for ease of operation. Key improvements anticipated are:

FLOORING IN CHAMBERS AND PENS. At present the cattle are standing on rubber mats and so become surrounded by their own excreta over time. We intend to put raised floors for the cattle to stand on for welfare reasons but also to facilitate hosing out.

TEMPERATURE AND HUMIDITY CONTROL. Currently there is no temperature or humidity control in the chambers. This needs to be established before energetic studies can be entered into. We are anticipating this will be individual air conditioning units on each chamber.

DATA MANAGEMENT. While functional, the data collection and processing software is still requiring considerable manual processing. We hope to get this automated quickly and will work with the designer to do so.

2.11 Description of components and equipment suppliers

Pen design, construction	Local contractors
Chamber design and construction	UNE Sciences workshop
Air ducting system and fans	Selves with local contractor
Mass flow, and all analysis hardware	AZCO holdings, Auckland NZ
Data handling software	AZCO holdings, Auckland NZ

2.12 Costing of the facility complete system with ancillary equipment

ITEMS	US$
LABOUR	
Design of the system	In house
Cattle pens within chambers (10)	15,000
Building of chambers	80,000 includes materials
Piping air circulation and sample lines	33,000 includes materials
Wiring, data acquisition, software development	Included in equipment cost
Monitoring and commissioning	Included in equipment cost
Tests	4 weeks
MATERIALS	
Building materials	In labour
Pipes, cables, etc	In labour
EQUIPMENT	
Minor assets (sensors, pumps, etc)	2,000
Mass flow meters (10)	34,000
Gas analyser	29,000 (CH4, CO2,O2)
Sample drying system	34,000
Gas switching system	Included in dryer cost
Calibration gases	3,000
Computer and data acquisition system	6,000
TOTAL COST	**236,000**

Technical Manualon Respiration Chamber Designs

Chapter 3: ILVO's Ruminant Respiration Facility, Melle, Belgium

AUTHORS
Sam De Campeneere and Nico Peiren

Contents

Chapter 3: ILVO's Ruminant Respiration Facility, Melle, Belgium

3.1 Summary

The large ruminant respiration facility at the Institute for Agricultural and Fisheries Research (ILVO) comprises six airtight chambers located in a separate room (15.7 m × 7.0 m) of a stanchion barn. Wind-breaking nets are installed in the air inlets. The incoming air is not heated or cooled. The six chambers are made of polypropylene (PP) panels mounted on a stainless steel frame. Chamber volume is 12.3 m^3. The chambers operate at a slight negative pressure, with a 200−1300 m^3/h air flow. The six chambers are arranged two by two, oriented side by side along the length of the room. All chambers are monitored by one system, which performs dedicated gas sample conditioning, gas analysis, data logging and animal monitoring. Gas samples from all chambers (six) and the ambient air (two) are continuously sampled at 4 L/hour and a gas switching system delivers a sample stream to the gas analyser at intervals that vary from 5−60 seconds, depending on the measured gases. A multigas analyser measures concentrations of CH_4, CO_2, N_2O, NH_3 and water. The gas analyser is self-calibrated, with gas recovery from each chamber tested routinely. This facility has been designed for rapid and efficient feeding, milking, cleaning and animal entrance and exit, so gas emissions are monitored for more than 95% of the time under normal operational conditions. Chambers are designed for large ruminants, but can also be used for monitoring small ruminants or mono-gastric animals. Animal welfare and comfort were taken into account when designing the chambers.

3.2 Location of the facility

The physical address of the facility is:

Institute for Agricultural and Fisheries Research (ILVO)
Animal Sciences Unit
Scheldeweg 68, 9090 Melle
Belgium

Mailing address:

Institute for Agricultural and Fisheries Research (ILVO)
Animal Sciences Unit
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The cow respiration chambers are located in a separate room of a stanchion barn of the Institute for Agricultural and Fisheries Research (ILVO) Animal Sciences Unit site located in Melle, Belgium. ILVO employs about 560 personnel, whereas the Animal Sciences Unit site in Melle employs about 90 persons. The experimental farm houses approximately 90 dairy cows with young stock, 75 suckling cows with young stock, 140 sows with piglets and fattening pigs (1000 in total) and 4500 broilers and 1500 laying hens. The research complex (50°59'02"N, 30°46'49"E) is located close to Melle (3 km; 11 000 inhabitants) and Ghent (10 km, 245 000 inhabitants). The stanchion barn, housing the chambers, is surrounded by pasture and other barns. There are no major industrial sites within 5 km of the facility.

The stanchion barn housing the chambers was built in 1971. The respiration room was modified in 2011. It is a brick building with a concrete frame; only the roof and part of the walls are insulated. No cooling or heating system is available. Wind-breaking nets are installed in the air inlets.

The facility houses six chambers for large ruminants in one room (Plates 1 and 2). A separate room houses instrumentation for sampling, instrument calibration, measurement and data handling. Feed preparation and cleaning facilities are nearby (in the same building), as are animal pens and equipment required for handling livestock. The six ruminant chambers are situated two by two, oriented side by side along the length of the room. The room is 15.7 m long, 7.0 m wide and between 3.8 and 5.1 m high. Animal access from the adjacent tie-stall is through a large sliding door. One door provides direct access between the room and the outside. The room has two window openings for an air inlet (Plate 3). The system layout is presented in Plate 4.

3.3 Description of the chambers structure

The respiration chambers were designed not only to enable accurate measurement of methane emissions but also of carbon dioxide, nitrous oxide and ammonia, and to provide a comfortable and safe environment for the animals. The conceptual design was based on existing chambers in The Netherlands and United Kingdom, but was thoroughly revised. Each chamber can house one large ruminant (dairy or beef cattle), but can also be adapted to house smaller ruminants and monogastric animals. Each chamber provides sufficient room for the animal while allowing rapid air exchange (1–4 minutes depending on the ventilation speed).

The volume of each chamber is 12.3 m_3, with outside dimensions of 4.0 m length, 1.55 m width and 2.8 m height, and a total weight of 1 tonne (Plate 2).

The chambers are constructed with 50 mm thick polypropylene (PP) copolymer panels (Paneltim, Belgium) mounted on an internal frame made of 80 x 80 mm stainless steel tubing (Plate 5). The panels are welded together to make the construction airtight. The construction company was Protherm (Schijndel, The Netherlands). Each chamber has three doors: an entrance door in the back, a lateral door for milking the dairy cows and a front door for feed supply, which is also the animal's emergency exit. To reduce the feeling of captivity and improve visual contact between cows, natural lighting in the chambers was maximized by using windows of 6 mm polyethylene terephtalate glycol (PETG) in each door and in the side panels.

Additional fluorescent tubes outside the chambers increase the lighting to 50 LUX inside the chambers (for animal welfare). The three other openings in the chamber are the manure tray situated under the back door, the air inlet (67 × 97 cm) in the front door and the air outlet (diameter 35 cm) in the rear part of the roof. Nine beams are welded under the floor

Plate 1: Drawing of ILVO's Ruminant Respiration Facility composed of six chambers.

Plate 2: Four of the six chambers of ILVO's Ruminant Respiration Facility.

Plate 3: Openings for air inlet, equipped with a fixed wind-breaking net with a porosity of 30% on the inside and an adjustable tarpaulin on the outside.

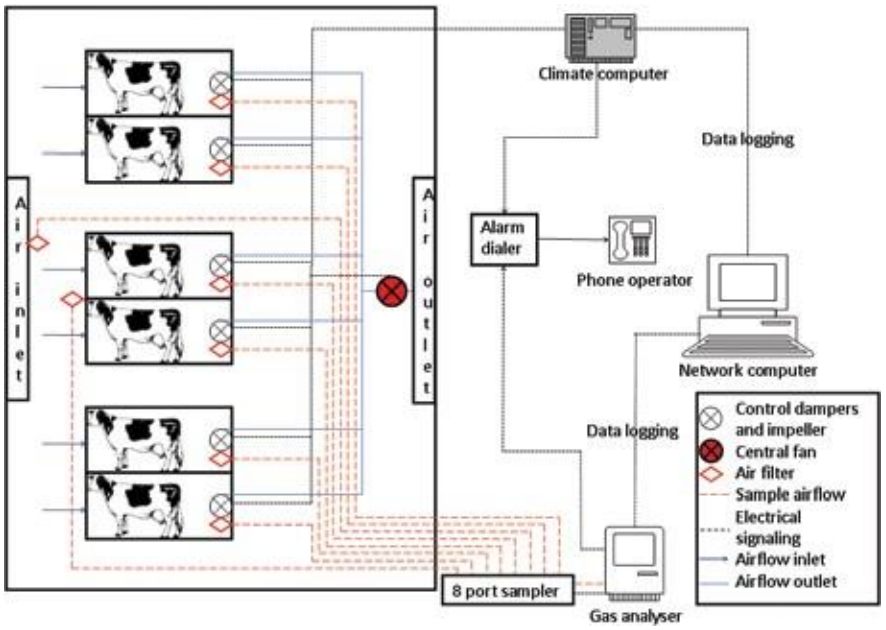

Plate 4: Layout of the ruminant facility. The system integrates 8 respiration chambers.

Plate 5: Inside view of a chamber made of polypropylene mounted on a stainless steel frame.

to make the chamber movable using a forklift or pallet jack. Inside the chamber, the floor is raised by 35 cm to allow for a manure tray in the rear part of the chamber (145 cm in the back). A custom made metal slatted grid is installed above the tray (Neirinck, Belgium) (Plate 5). In front, a polypropylene feed bin, 90 x 55 x 115 cm (inside dimensions) is placed with an opening for eating. A drinking bowl (F30A, La Buvette, France), with non-spill edge and water meter is attached to the side wall. To optimise the cow's comfort a Kew plus mat (Kraiburg, Germany), 183 × 130 cm, is placed on the floor. Cows are tied with a vertical chain tying system (where the tether junction could move up and down along a vertical girth/belt). The milking door is protected with bars with built-in springs for easy access (Plate 5).

3.4 Animal holding, feeding and cleaning

The group of cows is allowed to acclimate in the tie-stall for one week. The animals are accustomed to standing in a tie-stall during winter. The cows enter the chambers through the back door using a step mounted on a pallet jack and are then tied in the chambers (Plates 6 and 7). Installing the six animals in the chambers, including weighing, takes 30 minutes. Feeding, milking and cleaning the manure trays is done twice daily at fixed times. It takes two technicians 1 hour to complete.

The PETG windows allow the cows to see each other, which helps them to acclimatise quickly to the system as indicated by either eating, ruminating or lying. The chambers have a feed bin and a drinking water bowl, both easily accessible for the animal and suitable for mature lactating cows. Animals are usually kept in the chamber for two to four days.

The daily procedure enables more than 22 h of gas measurements to be made. The normal procedure is to milk the cow via the side door, to open the front door to remove any feed residues and to clean the (removable) feed bin. New feed is added, and the water supply is checked. At the rear, the faeces and urine collection tray is removed and replaced with a clean one. The removable exchangeable manure trays enable rapid cleaning of faeces and urine and minimises the daily frequency of opening the chambers. The doors are then closed and the chamber is allowed to equilibrate. Feed is usually provided twice daily, when lactating cows are milked, and leftover feed is removed once a day. We strive to minimise such refusals, by feeding just below maximal voluntary intake. Feeding is usually done at 0800 h and 1900 h.

After each experimental period, the cows leave the chamber and rooms are cleaned with a pressure washer.

3.5 Chamber airflow piping and measurement

The ventilation system is a temperature controlled mechanical central flow system (Plates 1, 4). The airflow through each chamber can range from a minimum 200 to a maximum 1300 m^3/hour (about 3 300–21 500 L/minute). The fresh air enters the room through two windows (2.3 × 1.4 m) equipped with an adjustable tarpaulin on the outside and a fixed wind-breaking net with a porosity of 30% on the inside.

The air enters each chamber via an adjustable opening (maximum 67 × 97 cm) in the lower panel of the front door, where chamber gas concentrations are monitored at 1 to 8-minute intervals, depending on the gases being measured. The air outlet is installed in the roof panel at the rear. A ventilation module with a diameter of 35 cm (ATM unit 35, Fancom, The Netherlands) is placed in the opening (Plate 8). This module connects with a 12.6 m long central ventilation duct (121 × 59 cm) made of coated plywood (Plate 1). Finally, the air is evacuated by an axial exhaust fan (FAN IF56M, Fancom, The Netherlands), with a maximum ventilation rate of 12 000 m^3/h, fixed in a chimney. In this way, one central exhaust fan creates

Plate 6: Cow enters the chamber through the back door using a movable step.

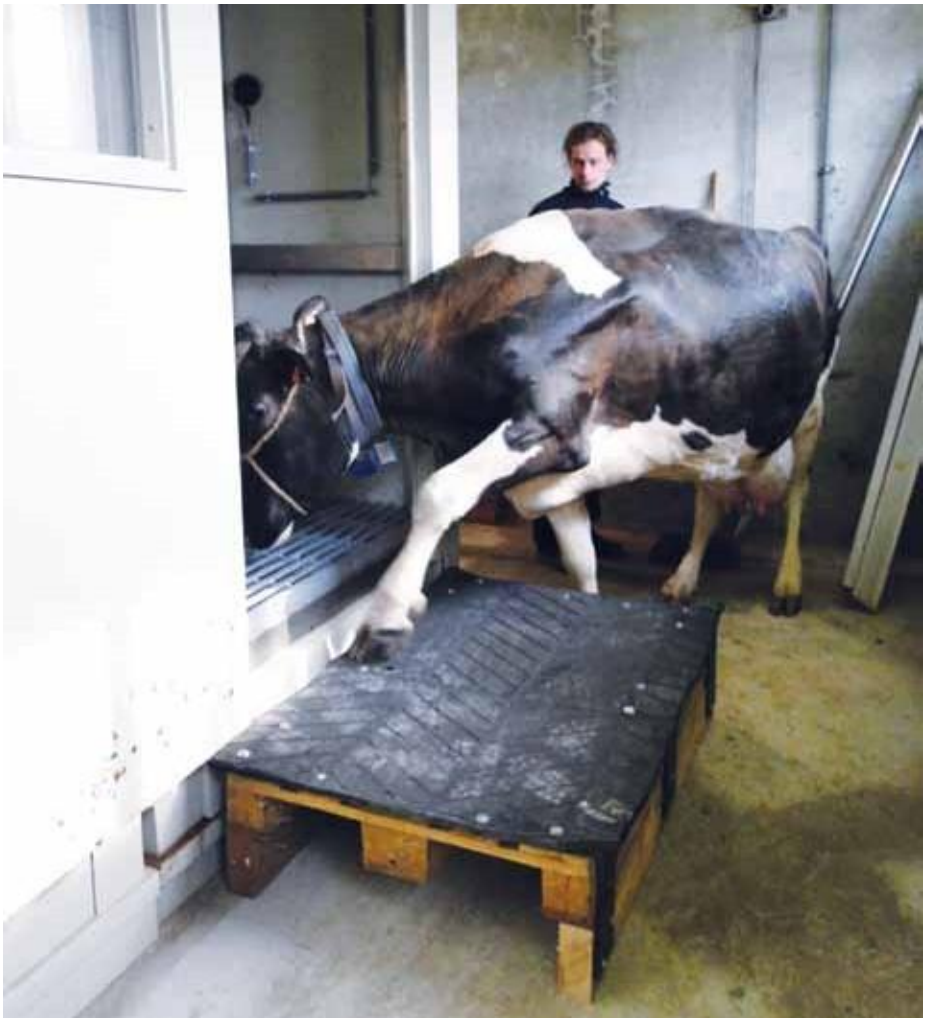

Plate 7: Cow tied in the chamber.

Plate 8: ATM-module with vortex damper **(grey)** and integrated impeller **(black)**. Gas sample point in ATM-module with sonic nozzle and 7 µm filter **(left)**.

Plate 9: Ventilation computer **(left)**, battery ventilation system **(right under)**, Octalarm **(right above)**.

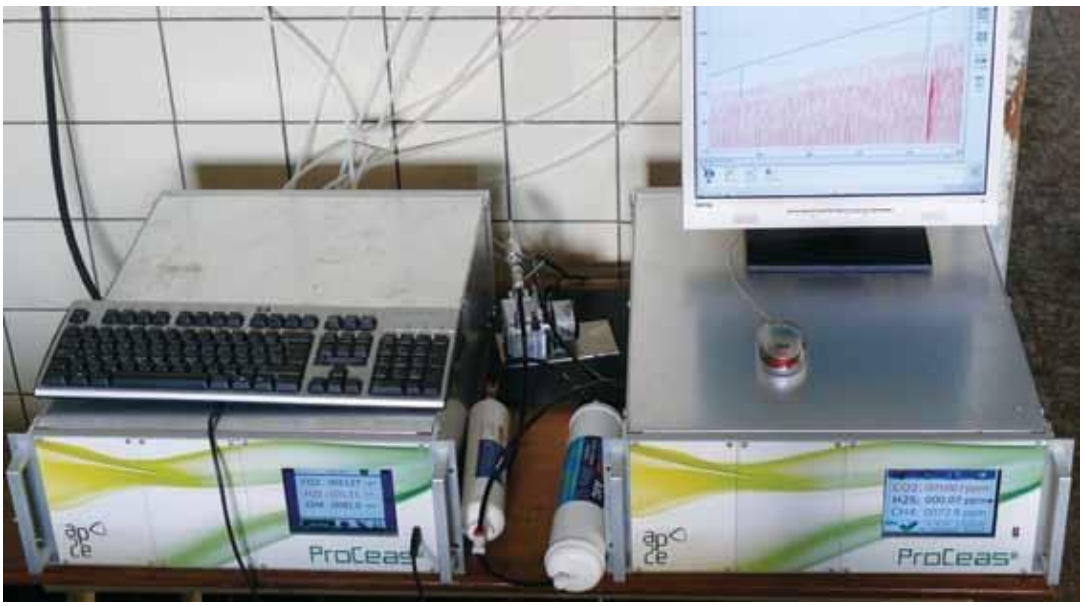

Plate 10: ProCeas gas analyser **(right)** and 8-channel multi sampler **(left)**.

air flow through all six chambers. However, each chamber has its own ATM module with a control damper that regulates the amount of air, and an integrated calibrated full size free-running impeller that continually measures the airflow. In this module the gas concentrations are measured every 1 to 8 minutes, depending on the measured gases. The chamber with the highest demand determines the speed of the central fan. To achieve the correct volume of air in chambers with a lower demand, the vortex damper damps the air exhaust in these chambers. The unique aspect of the central flow control is that it does not simply take the section with the highest demand into account, but also continually checks the positions of all the dampers. Because of this, all the chambers have enough fresh air without any unnecessary damping. This system is managed by a F21 ventilation computer (Fancom, The Netherlands), which is temperature controlled (Plate 9). Each chamber is fitted with a sensor, close to the outlet (Fancom, The Netherlands), to monitor changes in temperature (Plate 2).

3.6 Sampling, sample conditioning and analysis

The gas concentrations of CH_4, CO_2, N_2O, NH_3 and water are measured with an infrared laser optical-feedback cavity-enhanced absorption spectrometer (IR-OF-CEAS) (ProCeas, AP2E, France) (Plate 10) in the exhaust channels in the ATM module between the impeller and the vortex damper. See Morville et al. (2005) for the principles of this technique. In each ATM-module a sample point is placed, with a stainless steel sampling probe (AP2E, France), equipped with an inline 7 µm filter and a sonic nozzle (critical flow venturi), right next to the module. The six probes are connected to an eight-channel multi sampler (AP2E, France) via 25 m of 6-mm PFA tubing. The background air concentration is measured every 1 to 8 minutes on two locations in the room, depending on the measured gases. The two probes are connected to the multi sampler as described above. The sampler is connected to the gas analyser (Plate 4). The cycling time to measure gas concentrations from the eight sampling points is completed within an 8-minute period if all gases are measured, this because of the memory effect of ammonia. If CH_4, CO_2 and N_2O gases are only measured this can be done correctly within 1 minute with three repeats per measure point. The whole measuring system functions with a continuous negative pressure of 110 mbar. This system has several advantages: no condensation, no heated lines, excellent compositional sample representation, low response time, no false positive responses, high selectivity, a measuring range from ppt to ppm in one calibration, simultaneous measurement of several gases using the same laser spectrometer and no need for coolers or driers.

The detection ranges for CH_4, CO_2, N_2O and NH_3 are 0–700 ppm, 0–5000 ppm, 0–5 ppm, and 0–70 ppm respectively, with corresponding accuracies less than 1% of the full scale. If necessary, the maximum detection limits can be lowered, to improve the accuracy of the measurements.

The gas analyser has no instrumental drift and because of the self-calibrating direct measurement, calibration is checked every 2–3 months using two standards containing a mix of gases: CH_4, 200±4 ppm; CO_2, 2000±40 ppm; N_2O, 2±0.1 ppm in N_2 and NH_3, 30±0.9 ppm in N2. The calibration gases are supplied by Air Liquide Belge (Liege, Belgium).

3.7 Gas recovery test

Chamber methane recovery tests are performed every two-three months in each chamber or if changes are made. When the system is running, pure methane (99.9%) is bled in the chamber at a rate to achieve a concentration in the air of 100 ppm. This is done through a gas mass flow meter with display (Bronkhorst High-Tech, The Netherlands), connected to a gas cylinder, for 30 min at minimum and maximum airflow rate.

3.8 Emissions calculation

Calculations of enteric emissions of gases are based on measurements of the chamber ventilation rate (Q), the net concentration of gas (C) (above the background concentration), and the percentage of gas recovery in the entire system (if implemented for this gas).

The method to measure concentrations of gases with IR-OF-CEAS gives a "wet" measurement: the air pressure is reduced to 120 mbar, but the moisture content remains volumetrically the same and the composition does not change. The dew point decreases, but that is a physical phenomenon which not affects the volumetric proportions.

In the following formulas, there are no corrections needed for temperature and atmospheric pressure, because the ventilation rate is measured with an impeller which gives a direct volumetric measurement and the gas concentrations can be displayed under standard temperature and pressure (STP; 273.15 K, 1013.25 hPa) on IR-OF-CEAS, because pressure and cell temperature are continuous monitored.

Formula "wet" emission:

$$E_{wet\ STP} = Q_{wet} \mathbf{x}\ [(C_{wet\ i} - C_{wet\ o}) \mathbf{x}\ 10^{-6}] \mathbf{x}\ \frac{M}{V_m}$$

$E_{wet\ STP}$ = emission concentration under wet standard temperature and pressure (g h^{-1})

Q_{wet} = ventilation rate (m^3 h^{-1})

$C_{wet\ i}$ = gas concentration air outlet (ppm)

$C_{wet\ 0}$ = gas concentration air inlet (ppm)

10^{-6} = conversion ppm (parts per million)

M = molar mass gas (g mol^{-1}; CH_4, 16.042; CO_2, 44.410; NH_3, 17.031; N_2O, 44.013)

Vm = molar volume gas at 0°C and 1 atm (0.022414 m^3 mol^{-1}), $V_m = \frac{R \mathbf{x} T_{STP}}{P_{STP}}$

R= 8.314472 10^{-2} m^3 hPa K^{-1} mol^{-1}

T_{STP}= standard temperature (273.15 K)

p_{STP}= standard atmospheric pressure (1013.25 hPa)

Formula "dry" emission is:

$$E_{dry\ STP} = E_{wet\ STP} \text{ x } \left(\frac{100}{100 - \%\ H_20} \right)$$

$E_{dry\ STP}$ = emission concentration under dry standard temperature and pressure (g h^{-1})

$E_{wet\ STP}$ = emission concentration under wet standard temperature and pressure (g h^{-1})

% H_2O = % H_2O in air sample

Formula after recovery correction (if performed):

$$E_{dry\ STP\ grr} = \frac{E_{dry\ STP}}{grr}$$

$E_{dry\ STP\ grr}$ = emission concentration under dry standard temperature and pressure with correction for gas recovery rate (g h^{-1})

$E_{dry\ STP}$ = emission concentration under dry standard temperature and pressure (g h^{-1})

grr = % percentage gas recovered during recovery test

Ventilation rate, gas concentrations and environmental parameters are continuously monitored. When these data are brought together, an emission value for any chamber within the system can be calculated every minute if the focus is on CH_4, CO_2, and/or N_2O, and every 8 minutes if NH_3 is taken into account.

Missing data, from the moment the chambers are opened until they are stabilised, are estimated by interpolation based on 20 values (20 minutes) immediately before the chambers were opened.

Once instantaneous gas emissions (g h^{-1}) are calculated for each time interval for each chamber, and the missing values have been estimated, the daily emission (for a 24-hour period) from a particular animal housed in a given chamber is calculated by time integration (area under the curve) and expressed in g d^{-1}.

3.9 Animal welfare and operators' safety

All animal experimentation must conform to good ethical considerations and are approved by the ethical commission of our institute. These chambers are designed to maximise both animal and operator safety.

The system ensures that all environmental conditions are within the thermoneutral zone for the animals, with minimal exposure to stress or risk. The system is monitored for temperature, air flow, water and gas concentrations. Alarms will activate when abnormal conditions are detected (Plate 9). If the conditions fall outside the following threshold values, the alarm system goes off and an operator is warned: temperature (higher than 26 °C), air flow (lower than 200 m^3/h), and CO_2 (higher than 5000 ppm).

Because the chambers are airtight and operate under a slight negative pressure, the most critical risk to animal safety is lack of ventilation. Consequently, power failure or malfunctioning of the ventilation system could cause CO_2 intoxication. Two safety measures have been incorporated to overcome this risk:

1. In the event of power failure, the system is designed that the damper valves of the ATM-module open completely; by this a natural chimney effect is created so the CO_2 concentrations are not harmful.
2. The respiratory chamber system is connected to ILVO's computer network, which enables remote monitoring. Abnormal readings trigger telephone message notification to the operator or caretaker. These personnel can respond within five minutes.

Operator safety is also important. Although stainless steel frame construction is more expensive than steel, the lack of rust and easy cleaning lowers the risk of injuries and reduces labour costs. The stainless steel bars in the milking door also reduces risk of injury to the milkers and the animals. The movable step has worked very well, as has a removable manure tray (Plate 11).

Plate 11: Removable manuretray with wheels on pallet jack.

3.10 Weaknesses of the system

The main weakness of the system is the inability to provide uniform environmental conditions because it lacks conditioned air. Air-conditioning in the room can be installed if it should be necessary or an air duct with conditioned air can be attached at the air inlet of each chamber.

Polypropylene insulates quite well but double glazing would improve the insulation even more.

A typical calorimetric chamber could be more precise, but would be much more costly. No airlock exists between the room and the chamber.

A feed weighing device is absent but can be incorporated with minor changes. Fans inside the chamber

would mix the air better, with faster response time.

The chosen milking system, with tubing outside the chambers, makes it possible to work with dairy cows, but the milking door has to be opened during milking.

3.11 Description of components and equipment suppliers

Chamber structure

- Chamber frame is 80 x 80 x 3mm stainless steel tube
- Polypropylene plates; Paneltim, Lichtervelde, Belgium
- Construction structure chamber; Protherm, Schijndel, The Netherlands
- Drinking bowls F30A; (La Buvette, Charlesville-Mezieres, France), distributed by Laborim, Kortrijk, Belgium
- Custom made manure grid, Neirinck, Ruiselede, Belgium
- KEW plus laying mat; (Gummiwerk Kraiburg Elastik GmbH, Tittmoning, Germany), distributed by Vanloot, Vlamertinge, Belgium

Air Circulation and pumps

- Ventilator fan, FAN IF56M, Fancom, Panningen, The Netherlands Damper with intergrated impeller, ATM unit 35, Fancom, Panningen, The Netherlands
- Managing computer, F21, Fancom, Panningen, The Netherlands
- Chimney; Fancom, Panningen, The Netherlands.
- Fancom is distributed by De Jaeger, Aalter, Belgium

Analyser, sampling and sample conditioning

- PFA tubing 6 mm for sampling; (Du Pont de Nemours, Mechelen, Belgium), distributed by CW-Technics, Lokeren, Belgium
- Gas exchange unit: 8 channel unit; (AP2E, Aix-en-Provence, France), distributed by CW-Technics, Lokeren, Belgium
- Gas analyser: ProCeas; (AP2E, Aix-en-Provence, France), distributed by CW-Technics, Lokeren, Belgium
- Micro-diaphragm pump for sample delivery to analyser: N813.3; (KNF Neuberger Inc, Freiburg, Germany), distributed by CW-Technics, Lokeren, Belgium
- Mass flow meter: EL-FLOW F-201CV-5K0-AGD-22-V; (Bronkhorst High-Tech, Ruurlo, The Netherlands) distributed by Gefran Benelux, Olen, Belgium

Data acquisition, logging and alarm system

- Standard PC, tower case (full height PCI slots to accommodate relay cards).
- Hardwire card integrated in ProCeas for communication with alarm system
- Web Link Box; Fancom, Panningen, The Netherlands
- F-Central Farm Manager; Fancom, Panningen, The Netherlands
- Backup Power Supply: 1000VA UPS
- Telephone alarm, Octalarm; (Adesys, Wateringen, The Netherlands) distributed by De Jaeger, Aalter, Belgium

References

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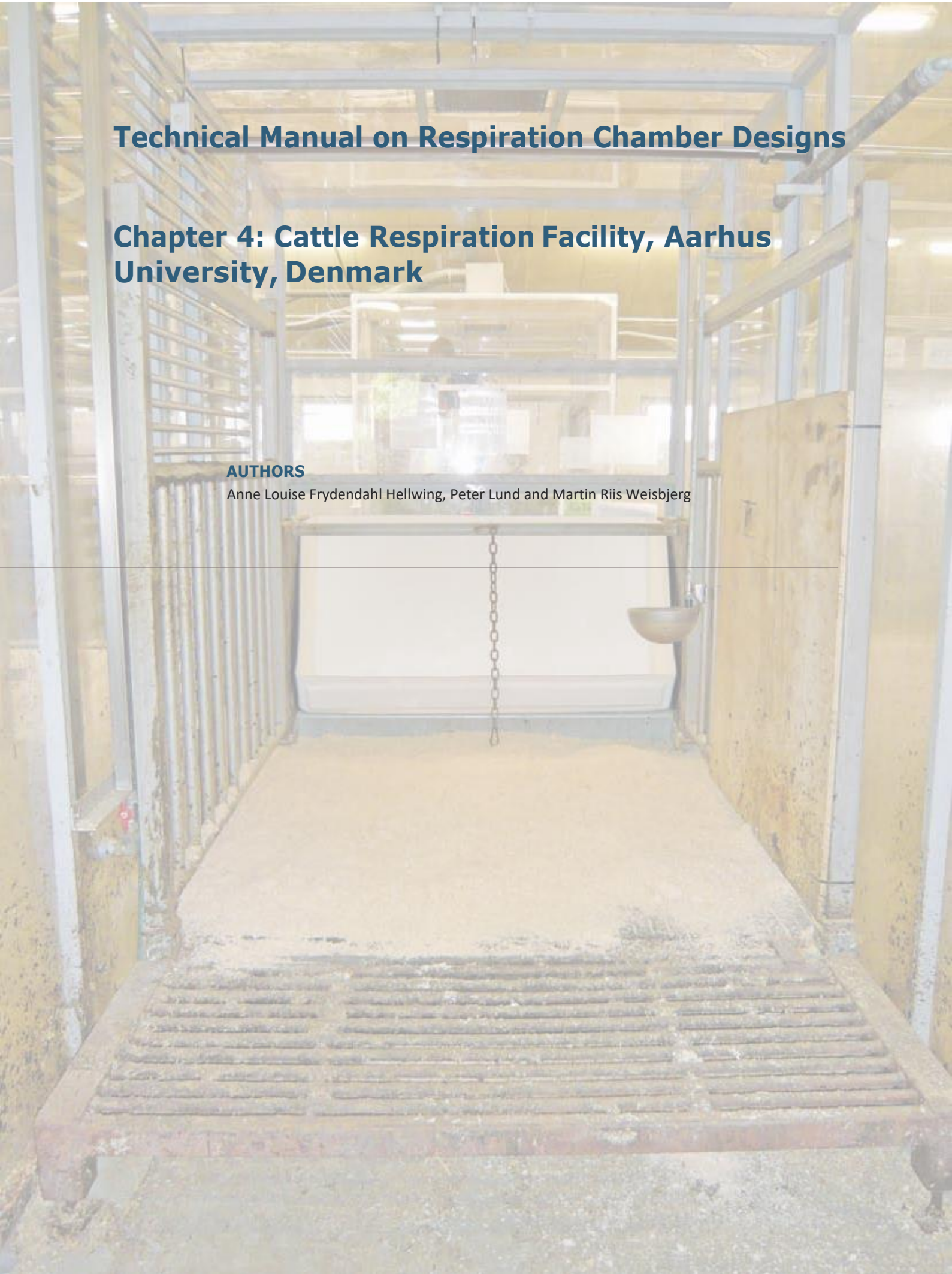
3.12 Costs of the facility

A complete system six ruminant respiration chambers and the ancillary equipment required#.

ITEMS	EURO	USD
LABOUR	**48,000**	**67,600**
Design of the system	3,000	4,200
Building chambers	20,000	28,000
Ventilation system	5,000	7,000
Wiring, data acquisition, software	5,000	7,000
Tests	4,000	5,600
Monitoring	7,000	9,900
Milk tubing	1,000	1,400
MATERIALS	**51,000**	**72,000**
Building materials chambers	29,000	41,000
Ventilation system	15,000	21,000
Tubing, pipes, cables, etc	3,000	4,200
Milk tubing	4,000	5,600
EQUIPMENT	**7,300**	**10,300**
Minor assets (sensors, pumps, etc.)	2,000	2,800
Gas analyser	45,000	63,000
Gas switching system	22,000	31,000
Calibration gases	1,000	1,400
Computer and data acquisition system	3,000	4,200
TOTAL COST	**170,000**	**387,200**

Excludes value added tax.



Technical Manual on Respiration Chamber Designs

Chapter 4: Cattle Respiration Facility, Aarhus University, Denmark

AUTHORS
Anne Louise Frydendahl Hellwing, Peter Lund and Martin Riis Weisbjerg

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Chapter 4: Cattle Respiration Facility, Aarhus University, Denmark

4.1 Summary

In Denmark, the emission rate of methane from dairy cows has been calculated using the IPCC standard values for dairy cows in Western countries, due to the lack of national data. Therefore, four respiration chambers for dairy cows were built with the main purpose of measuring methane, but also emission of carbon dioxide, hydrogen and consumption of oxygen. The chambers are constructed of steel and polycarbonate. The outside dimensions of the chambers are 183 × 382 × 245 cm, and the volume is approximately 17 m^3. The air inlet is a small gap between the floor and bottom of the chamber. The air outlet is placed at the top of the chamber. The flow rate in the chambers is measured with a mass flow meter. The concentration of gases is measured every 12 minutes with a chemical hydrogen sensor, a paramagnetic oxygen sensor and infrared carbon dioxide and a methane sensor. The ventilation rate for dairy cows is between 800 to 1500 L/min depending on the milk production and liveweight. This gives an average concentration of 5000−6000 ppm of carbon dioxide and 500−600 ppm of methane in the chambers.

4.2 Location of the facility

The physical address of the facility is:

Department of Animal Science
Aarhus University
Blichers Allé 20
DK-8830 Tjele
Denmark

The mailing address:

Department of Animal Science
Faculty of Science and Technology
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The cattle respiration chambers are located at the research centre in Foulum near Viborg which is a part of the Faculty of Science and Technology at Aarhus University. The research centre employs 600 people divided into four different departments. The main campus is located 60 km south east of the centre.

The cow herd counts 200-250 milking Holstein cows plus heifers but no bull calves. The Danish Cattle Research centre is located 1 km from the centre and houses a herd of 210 Holstein and Jersey cows. There is a strong collaboration between the two institutes, and cattle from the Danish Cattle Research centre have been used for experiments in the respiration chambers.

The building housing the respiration chambers was built in 1984 with the purpose of intensive cattle studies (Barn K-33 – see Plate 1). The building is made of bricks with a roof of fibre cement. The walls and roof are insulated according to Danish regulations. The floor is made of concrete, and a slurry drain is placed behind the cows. The slurry drains are covered with gratings behind the cows and with concrete in other places. The slurry drain is emptied two to three times daily with a scraper. The buildings are connected with the other cow barns and feed preparation facilities.

The building can be heated during winter, and the barn has mechanical ventilation. The temperature in the barn during wintertime is normally kept at 14 °C.

The biggest room (K33-1) in building K33 measures 14.4 m x 19.4 m (280 m^2). The height is 2.25 m at the outer wall and 6.6 m in the middle. The volume is approximately 1160 m^3. The barn has room for 2 x 7 tied dairy cows, but normally only eight fistulated cows are housed in this barn section. The other end of the barn had enough room for four respiration chambers. The respiration chambers were established in this barn to reduce the risk of decreasing the feed intake when animals are moved from the tie stall to the chamber. The barn houses other cows, and the barn workers come in to the barn regularly to feed and milk the other cows. Instruments and computers are located in a laboratory right next to the chambers (Plate 1). The laboratory facilities in the barn are used for sample preparation and determination of dry matter.

4.3 Description of the chambers structure

The design of the chambers had to fulfil three criteria:

- Inexpensive to build.
- Ensure an environment without a negative impact on the animal welfare.
- The air inlet had to come from a gap between the floor and the chamber bottom.

The chambers are built for milking cows but calves larger than 250 kg can also be measured in the chambers.

The chambers are built around a platform which was designed for the housing of cattle during intensive metabolic studies (Plate 4).

The laying area is 170 cm long and 165 cm wide and is covered with a rubber mat. Behind the platform there is a slurry grate at 67.5 cm, and underneath the slurry grate a four wheel wagon is placed for manure and urine collection (Plate 5).

The feeding box is placed on a four wheel wagon (Plate 6) with a scale for online recording of feed intake. Water intake is also recorded online.

The chamber is constructed on a frame of 40 x 40 mm steel tube, built around the platform with little space to each side, in the front and behind the platform.

Plate 1: Sketch of the building containing the respiration chambers. The building is connected with the other cattle facilities with a central lane. The cattle facilities include barns for both loose and tied up cattle as well as barns for milking and feed mixing. The respiration chambers are placed in room K33-1 together with fistulated dairy cows. Sensors are placed in laboratory 1.

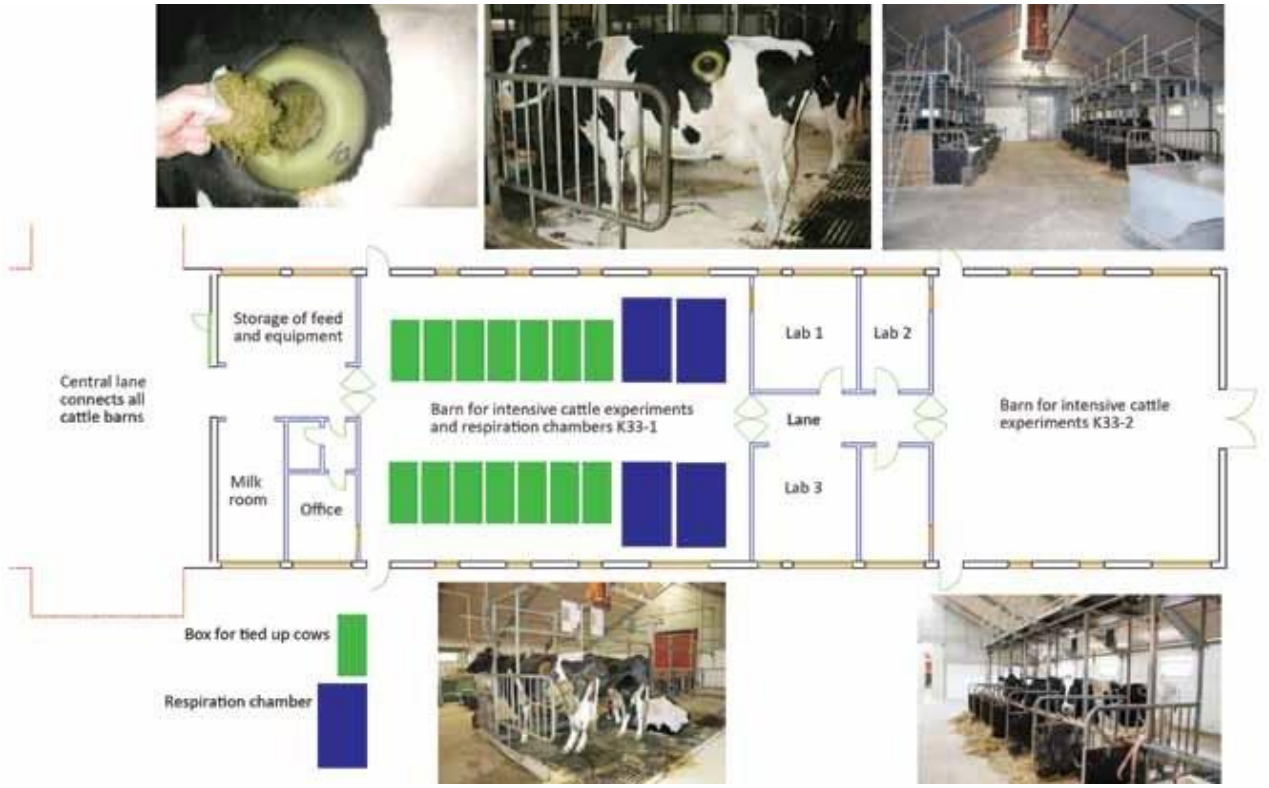
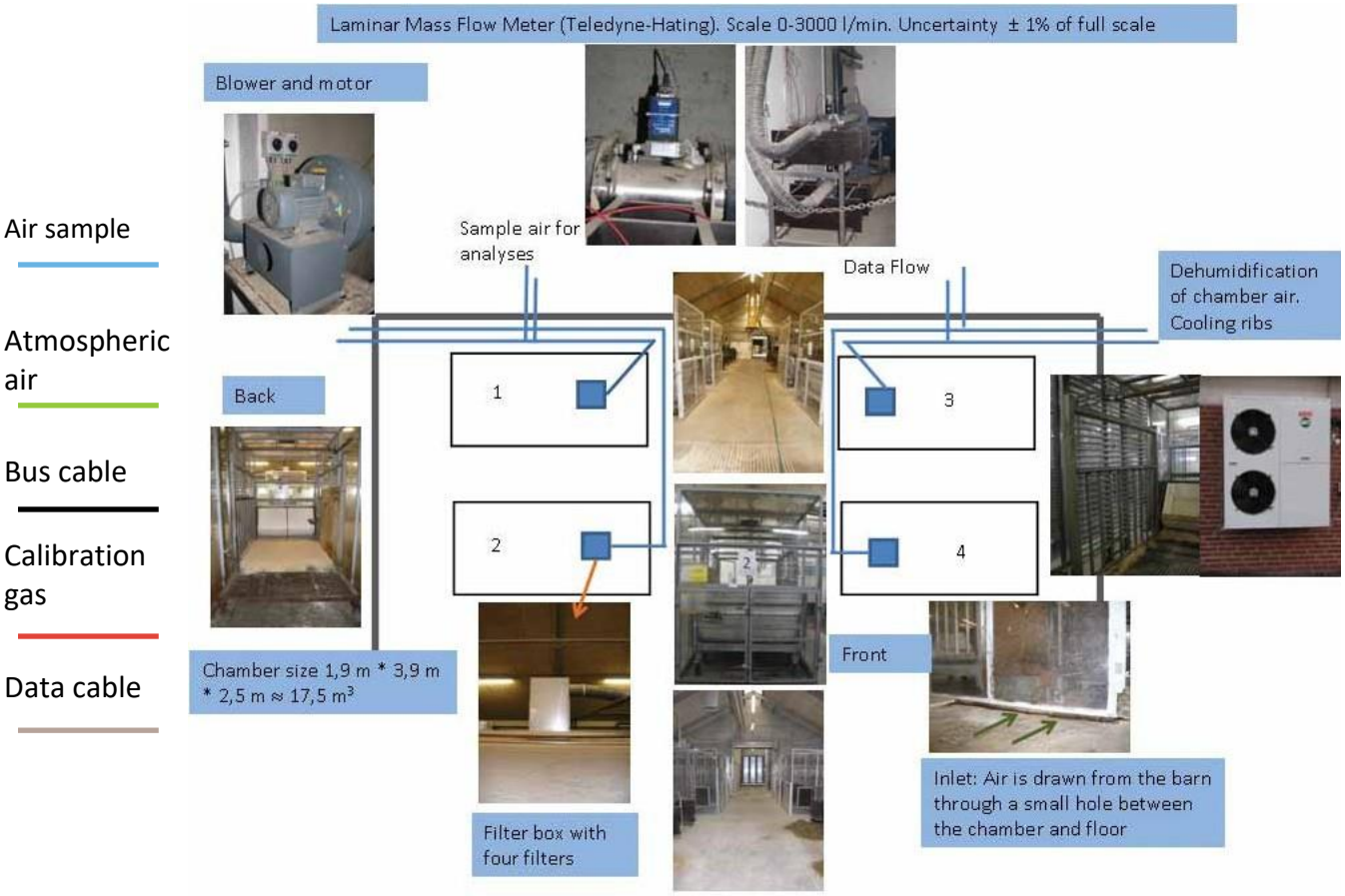


Plate 2: A sketch of K33-1.



The outside dimensions of the chambers are 183 cm (width), 382 cm (length) and 245 cm (height). The chamber volume is approximately 17 m^3.

A 177 cm high double door is placed in the back and a 122 cm double door is placed in the front. Both in the back and front, the doors are closed by a sliding latch and the second door is closed by a lock clamp against the first door (Plate 7) to ensure that the doors are closed. The rear door is for entering during milking, and the front door is for feeding. The edges on the front and rear doors are covered with draught strips.

The chambers are built in two parts which make the transport easier. The metal frame was painted by a professional painter before polycarbonate was mounted. The 6 mm thick polycarbonate is fixed with glue and stopper pins on the steel frame. Polycarbonate and steel have different linear expansion coefficients, and stopper pins are not flexible enough to allow movement of the polycarbonate when the temperature increases. The edges of the two sections were covered with draught strips and screwed together.

The chamber is screwed onto the floor, and the first ⅔ of the side from the front is sealed. The last ⅓ is used as an air inlet. During experiments, the air inlet is sealed with foam rubber (Plate 8) to increase the resistance and thus the air speed (Plates 3 and 8). A smoke test shows that the air movement is slow and that there is no risk that the cow will stand in a draft. It is possible to have a negative pressure when the flow is greater than 1000 L/min. This indicates that the chambers are tight enough and that there are no leakages of air from the chambers to the barn. A smoke test of the chambers shows that the movement of air follows the airstream from the inlet to the outlet.

Each chamber has access to the vacuum and the milk pipe line.

4.4 Animal holding, feeding and cleaning

There is no strict protocol for the operation of the system, and the duration and procedures are planned for each experiment depending on aim, other experiments and labour. Instead, we try to follow a number of guidelines:

- The measurement period should be at least two days and preferably four days. Animals which are measured for only two days will stay in the same chamber. If they are measured for four days they will change chambers (see section 4.10).
- The moving of animals takes place either before or just after the morning or evening milking to reduce the opening time and to ensure the proper estimation of the feed intake.
- The barn workers are instructed to open one chamber and finish work in this before proceeding to the next. The order for fistulated cows is: milking, cleaning, new feed and in the morning also care of fistulas. The total time averages 15–20 min for each cow. The new feed is given just before the chambers are closed. The workers fill in a log book when they open and close the chambers.
- When the chambers contain milking cows, they are usually opened twice a day but only once a day with non-milking animals. On average, we process data for 22 hours/d for milking cows and 23 hours/d for non-milking animals. Milking will normally take place between 05:00 and 07:00 h and between 15:00 and 18:00 h. We always plan an interval of time of at least 10 hours between the milkings.
- All animals are weighed on the way to and from the chambers. The daily milk yield and feed intake are recorded.

Plate 3: Diagram of tubes and cables in the lab.

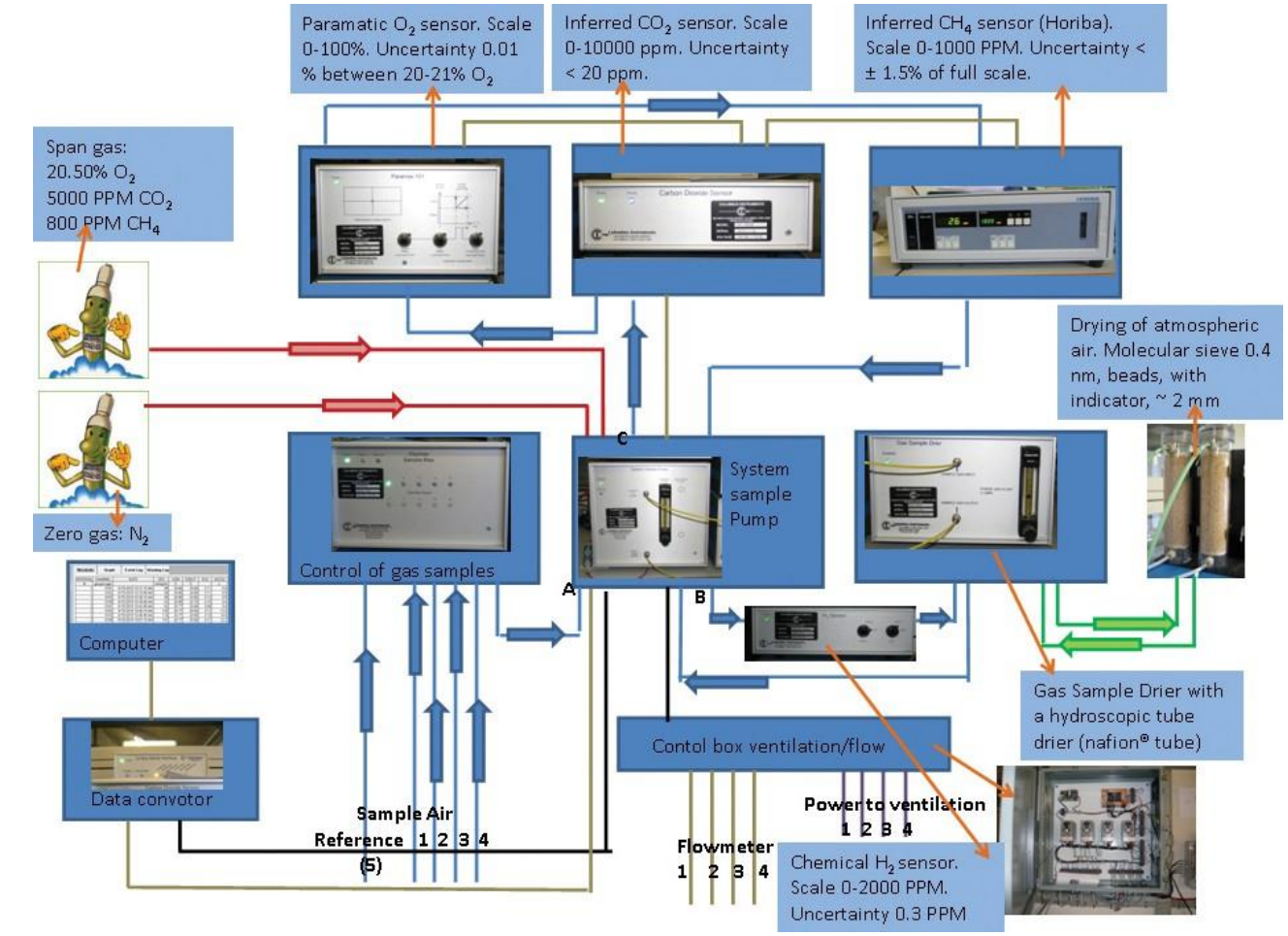
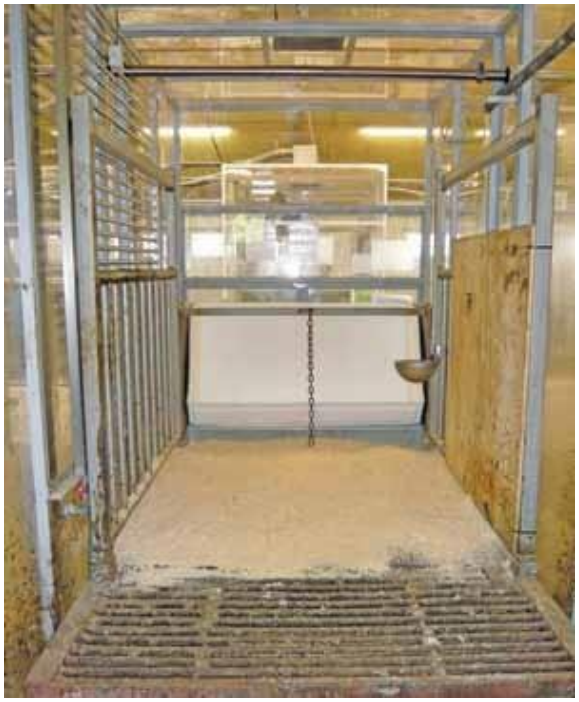


Plate 4: Platform seen from the back.



4.5 Chamber airflow piping and measurement

The flow system consists of:

- A Dayton 2C820 high pressure direct drive radial blade blower with a 0.9 KW motor from VEM motors GmbH (Plate 9).
- The speed of the motors is regulated by an Altivar 31 motor controller (Plate 9) from Schneider Electric by changing the frequency inverters to the motor.

Each motor can be regulated separately. The relationship between the flow and the frequency of the signal to the motor is non-linear. An increase of one hertz will increase the flow more when the flow is increased for instance from 50 hertz to 51 hertz than from 20 to 21 hertz. The flow rate from each chamber depends on the weight of the animal and the production level (milk production or growth) (see Table 1).

Each chamber is fitted with a HFM-200 flow meter, with a laminar flow element from Teledyne-Hating (Plate 9). The flow meters can measure a flow up to 3000 L/min. The laminar flow element has a diameter of 10.2 cm and an accuracy of 1% of full scale, and the repeatability is 0.05% of full scale. The mass flow meter has a shunt and a valve to regulate the air flow.

Table 1: Ventilation rates (L/min) used for animals differing in liveweight and milk production, with expected net concentration of CO_2 of 5000 ppm.

	COW LIVEWEIGHT (KG)						
Milk kg/d	**450**	**500**	**550**	**600**	**650**	**700**	**750**
10.0	492	516	540	564	588	612	636
12.5	534	564	588	612	636	660	684
15.0	582	606	630	654	678	702	726
17.5	624	654	678	702	726	750	774
20.0	672	696	720	744	768	792	816
22.5	714	744	768	792	816	840	864
25.0	762	786	810	834	858	882	906
27.5	804	876	858	882	906	930	954
30.0	852	876	900	924	948	972	996
32.5	894	924	948	972	996	1020	1044
35.0	942	966	990	1014	1038	1062	1086
37.5	984	1014	1038	1062	1086	1110	1134
40.0	1032	1056	1080	1104	1128	1152	1176
42.5	1074	1104	1128	1152	1176	1200	1224
45.0	1122	1146	1170	1194	1218	1242	1266
47.5	1164	1194	1218	1242	1266	1290	1314
50.0	1212	1236	1260	1284	1308	1332	1356
52.5	1254	1284	1308	1332	1356	1380	1404
55.0	1302	1326	1350	1374	1398	1422	1446

Plate 5: Manure wagon.

Plate 6: Feed wagon.

Plate 7: Lock clamp used to ensure that the front and back doors are closed, and a picture of the front door.

Three things have to be fulfilled to get correct measurements from the flow meter:

- The flow meter must be level. This was ensured when the flow meters, blower and motor were mounted.
- The flow must be laminar through the element. The air runs in a PVC tube before and after it enters the flow meter to ensure a laminar flow. The length of the PVC tube before the flow meter is 60 cm and after 120 cm, which is more than the recommend minimum of 50.7 cm (5 times the diameter of the flow meter).
- The air must be dust free. A 31 x 31 cm filter box is placed at the top of the chamber (Plate 10) to ensure a dust free air. The air from the chambers is drawn through four filters (Plate 11). The first filter is a reticulated foam slab. The next three filters are panel filters. The reticulated foam slab can be washed and the filters are checked regularly and are changed if necessary.

The filter box and PVC tube are connected with a flexible hose.

The chamber air is dehumidified to get rid of the water production from the cow. This is necessary in Denmark, where the humidity is high all year round.

An Ascon Internationel® R407C refrigerating plant at 11.28 kW cools down the water which is circulated through ribs in the chamber (Plates 13 and 14). One refrigerating plant serves two chambers. Furthermore, the dehumidified air decreases the temperature in the chamber by 2−4 °C because the heat from the condensation of water on the ribs is removed. The temperature is approximately 1−2 °C above the barn level, and the humidity is approximately the same as in the barn.

Other systems

The temperature, humidity and differential pressure are registered every fifth minute in each chamber during the whole experiment to monitor the climate condition of each cow.

The CO_2 level is monitored in each chamber. If the CO_2 level exceeds 9000 ppm in the chamber or there is a power cut, a GSM modem with an external power supply will phone one of the persons responsible for the experiment (for further details see section 4.9). This CO_2 sensor is independent of the rest of the measuring system.

4.6 Sampling, sample conditioning and analysis

The system can measure five lines. One is used for reference in the barn. We divided the reference line into four tubes in the barn. The tubes are placed the furthest away from the laboratory facilities near the air inlet. The other four lines are used for the chambers. The whole measuring system including the flow meters, the control system for motors and the data acquisition system are delivered by Columbus instruments (Ohio, USA). The measuring system consists of multiport unit switches, a system sample pump, a hydrogen sensor, a sample drier with two drying columns, an oxygen sensor, a carbon dioxide sensor, a methane sensor and a CI bus serial interface. In Plate 3, a schematic outline of the system is presented.

A small sample of air is taken in the middle of the PVC tube between the flow meter and blower (Plate 9). Before entering the measuring system, air is drawn through a Balton DFU® disposable filter. The multiport unit switches the air between barn air (reference) or air from the chambers.

A filter binds the ammonium before it enters the system sample pump. The air is drawn through the system by the system sample pump. The flow in the system is 0.5 L/min.

Plate 8: Air inlet with isolation foam rubber.

Plate 9: Flow system and flowmeter.

Left – Plate 9a: The system. Right – Plate 9b: Flow meter.

Left – Plate 9c: Motor and blower. Right – Plate 9d: Control box with individual motor controller.

Plate 10: Filter box at the top of the chamber.

Plate 11: Filters.

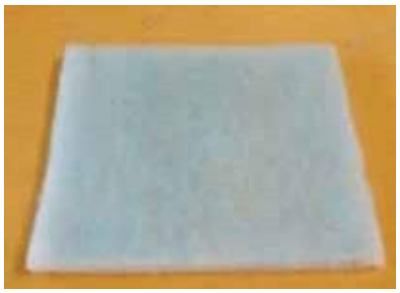


Plate 12: Cooling ribs in the chamber.

After the sample pump, the concentration of H_2 is measured with a chemical sensor.

The next step is the drying of air. The air sample is pumped through an inner tube in the Nafion® tube and dry atmospheric air in the outer tube with a flow of 2 L/min. The atmospheric air is dried in two bottles filled with a molecular sieve (0.4 nm beads, with indicator) that binds water.

After drying, the air is pumped through a paramagnetic O_2 sensor and infrared CO_2 and CH_4 sensors (see Table 2 for details of sensors).

The data from the flow meters and sensors are sent through the CI bus to the computer.

Calibration

Before starting an experiment, all four sensors are calibrated.

The zero point is calibrated with pure nitrogen.

The span point of each sensor is calibrated with calibration gas with a concentration of 20.5% O_2, 0.5% CO_2, 800 ppm CH^4, 150 ppm H_2 in nitrogen as carrier (Table 2). The gas company measures the composition of the gas in the cylinder, and the measured values have a relative uncertainty of 0.2% for O_2 and of 1% for the rest.

Each time the system is calibrated, a log book is filled out to document that the sensors measure correctly. During an experiment, the system is calibrated every second day and always in connection with feeding and milking routines to enable maximum collection of data.

Set up of data acquisition program before the start of an experiment:

- Rinsing of tubes (default 2 min).
- Measuring time (default 30 sec).
- Measurement of the reference (default every fourth time).

The default values of the total measuring time for all chambers and the reference is 12.5 minutes.

The speed of the motors is set to an average CO_2 concentration of 5000 to 6000 ppm in the chambers. This level is chosen because the difference between the outside (barn) and the inside (chamber) has to be as big as possible to reduce the uncertainty of the individual measurements without crossing the upper measuring level of CH_4 and CO_2 sensors in peak periods during the experiment.

Table 2: List of sensors, measuring range, calibration and uncertainty of sensors

Sensor	Principle	Range	Zero/span calibration	Uncertainty
H_2	Chemical	0–2000 ppm	0/150 ppm	0.3 ppm
CO_2	Infrared	0–10,000 ppm	0/5000 ppm	< 20 ppm
O_2	Paramagnetic	0–100%; most accurate between 20–21%	0/20.50 %	0.01 % between 20 and 21%
CH_4	Infrared	0–1000 ppm	0/800 ppm	± 1.5% of full scale

4.7 Gas recovery test

The system is checked regularly, and 35 gas recovery tests have been conducted since establishment in March 2010, until June 2011. The main recovery has been with CO_2 but CH^4 has also been used. The protocol for the gas recovery test is as follows.

Calibration of the system

Adjustment of the gas flow to the chamber with a flow meter to produce an appropriate concentration in the chamber.

The gas cylinders are weighed and placed in the barn, and the tubes are put into the chamber. The CO_2 tube is placed at the top of the chamber. The CH_4 tube is divided into two and placed in the bottom of the chamber.

The measurements are started, and the gas cylinders are opened. To ensure the correct estimation, the decrease in weight of the gas cylinder has to be at least 200 g. The time needed to reach that level depends on the flow from the chambers and the gas concentration in the chamber. Tests with CH_4 take longer than with CO_2.

When the cylinders are closed, the system is given at least one hour to return to barn concentrations and the cylinders are reweighed. The measured accumulations of gases passing through the chambers are compared with measured amounts.

The slope of the decline in gas concentrations after the gas cylinder has been closed, is used to check the chamber volume.

The test results have shown a variation in recovery from 95% to 108% for CO_2 and 90%–110% for CH_4. On average, the recovery has been a little above 100% for both CH_4 and CO_2. (See section 4.10 for further discussions of recovery).

4.8 Emission calculation

The calculation of the methane emission is based on the flow and concentration of the barn air and the outgoing air of the chamber. Until now, no gas recovery correction has been made because the average recovery is approx. 100%. Data deriving from the opening time of the chambers are removed.

CH_4 (L/min) = V_{out} [L/min]* $C_{CH4(out)}$ [ppm]– V_{in} [L/min]* $C_{CH4(barn)}$ [ppm] + delta CH_4 chamber.

Where:

V_{out}: The volume of outgoing air in STP

V_{in}: The volume of ingoing air in STP

$C_{CH4(out)}$: The CH_4 concentration of outgoing air

$C_{CH4(barn)}$: The CH_4 concentration of the reference air/barn air

The Delta CH_4 chamber: The difference of the CH_4 volume in the chamber at the start and at the end of the measurements. The influence on the result will decrease with the increasing duration of the experiment. After 24 hours of measurement, the number is negligible.

The air flow is measured in litres per minute at STP. Only the outgoing air flow is measured. The ingoing air flow can be calculated when the CO_2 and O_2 contents of the outgoing and ingoing air are known. Nitrogen is not used by the animal or excreted by

the animal and will be constant. The ingoing airflow can be calculated as follows:

V_{in} [L/min] = V_{out} [L/min]* $C_{N2(out)}/C_{N2(barn)}$

Where:

V_{in}: The volume of ingoing air

V_{out}: The volume of outgoing air

$C_{N2(out)}$: The concentration of nitrogen in outgoing air

$C_{N2(barn)}$: The concentration of nitrogen in ingoing air/barn air

If the respiratory coefficient is one $V_{in}=V_{out}$. The more the respiratory coefficient differs from one the greater the difference between the volume of ingoing and outgoing air.

The methane production (as well as CO_2 and H_2 production and the O_2 consumption) is calculated for every measuring interval and multiplied with the length of the measuring interval (normally 12 minutes). The calculated production and time of each experiment are summed, and the 24 hour production is calculated.

4.9 Animal welfare and operators' safety

All experiments are approved by the Animal Experiments Inspectorate. The temperature and humidity of the system are automatically registered. If the CO_2 concentration exceeds 9000 ppm or a power cut occurs, the person responsible for the experiment is called. If the blowers stop, the CO_2 concentration will increase at approximately 1 percentage unit per hour, and O_2 will fall at a similar rate. There is sufficient time for the person responsible to get to the barn.

4.10 Weaknesses of the system

There are two main problems in our system. The first is that it is placed in the barn. This causes a fluctuation of the background gas concentrations during the day, so the concentration is not the same for all four chambers. The main reason for placing the system in the barns was to keep the cows in their normal daily environment to ensure that the feed intake was not negatively influenced during experiments. The differences in background gas concentrations are small, and in order to reduce the risk of any influence from background values, the cows change chambers halfway during the four day measuring period. The change is always diagonal so that cows in chamber 1 will be moved to chamber 4. Furthermore, the background problems can be further reduced by increasing the ventilation rate in the barn. This is important during the winter time, when the ventilation rate is low, and the level of methane in the barn can be above 100 ppm. A better mixing of the barn air by a fan will also reduce the problem. The two doors in the barn must be either closed or open to ensure the same concentration all over the barn.

The second problem is the internal mixing of air in the chamber. The only circulation of air in the chamber is generated by the heat from the cow and air change caused by the ventilation system. This is not enough to ensure a total mix of air in the chamber. There are two risks: First, the separation of air in the chamber. Methane is lighter than air and can rise to the top of the chamber, and carbon dioxide can fall to the bottom of the chamber because it is heavier than air. Second, most of the methane and carbon dioxide are expired through the mouth and nose of the cow which will cause a higher concentration in front of the animal. At the rear, the concentration of methane and carbon dioxide is lower because of the air inlet.

The second problem is the most important; there is a little risk of overestimating the methane and carbon dioxide production and differences between treatments will still be correct. The problem could be reduced if a small fan is installed in the chamber for a better mixing of air. These should be placed in such a way that the air from the chamber is not directed out of the chamber through the inlet.

The problems with the background level can explain the fact that the recovery of gasses is not always 100 %. If the background level of CO_2 is ±50 ppm of the measured value it can explain 1–2 percentage points of the difference in the recovery rate. Further, the recovery of gas is dependent on an accurate estimation of the flow. If the uncertainty of the flow meter is 1 % of full scale it would influence the recovery rate with 3 at a flow of 1000 L/min. Therefore, a recovery rate of 90–110 % could be acceptable, but this must always match the flow rate used, the background level in the barn and the chamber concentration of test gas; for instance, if the flow is high, the deviation in recovery must also be low. If the carbon dioxide and methane are run simultaneously, the recovery rate must be nearly the same for the two gasses. If not, it indicates a problem with one of the sensors.

4.11 Description of components and equipment suppliers

Chambers

The chamber frame consists of 40 x 40 mm steel tube covered with 6 mm polycarbonate walls.

The followings parts were delivered by Columbus instruments:

- Four flow meters model HFM-200 with a laminar flow element from Teledyne Hastings instruments that can measure up to 3000 L/min
- Sample controller with valves for control of the gas stream
- System sample pump
- Chemical hydrogen sensor
- Drying system – 2 canisters with drying material
- Infrared CO_2 sensor
- Paramagnetic O_2 sensor
- Infrared Methane sensor VIA 510 from Horiba Instruments INC
- Four Dayton 2C820 high pressure direct drive radial blade blowers
- Four 0.9 KW motors from VEM motors GmbH
- Four inch pvc tubes
- Four inch flexible hose

Other parts

- Cooling plan from Ascon Internationel® R407C at 11.28 Kw
- Stainless steel ribs for cooling water
- Multisensor with 6 channels for CO_2
- Alarm
- Battery for alarm
- Telephone/GSM modem
- Four humidity sensors
- Four temperature sensors
- Four pressure sensors
- Software for locking climate data
- Electrical water counter
- Load cells for feed wagon

4.12 Facility costs

Table 3: Costs of the system, but does not include internal scientific and practical labour costs with respect to planning, establishment and validation of the system.

ITEMS	US$
Four platforms	16,000
Four chambers	40,000
Four air conditioners	32,000
Manure handling	8,000
Feed registration	17,000
Sensors, flow meters, data programme	133,000
Labour (external)	12,000
Test gasses	2,000
Alarm system plus temperature, humidity	20,000
Computer	2,000
Water	2,000
Other expenses	25,000
TOTAL COSTS	**309,000**

Technical Manual on Respiration Chamber Designs

Chapter 5: Sheep Methane Chambers at Aberystwyth University (UK) and CSIC (Spain)

AUTHORS
KJ Hart, DR Yáñez-Ruiz, AI Martín-García and CJ Newbold

Contents

Chapter 5: Sheep Methane Chambers at Aberystwyth University (UK) and CSIC (Spain)

5.1 Summary

Four open circuit respiration chambers have been constructed to quantify sheep methane production. There is no intention to determine energetics. The chambers are of approximately 5 m^3 internal volume with an air flow rate of 2 m^3/min and consist of a polycarbonate (4 mm) shell affixed to a powder coated steel frame (25 × 25 mm box section; 1.80 × 1.80 × 1.53 m) with 90 × 90 mm steel mesh lining the sides. The front of each chamber has 2 doors where there is an air gap of 33 cm not covered by polycarbonate along the width of the front. The chambers are sited on a concrete floor that is covered by a 12 mm rubber mat. Waste is removed by daily scrapping through the door. Chamber airflow is measured twice daily by a hotwire flow meter. A continuous subsample of gas (1 L/min) is drawn from the air exhaust and ambient air supply, through a moisture trap (silica) into a gas analyser (MGA3000; ADC analyzers) with an integral 8 channel multiplexor. Methane concentrations are measured every 60 seconds for 5 minutes from each channel. Gas concentrations and air flow measurements are loaded into an Excel spreadsheet for each chamber allowing methane measurements every 30 minutes. Methane recovery through chambers is measured by injection of a known dose of methane over a 24 h period prior to each experimental run.

A set of four open circuit respiration chambers for sheep and goats have been constructed at CSIC (Spain) based on the Aberystwyth's design, with slight modifications.

5.2 Location of the facility

The physical address of the facility is:

Aberystwyth University
Gogerddan Campus
Aberystwyth
Ceredigion
SY23 3DA
Wales

Consejo Superior Investigacion Cientifica (CSIC)
Camino del Jueves s/n
E-18100 Granada
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The chambers are located in an enclosed barn in Aberystwyth in Wales, United Kingdom at 31 m altitude. This is on the University of Aberystwyth's Gogerddan Campus and is within 100 m of the main sheep housing and handling facilities. The location is situated away from public rights of way. Limited penning of 20 sheep exists within the barn to facilitate moving blocks of sheep in and out of the chambers. If required, animals are transported to and from the facility in a trailer towed by an ATV. The barn is constructed of blocks with slatted wood ventilation and the front of the building has a large roller door to allow vehicular access. The barn is not insulated and not heated and runs at ambient pressure and temperature. A mobile sheep weigh crate is located within the building to weigh the sheep before and after each methane run. Rumen fluid and blood samples can be taken whilst the sheep are manually restrained in their individual pens or within the handling system of the main sheep housing in accordance to the Animals (Scientific Procedures) Act 1986.

The chambers in Spain are located in Granada at the Institute of Animal Nutrition from the Spanish Research Council (CSIC) at 723 m altitude. The facilities include a barn that can hold 55 sheep or goats and an animal trial building housing the chambers. The facility is part of research Institute devoted to Animal Nutrition studies employing around 40 personnel. Within the animal trial building, there are two rooms (65 m^2 each) connected through a corridor with positive air pressure to avoid air cross-contamination between them. One room is used for the adaptation of the animals to the cages and the other where the set of chambers is placed. Both rooms are air-conditioned to maintain the temperature about 25 ^oC (±5 ^oC). An adjacent third room houses instrumentation for sampling, instrument calibration, measuring and data processing.

5.3 Chamber airflow piping and measurement

The four chambers at Aberystwyth are arranged in 2 rows of 2, with the chamber doors opening onto a common aisle that runs between the 2 rows (Plate 1). The chambers are constructed of 25 mm × 25 mm powder coated steel box section modular frames to give length, width and height of approximately 1.80 × 1.80 × 1.53 m and an approximate volume of 5 m^3. The panels consist of a roof, 3 sides and a bisectional front door. To the internal frame a 90 mm × 90 mm steel mesh was welded to protect the exterior polycarbonate sheeting from being damaged. The bottom of the 3 sides had a 330 mm high section of stainless steel sheet welded to them in order to facilitate cleaning. Polycarbonate (4 mm) sheeting is attached to the external frame using foam pads and tech screws in order to completely cover the 3 sides and roof. However, at the bottom of the 2 doors had an air gap of 33 cm left in order to allow air entry to the chamber (Plate 3). The air gap also facilitates as a safety mechanism in case of power failure to the fans. Each panel was bolted together and joints sealed with silicone. The front doors of the chamber are half width and have a baton running from the top to the bottom to allow the doors to be fastened securely once the sheep is inside. Feed and water are manually placed within each chamber in buckets.

Chambers at CSIC are arranged in a single row (Plate 2) and have a single front door that can be dismantled and closed mounting it on two bolts at the bottom and secured by means of two springs in the upper part (Plate 4)

5.4 Animal holding, feeding and cleaning

At Aberystwyth, the animals to be measured for methane production are adapted to diet for at least 21 d in individual pens in the main sheep housing and are fed a diet at 1.05 x maintenance requirements (AFRC, 1993). Sheep are blocked in groups of four according to live

Plate 1: Aberystwyth University sheep respiration chamber facility showing the 4 chambers.

Plate 2: CSIC sheep and goats respiration facility composed of chambers and crates to hold the animals.

Plate 3: Front chambers at Aberystwyth University showing the gap at the bottom of the door to allow air to flow in.

Plate 4: Front of two chambers at CSIC Granada showing the gap at the bottom of the door to allow air to flow in.

Plate 5: Mobile crates at CSIC to hold the animals during adaptation and in the respiration chamber.

weight, age and gender. Animals are bedded on sawdust and have *ad libitum* access to water at all times. Animals are fed in 2 equal portions at 09:00 and 16:00 h daily with refusals being recorded each morning. The feed intake of each block of sheep is averaged for the 7 days preceding entry into the chambers and this amount used to feed each sheep within the block whilst within the chambers. Sheep are moved into the chambers prior to the morning feed and remain resident within the chambers for three 24-h periods. Whilst within the chambers a fine sprinkling of sawdust is placed upon the mats to soak up urine and this is replenished twice a day following manual scrapping of the chamber area. Once the sheep have been removed they are sampled as required and then the mats are scrapped and hosed down and allowed to dry for 24 h prior to the next block of sheep. The chambers are permanently affixed to the floor. Wash water and waste material is directed to a sunken drain covered with a grill that drains away outside the shed.

At CSIC, the sheep are placed in crates (1.0 m wide, 1.5 m long and 1.3 m high) (Plate 5), which are wheeled into the respiration chambers. The crates are made of the same components as the pens in the barn to help animal acclimatisation. Normally, animals start the acclimatisation period to the experimental treatment in the stable and are moved on day 14 to the crates in the acclimatisation room where they stay for at least a week. Then, they are moved into the chambers for gas measurements over three 24-h periods. Keeping the animals in the same crates during adaptation and measuring periods has been proven to prevent animals' distress and potential drop in dry matter intake. Collection trays for feed refusals collection that are disposed at the front of the crate allows for accurate measurement of daily feed intakes.

5.5 Chamber airflow piping and measurement

The respiration chamber air flow is reliant upon negative pressure in the system, achieved by high speed, in line fans placed at the exhaust of each chamber. A common stream of ambient air is drawn from the common aisle between the chambers within the shed through the open section of the front doors. The air is naturally circulated within each chamber and exhausts through a 10 cm diameter outlet hole in the centre of the roof section at the opposite end to the doors. Air passes through a 50 cm length of flexible 10 cm diameter plastic hose into

Plate 6: Top of the chamber at Aberystwyth University showing hand held flow meter inserted into the exhaust pipe.

an inline fan and the air exiting the fan is connected to a 120 cm length of the same flexible pipe which joins onto a 360 cm length of 10 cm diameter rigid pipe containing two 90 degree bends prior to being vented to the outside of the shed through plastic 10 cm diameter tubing. Within the first section of rigid pipe that runs across the front of the chamber is a 2.5 cm hole drilled, approximately half way down the length of pipe, which allows insertion of the hot wire anemometer (Plate 6), when not in use a rubber bung is inserted. A subsample of exhaust gas is continually withdrawn from half way down the sections of straight pipe through a 6 mm diameter plastic pipe. Four similar lines are located within the common aisle to sample the ambient air. A variable speed fan controller (Plate 8) is fixed to the top of each chamber in order to regulate air flow for the readings of exhaust gas to be less than the maximum range of the analyser.

At CSIC Granada, exhausting pipes are made of aluminium and contain three 90 degree bends prior to being vented. Air flow out of the chambers is measured using a 50 cm length tube inserted after the ventilation fan and connected to a flow-meter that contains a dynamic differential-pressure sensor and an electronic measuring and control unit (VAV-Universal VRD2, Belimo Automation AG, Brunnenbachstrasse, Switzerland) (Plate 7).

5.6 Sampling, sample conditioning and analysis

Each of the 8 sample lines (1 L/min) are connected to an individual channel of the gas analyser. Prior to the analyser, each sample line passes through a desiccant (silica) and then a membrane filter to remove any particulates that would affect the analyser. When the solenoid for a chamber (or the ambient line) is open, dried air from that sample line is directed internally to the analyser.

The analyser itself is an ADC MGA3000 (Plate 9) fitted with an infrared detector for methane (50 or 200 ppm) (CSIC: 300 and 1,000 ppm respectively for CH_4 and CO_2). Every 4 h the analyser auto zeros on oxygen free nitrogen. Calibration of the analyser is completed daily using a 50 ppm (Aberystwyth, UK) or 240 ppm (CSIC, Spain) span gas following a manual zeroing both calibration gases being dispensed at 5 psi from the cylinder into the back of the analyser.

5.7 Gas recovery test

Recovery of methane through the chamber is quantified over a 24 h period. A bag containing a known volume and concentration of methane is connected to a multichannel pump dispenser to which is also connected a reservoir of water that is pumped at the same rate into a measuring cylinder. The gas is piped into the chamber through the front air grill and normal measurement cycles are followed. Following the 24 h the recovery of the gas is calculated and if it falls between 98–102% the chambers are ready for use. If the recovery is <98% all components are checked and the test re-run and if recovery is > 102% the analyser is recalibrated and verified against the span gas prior to rerunning the test for a further 24 h.

5.8 Emissions calculation

Average methane concentrations are determined for each chamber and ambient line within an Excel spreadsheet. Air flow is calculated from the 2 mean airflow readings from the anemometer which has been pre-set to calculate air volume from air velocity measurements assuming an internal pipe diameter of 100 mm. The mean methane concentration from each chamber is corrected for mean ambient methane concentration and total methane emissions are calculated based on the total air volume. Methane production is calculated as g/d.

Plate 7: Top of the chamber at CSIC showing the flow meter and electronic measuring unit.

Plate 8: The variable fan speed controller and the inline fan positioned on the top of each chamber, at Aberystwyth University.

Plate 9: Gas analyser used by both Aberystwyth University and CSIC which includes an integral switching unit (lower photo).

Feed intake and DM content are recorded daily to enable emission to be reported as g methane and g methane/kg DM intake.

5.9 Animal welfare and operators' safety

Operator safety

The chambers are designed to allow sheep to be guided into them. Two handlers are always present when moving sheep in and out of the chambers. This is achieved by:

- A race of hurdles is constructed allowing easy movement of the sheep from the pens or loading bay.
- For getting animals into chambers, one side of the chamber door (framed polycarbonate) is opened only allowing the animal into the correct chamber or return the way it has come. The animals are followed by the operators in order to prevent them from returning. The door is sealed manually and locked using a fold-over catch.
- The door on the opposite chamber is then opened and that chamber filled in like manner.

Animal welfare

There is a small risk of asphyxiation which may occur in the event that air flow through the chambers is stopped (power blackout or fan fault). However, this is unlikely due to the air gap which allows air entry into each chamber. The analyser and computer are connected to a UPS which enables the system to carry on logging following resumption of power to the fans. At CSIC air velocity is always set to allow a maximum CO_2 concentration threshold in chambers around 1200 ppm.

5.10 Weaknesses of the system

The system at Aberystwyth was one of the first to be constructed and therefore has some notable problems that when finances allow will be retrofitted. Key improvements anticipated are:

- The air flow is only recorded twice daily, however ,10 readings are taken at each time point from each chamber in order to get an accurate representation of the air flow. Constant monitoring every 30 min of a single chamber, containing a sheep, demonstrated little change in an 8 h day. There is potential of inaccurate readings due to the necessity of the hot wire to be perpendicular to the air flow which cannot be seen once the probe is located within the pipe.
- The number of readings recorded is 10 readings per chamber per hour; this is predetermined by the analyser. This does not allow for flux measurements to be recorded over a short period.
- While functional, the data collection and processing software still requires considerable manual processing. We hope to get this automated quickly and will work with the designer to do so.
- It is anticipated that a cylinder of known methane concentration could be connected to a channel on the analyser. However, a way needs to be designed to allow gas to flow out of a high pressure cylinder at 1 L/min in order that the cylinder does not empty in a short time period.
- At CSIC Granada, signal from dynamic differential-pressure sensor (0 to 10 V) is manually recorded (4 times/day) from the connected potentiometer. There is a good linear correlation within these values and those from an anemometer (m/s) fitted in the centre

of the exhaust pipe. Despite the stability of air flux produced by the fan, automatic continuous record of measurements in the lab computer via the suitable data-logger should increase the accuracy of CH_4 emission measurements.

5.11 Description of components and equipment suppliers

Chamber design and construction:	Local contractor
Air ducting system and fans:	Flakt Woods and local builders merchants
Gas analyser and switching unit:	ADC Gas analysis, UK
Mass flow:	TSI Airflow instruments VAV-Universal VRD2, Switzerland (CSIC)
Data handling software:	Microsoft Excel

5.12 Costing of the facility

ITEMS	US$
LABOUR AND MATERIALS	
Design of the system	In house
Building of chambers	7,500
Piping air circulation and sample lines	700
Mobile animal crates	6,600 (CSIC only)
EQUIPMENT	
Air flow meter	1,800[1]
Gas analyser[2], inc switching unit	11,500 (CH_4)
Calibration gases	600
Analyzer control software	2,990
Computer and data acquisition system	1,000
TOTAL COST	26,090 (UK) or 32,690 (CSIC)

[1] Cost for a single hand held meter (UK) or 4 inline meters (Spain).

[2] Analyzer can handle up to 4 gases at additional cost.

Technical Manualon Respiration Chamber Designs

Chapter 6: Metabolic Centre of the University of Zurich and ETH Zurich (under construction)

AUTHORS
Kathrin Buehler and Marcel Wanner

Contents

Chapter 6: Metabolic Centre of the University of Zurich and ETH Zurich (under construction)

6.1 Summary

At the time this manual was written (June 2011), the metabolic centre of the University of Zurich and the Swiss Federal Institute of Technology Zurich is still under construction. In its first stage it will be situated at a temporary location and it will consist of two large chambers for cattle, two medium chambers for sheep and goats and two small chambers for smaller animals (piglets etc; not discussed in detail in this manual). It is planned to enlarge the metabolic centre to four chambers per size. Facility air flow is about 910 m^3/h and temperature ranges from 20 °C to 26 °C without humidity control. The chamber casings are constructed with insulated sandwich panels. Windows are inserted into the casing on each side of the chamber. Chamber volume of the large and medium chambers is about 22.4 m^3 and 9.2 m^3, respectively. The chambers will be operated at a slight negative pressure (maximal 100 Pa below ambient pressure). Air flow through the chambers will be between 300–1000 L/min (large chamber) and 40–250 L/min (medium chamber). Each of the chambers forms an independent system with its own flow generator, air conditioning unit, animal welfare monitoring and data logging. The two large chambers share a common gas analysis and data acquisition system, so do the two medium chambers. From the flow generators a sub sample is directed to the analyser system by the means of a multiplexer at regular intervals. The duration of these intervals is not yet known. Concentration of water vapour, CO_2, CH_4 and O_2 are measured with a four-gas analyser. Intervals of gas analyser calibration and recovery tests are not yet defined and will be set according to the dynamics of the chamber and the gas analysers. The large and medium chambers are planned to suit the needs of research animals and operators while enabling monitoring of CH_4 emissions for a maximal time during measurement.

6.2 Location of the facility

The physical address of the facility is:

Strickhof
Eschikon
Postfach
8315 Lindau
Switzerland

Mailing address:

University of Zurich, Vetsuisse-Faculty
Institute of Animal Nutrition
Winterthurerstr. 260
8057 Zurich
Switzerland

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The metabolic centre is temporarily situated in a part of the agricultural outbuilding of the Strickhof Lindau (Plate 1). This is a centre of excellence in agricultural education, services as well as information and belongs to the Department of Landscape and Nature of the Canton of Zurich. The metabolic centre itself is led and financed by the University of Zurich in close collaboration with the Swiss Federal Institute of Technology (ETH Zurich). For research conducted in the metabolic centre, the animals and stables of the Strickhof Lindau can be used.

Strickhof Lindau is situated about 20 km from the University of Zurich and ETH Zurich (35,000 students in total) and about 10 km from the city of Winterthur (100,000 inhabitants). A highway (A1) is approximately 2 km from the location but there is no industry nearby.

The part of the outbuilding where the metabolic centre is located was separated from the rest of the building by a plywood wall. The floor, walls and ceiling were insulated and floor heating was installed. Heating and cooling maintain a temperature between 20 °C (winter) and 26 °C (summer) within ± 2 °C without control of humidity. The pressure in the facility is equal to the ambient pressure. Air circulation in the metabolic centre is maintained with a flow rate of 910 m^3/h by the means of ceiling vents. Ventilation is only turned on when the chambers are in use or when people are present.

All chambers are located in the same room. A separate room contains the equipment for gas analysis, calibration, chamber control as well as data acquisition and processing. Both rooms are air conditioned and ventilated by the facility's system. A dedicated room on the second floor of the outbuilding contains the equipment for the centralised cold and heat production. Feed preparation, stables and cleaning facilities are within 300 m of the metabolic centre on the premises of Strickhof Lindau.

Each respiration chamber is an independent unit with its own flow generator, air conditioning unit (connected to the centralised cold and heat production), data sampling and animal welfare monitoring. The two large chambers are grouped together and share one gas analyser chain and one data acquisition and analysis system (Plate 2). The same design is used for the medium chambers.

6.3 Description of the chambers structure

The respiration chambers were designed to be suitable for a wide range of animal species while guaranteeing accurate measurements of whole animal metabolism and especially of CH_4 production. In addition, the chambers should be as safe and as comfortable as possible for animals and operators.

The design of the chambers is a mixture of existing large and medium chambers in Switzerland, Germany, The Netherlands and New Zealand. It was also taken into account that the new respiration chambers should be suitable for a wide range of research questions. Furthermore, the chambers were built to not just accommodate cattle, sheep and goats but also other animals such as ponies (large chamber) or pigs and camelids (medium chamber). The chamber design also allows easy addition of further measurement devices within the chamber (for example, activity detector) or to the analyser chain (for example, H_2, NH_3). The metabolic centre also includes two small chambers for piglets, cats and rabbits but these chambers will not be discussed further in this manual.

LARGE CHAMBER (Plate 3)*:* The outside dimensions (cm) are 550 long, 250 wide and 310 high. These numbers include the air conditioning unit, the air lock and the 'chamber core'. The inner dimensions (cm) of the 'chamber core' are 354 long, 230 wide and 275 high (V = 22.4 m^3) with a suspended ceiling 42 cm below the chamber casing. The 'chamber core' consists

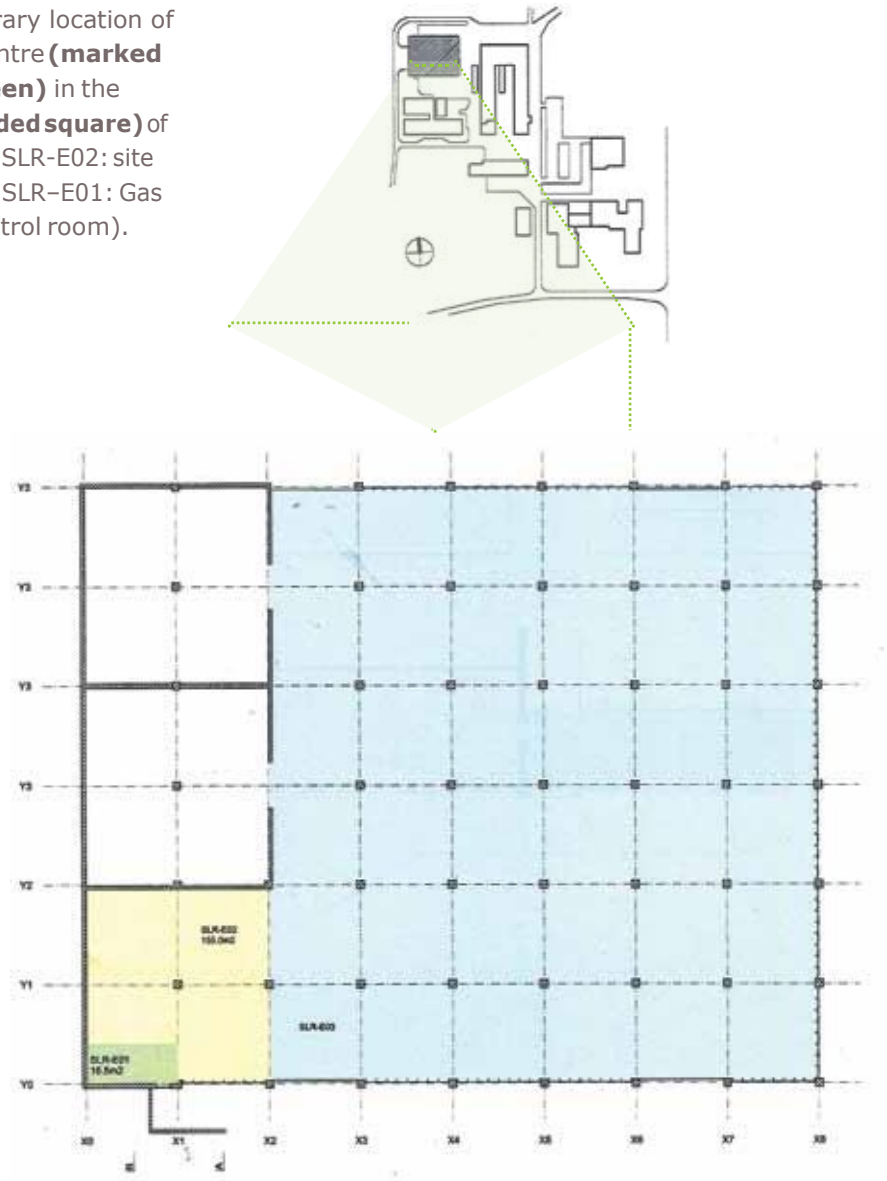


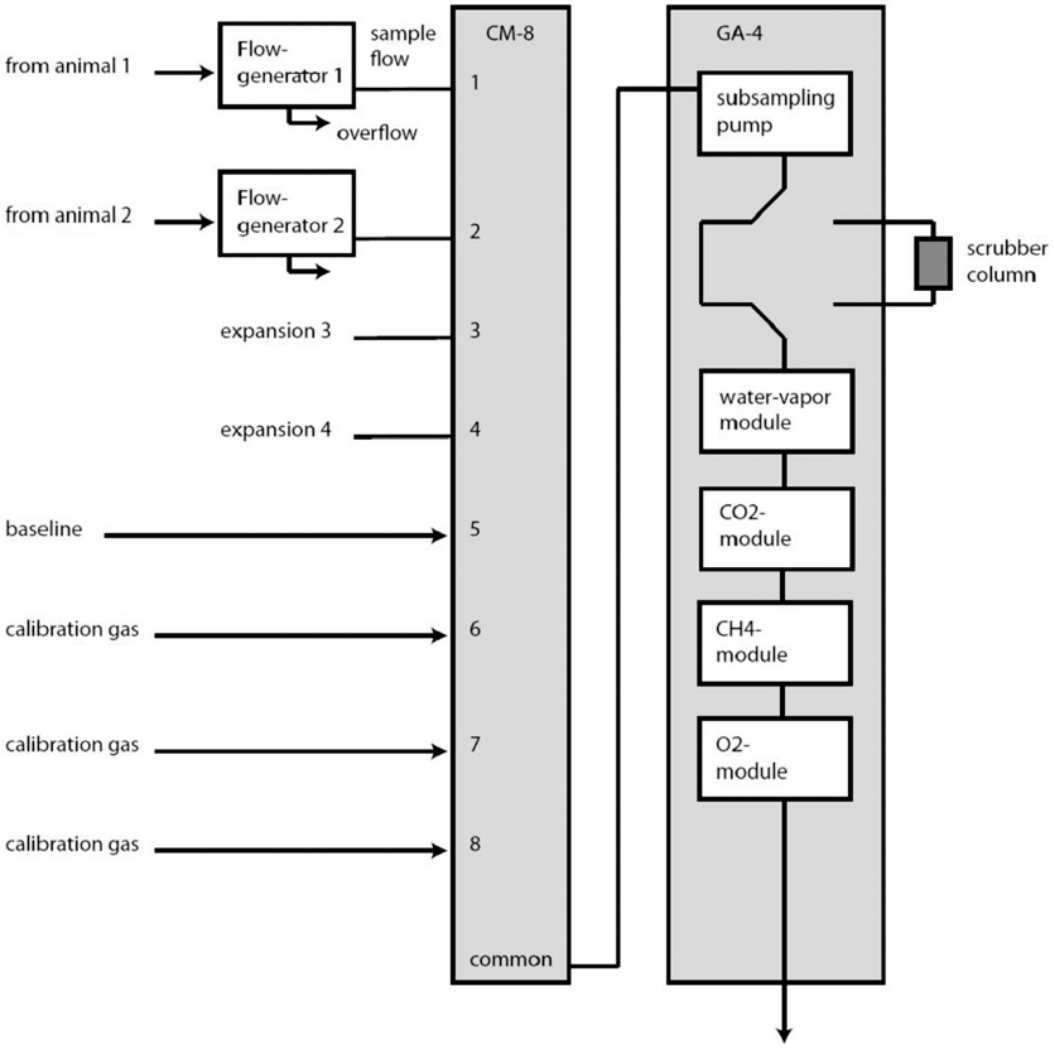
Plate 1: Temporary location of the metabolic centre **(marked yellow and green)** in the outbuilding **(shaded square)** of Strickhof Lindau (SLR-E02: site of the chambers; SLR–E01: Gas analysis and control room).

of the animal area (240 cm length, 130 cm width, 200 cm height) and a maintenance aisle for easy access to the animal.

The chamber has a double rear door, closing onto the chamber casing. This door is used to get the animal into and out of the chamber. Rubber seals on both doors and the casing ensure air-tightness. For feeding, milking (if necessary) and exchange of the excreta trays, the chamber can be accessed through the airlock on the far side. The airlock is connected to the chamber by a sliding door sealed with rubber seals. All doors are operated manually. Cables leave (or enter) the chamber through a special airtight sheet for cables, and a tube ensures that additional cables can be brought into the chamber without compromising its tightness in the future.

Temperature (0–40 °C, ± 0.5 °C) and relative humidity (20–100 %, ± 3%;) are controlled for each chamber separately by an air conditioning unit (monoblock) placed on front of the air lock, and connected to the centralised air conditioning units. Humidity is not regulated

Plate 2: The diagram shows the general set up used for a group of chambers. Only air/gas flows are shown. Data from the GA-4 and data from sensors in the chambers are sent to a computer and are recorded by the data acquisition system. The multiplexer (CM-8) is also controlled by the computer and follows a macro defined by the operator. This set up is repeated for each chamber size. Expansion 3 and 4: shows where the planned additional two chambers will be connected to the multiplexer and the analyser in the future.



below 10 °C. The air conditioning unit is completely integrated into the chamber casing to prevent leakages out of the chamber. Measures to reduce noise and vibration due to the air conditioning unit are in place and will be used if necessary to minimise animal stress associated with noise.

The chamber walls are made of insulated sandwich panels. The inside consists of stainless steel (EN-Standard 1.4301) and the outside is made of PVDF coil coated galvanised sheet, painted white (RAL9010). Insulation is achieved with 100 mm of polyurethane foam and the panels are glued together (inside and outside) with silicone adhesive. Triple pane windows are inserted into the casing (float glass, Ug-value 0.7, ED 36 mm). The window frames are electrically heated (dew point controlled) to prevent condensation. The whole chamber is protected against thermal bridges with key and slot joints.

The chamber has a small window in the rear door and a medium sized front window (75 cm wide, 89 cm high). Two large windows (104 cm wide, 153 cm high) on either side of the chamber allow the animal to see its surroundings. Two of these windows are fixed whereas the other two are held shut by an electromagnet and serve as emergency openings. In case of a power failure or violation of set thresholds in temperature, humidity, CO_2, CH_4 or NH_3 the windows are pushed open by gas rams. One window opens from above, the other from below to allow good circulation of fresh air and to avoid accumulation of toxic gases on the bottom of the chamber.

MEDIUM CHAMBER (Plate 3)*:* The outer dimensions of the medium chamber are 400 cm long, 180 cm wide, 310 cm high. The 'chamber core' (inner dimension) is 250 cm long, 160 cm wide and 230 cm high (V = 9.2 m^2) and has a suspended ceiling 22 cm below the chamber ceiling. The area where the animal is kept measures 170 cm length, 90 cm width, 200 cm height. As with the large chamber, the remainder of the space in the 'chamber core' is used for accessing the animal.

Apart from some minor modifications, the medium chamber is built the same way as the large chamber. The main differences are:

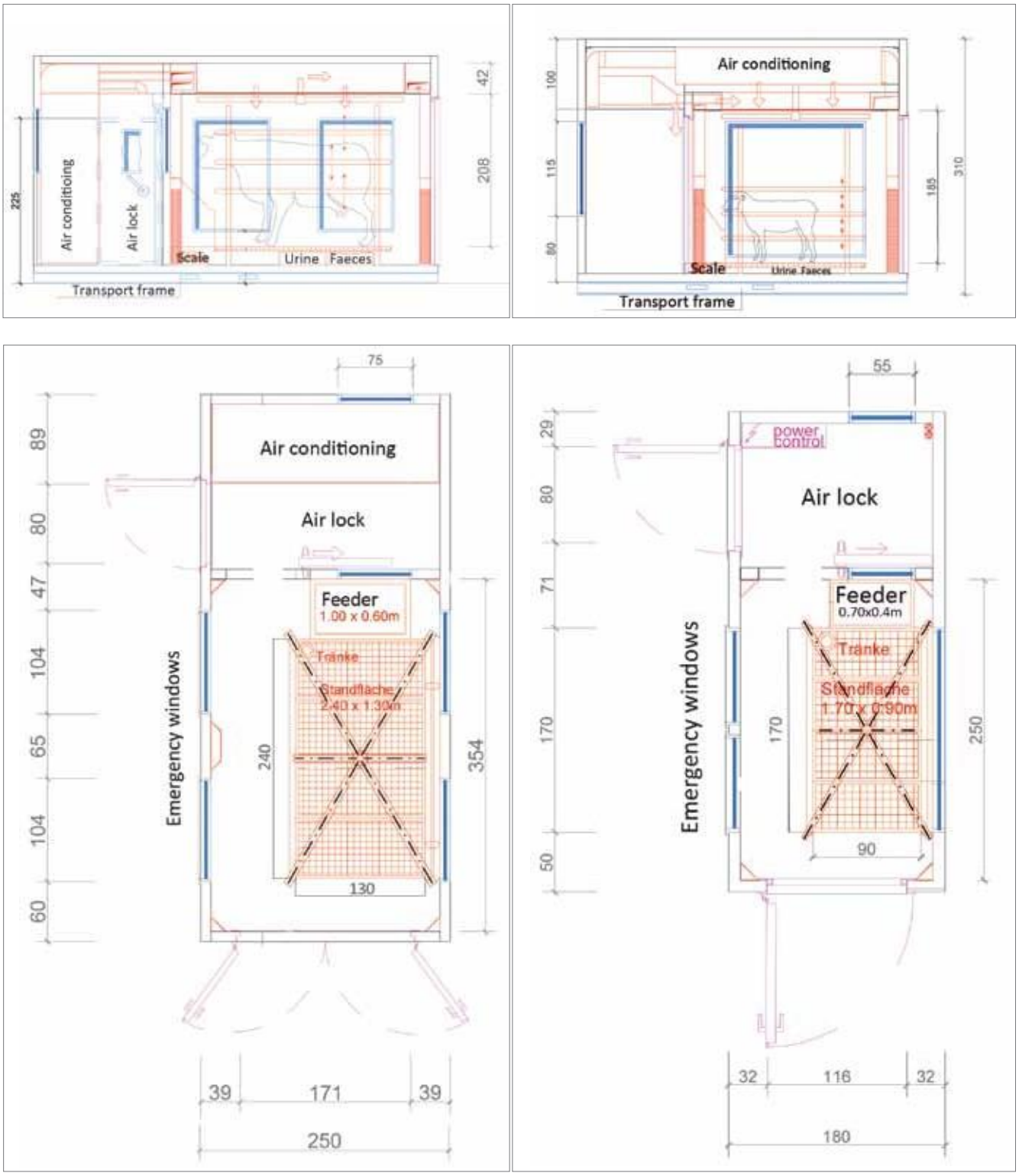
- There is a single rear door, and this entrance is used to get the animal into and out of the chamber.
- The air conditioning unit is on top of the chamber and not in front of the air lock.
- The dimensions of the front window of the medium chamber are 55 cm wide and 115 cm high. Two large windows (170 cm width, 168 cm high) are placed on each side of the chamber. On one side, the window is divided in two halves and serves as emergency opening (same opening mechanism and alerts as in the large chamber).

The large and medium chambers are welded onto a transport frame (stainless steel) with openings on the long side to allow transport with a forklift. The chambers themselves are too heavy (several tons) to be toppled by a rampaging animal. Nevertheless, for additional safety square plates were glued to the room floor to prevent movement.

6.4 Animal holding, feeding and cleaning

The animal containment area consists of a highly adaptable stanchion welded to an elevated floor and includes the feeder and water bowl (Plate 4). All parts of the stanchion are made of stainless steel with the exception of the automatic water bowls (enameled) for easy cleaning and resistance to corrosion. At the top of the stanchion metal bars connect the two sides of the stanchion to dissipate any powerful movements of the animal. It was ensured that the adaptation of the stanchion to the animal's size can be easily performed. The animal

Plate 3: Views and dimensions of the large chamber (left hand side, this page) and medium chambers (right hand side, this page). Windows are marked blue.



containment does not cover the complete 'chamber core' and a maintenance aisle allows easy access to the animal for feeding, milking, removing of faecal and urine trays or for minor veterinary treatments.

LARGE CHAMBER: A ramp (stainless steel) with an adjustable gradient is wheeled to the chamber and used to lead the cattle onto the elevated floor of the stanchion (29 cm above chamber floor). Once the animal stands in the stanchion, a chain attached to a collar is used to prevent the animal from turning around, but does not restrain head movement or lying down.

The floor is divided into several segments covered with a rubber mat (2 cm thick). The floor segments are between 37.5 and 39.0 cm wide. Depending on the size of the animal, single floor segments can be removed (or added), adjusting the floor to the size of the animal. This system promises easy collection of faeces and urine (female animals) independent of the animal's size. When measuring from male animals, the rubber mat is removed at the likely place of urination and urine is collected by means of a moveable funnel below the elevated floor, which leads to the urinary tray. To prevent the animal from stepping back on the chamber floor a vertical barrier is put in place behind the animal.

To narrow the area of defecation, the width of the stanchion can also be adjusted. A movable side wall can reduce the width of the stanchion, in increments of 6.6 and 8.0 cm. The maximal width of the stanchion can thus be reduced from 130 cm down to 92 cm.

Feeding and milking (bucket milking system) will be twice a day. The feeder is leveled with the stanchion's floor and it is designed to contain about 22 kg of dry matter. The weight of the feed and weigh changes are recorded by the means of load cells below the feeder. Water is provided and consumption is measured by a water meter. Light beams measure total time of lying or standing.

MEDIUM CHAMBER: Sheep and goats can either be measured when they are tied with a neck chain (similar to large chamber) or in a metabolic crate. If the animal is tied, the procedure is the same as for cattle: The animal is led up the ramp, tied to the stanchion and the stanchion is then adjusted to the animal's size. The principal construction of the stanchion in the medium chamber is the same as for the large chamber. The floor segments of the medium chamber measure between 25 and 36 cm. The side wall to reduce stanchion width can take one of five positions, each between 6.6 and 8 cm from the other. Thus the width can be reduced from 90 cm to 58 cm. However, primary tests in the factory showed that the minimal width actually used in the medium chamber will most probably be 66 cm, as otherwise the feeder cannot be accessed by the animal.

Feeding and milking (by hand) will be twice a day. Measurement of food and water intake and of standing and lying times are the same as in the large chamber. The feeder of the medium chamber can contain up to 5 kg of feed.

In case a metabolic crate is used, the whole floor of the stanchion can be removed and the crate can easily be wheeled in. The existing crates (Plate 5) will soon be modified to improve animal welfare, without disturbing the air circulation or animal handling.

On average, the animals are kept in the metabolic chamber (large and medium) for two days. Maximal duration of metabolic measurements in the respiration chamber will be four days. The large windows in each chamber allow the animals to see their surroundings, and each other to minimise stress. The windows also ensure daylight in the chamber. If necessary, lamps in the chamber (timer controlled) can be used to increase light intensity. An inbuilt radio in the chambers muffles other noises and keeps the animals calmer in case of sudden sounds.

Exact procedures and the time budget for exchanging animals and cleaning the chamber and metabolic crates are not yet known.

Plate 4: Stanchion for the large **(left)** and medium **(right)** chamber. It is adjustable in width and length to the size of the animal (in both pictures the last segment of the floor has been removed). The floor will finally be covered with a segmented rubber mat.

Plate 5: Existing metabolic crate for sheep. The feeding and water trough can be seen in the front. Urine and faeces are collected separately beneath the crate.

6.5 Chamber airflow piping and measurement

LARGE CHAMBER: Air flow through the large chamber can be set between 300 and 1000 L/min (rule of thumb: increase of CO_2 should not exceed 1 percentage unit and measurement of O_2 consumption should still be possible). At the metabolic centre, the facility's air ventilation system (incoming fresh air) serves as inlet source. Each chamber has its own inlet source. Fresh air from the ventilation system is ducted over a draught damper from the facility air conditioning and ventilation to the chamber is above the suspended ceiling into the chamber. The suspended ceiling is punctured with 8 mm holes, to avoid laminar flow and to reduce air speed. The aim is an air speed of maximal 0.2 m/min in the animal compartment to avoid draught. The air within the chamber is mixed with help of the air conditioning unit at a rate of 80 times per hour. For recirculation, the air leaves the chamber through five openings (20×70×6 cm) at the bottom of the chamber (one in each corner of the chamber and one in the middle of the long side of the chamber). The air is then recycled into the chamber the same way as the fresh air.

Because of the thorough mixing of the air within the chamber, the outlet (air going to the analysers) could be described as a 'spider' hanging just below the suspended ceiling (Plate 3). This 'spider' consists of 6 PVC tubes (inner diameter of 5.5−6.5 cm and capped ends) fixed to a central collector box. Four of these tubes measure 140 cm in length and two 60 cm in length. Air drawn out of the chamber enters through 5 cm holes drilled at 50 cm intervals and is connected to a 12 m long PVC tube with a diameter of 9 cm which takes all of the air (untreated, wet) to a filter (5 µm) and flow meter (one per chamber, capacity can be set between 300–1000 L/min). The flow meter consists of a sealed rotary pump with a controllable flow rate and a mass flow meter that provides flow measurements corrected for standard temperature and pressure. This keeps the chosen and set flow rates constant throughout the measurement period.

Sensors for monitoring temperature and relative humidity (for surveillance only, and not used for analysis) are located in the middle of the chamber, attached to the stanchion. Surveillance analysers to determine gas concentrations are placed in the recirculation air flow tubes

MEDIUM CHAMBER: Air flow through the medium chamber can be set between 40 and 250 L/min. Location of the air inlet, the function of the suspended ceiling, and the location of the air outlet are the same as in the large chamber. As in the large chamber, mixing of chamber air is by re-circulating the air 80 times per hour via the air conditioning unit through 4 openings (15×60×4 cm) at the bottom corner of each chamber.

The dimensions of the 'spider' (PVC-C tubes) in the medium chamber are: four arms with 100 cm length and two arms with 35 mm length. The inner diameter of the tubes is 5.5−6.5 cm. The diameter and distance of the holes as well as the working principle are the same as in the large chamber. From the medium chamber the air is drawn through a 15 m PVC tube with a diameter of 5 cm to the flow meter (one per chamber, capacity 40–250 L/min; same working principle as in the large chamber). From there the air is processed the same way as in the large chamber.

6.6 Sampling, sample conditioning and analysis

The flow meters are continuously drawing air out of the chambers at 300–1000 L/min (large chamber) and 40–250 L/min (medium chamber). This 'pull-system', together with valves at the inlet of the chamber, always ensures a closely controlled slight negative pressure in the chamber, thus preventing leakages out of the chamber.

An eight-channel multiplexer controls four chamber samples (two not used at the moment),

with three calibration gases (one not used at the moment), one baseline sample, and the flow meter either exhausts all air or sub-samples air leaving the chamber for analysis (Plate 2). This gas is then delivered by a diaphragm sub-sampling pump to the analysers, and is exhausted after passing through the O_2 analyser. All tubing from the flow generators and other gas streams to the multiplexer and to the gas analyser utilise Bev-A-Line tubes.

The multiplexer switches the channels at preset time intervals, defined by the operator. However, only one gas stream (from one of the chambers *or* baseline *or* calibration gas) is sent to the analyser at any time.

The analysers are equipped with in built 3 µm filters, after which there is no further treatment of gases (from chambers or baseline) to the analyser. However, for calibration some of the gas samples are dried and scrubbed from CO_2 (see below), so attention has to be paid to the possibility of condensation, which may harm the analysers. The analysers are heated to avoid condensation over a wide range of temperature and humidity, but under extreme conditions insulation, scrubbers (chemical or physical) or condensate traps may have to be installed.

Gas concentrations are measured serially in a "four gas" analyser. The analysers also have an internal temperature control and barometric pressure compensation, so the readout is already corrected to wet standard conditions (0 °C, 1,013 hPa). The detection ranges for CO_2, CH_4 and O_2 are: 0–10 % (accuracy 1 % of reading, resolution 0.0001 %), 0–10 % (accuracy 1 % of reading, resolution 0.001 %) and 0–100 %, accuracy 0.1 %, resolution 0.001 %), respectively. The fourth gas being analysed is water vapour (range: 0–100 % RH, accuracy: 1 %, resolution: 0.001 % RH, 0.1 dew point) and the water concentration in the air is used for "mathematically drying" the flow rate and the other gas concentrations to remove effects of water vapour. CO_2 and CH_4 are measured using infrared technology and O_2 is measured with a fuel cell. For measuring H_2O thin-film capacitive technology is used.

Calibration of the gas analysers (zero and span) will be made before the beginning of each measurement. During measurement the analyser regularly recalibrates automatically. A zero value of the H_2O analyser is achieved with dry air. The equation for spanning is:

$$\text{Water vapour pressure} = BP \times (F_iO_2(dry) - F_iO_2(wet))/ F_iO_2(dry)$$

For zeroing the CO_2, CH_4 and O_2 analyser, nitrogen (99.999 %) is used, and CO_2 and CH_4 are spanned by using a mixed gas (0.5 % CO_2, 0.1 % CH_4 in N_2 as carrier) Production accuracy: is ± 5% and analytical accuracy: ± 2 % . The O_2 analyser is spanned with dry, CO_2-free ambient air as it shows an almost constant concentration of 20.95 %. The air is scrubbed from water and CO_2 by using magnesium perchlorate and ascarite.

6.7 Gas recovery test

Gas recovery tests are planned to be done with propane burns but the method for methane recovery has not yet been determined. The propane torch is put on a portable precision scale (weight range: 4000 g; resolution 0.1 g) and left there burning with a small flame (duration not yet known) and the scale is connected to a notebook where the weights and exact times are recorded (probably between 1–5 seconds) in an excel file. Calculations will only use data where linear weight loss can be observed (regular, undisturbed burning). Calculation of gas recovery is then done by comparing the expected amount of O_2 consumption and CO_2 production to the values given by the analysers. The expected amount and the recovery rate are calculated using the equations:

$$C_3H_8 + 5\ O_2 \text{ à } 3\ CO_2 + 4\ H_2O$$

and:

Recovery rate (%) = 100 x $V(Gas)_{measured}/V(Gas)_{calculated}$

The frequency of gas recovery tests will depend on the dynamics of the chambers and the analysers. Until the chamber is properly working, frequency will be higher (possibly several times per day) to ensure that leaks can be detected and repaired. The first tests with animals will not start before good recovery rates are achieved. After a measurement routine has been established, test frequency will be 1–3 monthly. Additional gas recovery tests will be performed after alterations to chambers, piping, pumps or analysers, or malfunctions.

6.8 Emissions calculation

For the calculation of CH_4 emission, measurements of outflow rate (FR_e) and factorial gas concentrations in the air entering (baseline, F_iGas) and leaving (F_eGas) the chambers are needed. The flow rate is measured as mass but the output is given as L/min, corrected to 0° C and 1,013 hPa. The gas analysers are equipped with internal temperature and pressure sensors and they automatically record gas concentrations at standard temperature and pressure (0° C, 1013 hPa).

To correct for the effect of water vapour on FR and gas concentrations, barometric pressure (BP) and water vapour pressure (WVP) are also recorded. Calculations use the 'mathematically' dried values:

Dry flow rate (l/min): FR (dry) = FR x (BP – WVP)/BP

Dry gas (%): Gas (dry) = Gas x BP/(BP – WVP)

Rate of CH_4 production (l/min):

$$V_{dot}CH_4 = FR_e \times [(F_eCH_4 - F_iCH_4) + F_iCH_4 \times (F_iO_2 - F_eO_2) + F_iCH_4 \times (F_eCO_2 - F_iCO_2)]/(1 + F_iCH_4)$$

In the above equation F_i and F_e are the inflow and outflow fractional concentrations of the specified gas. F_i equals the values of baseline (ambient air) concentrations.

For each chamber flow rates, gas concentrations, temperature, humidity and barometric pressure are available and recorded during the measurement period. The data acquisition and analysis software visualises the recorded data in real time on the computer screen. To correct for slow washout times, an instantaneous correction (also called Z-Transformation) is applied to the raw data. This transformation results in very low response times and almost instantaneous values despite the large volume of the chambers.

A measurement cycle will normally show the following pattern: chamber one; chamber two; baseline; chamber one; chamber two; baseline…etc. The optimal duration of measurements in one chamber and of the total measurement cycle is not yet known. Most probably one chamber will be measured for 60 to 90 s and a cycle will be completed after 3 to 5 min. As soon as these values are known, instantaneous CH_4 emissions can be extrapolated to 24 h.

In the large and medium chambers, data recording and storing continues when the chamber is opened, so every time a door is opening is recorded. To keep disturbances of the chamber environment as small as possible, operators are asked to use the air lock as entrance and to keep the duration of their stay in the chamber to a minimum. An additional indication can be entered manually prior to and after entering the chamber which may also help (and simplify) analyses recorded during feeding, milking or similar disturbances.

6.9 Animal welfare and operators' safety

Animal welfare and animal safety strongly influenced the design of the respiration chambers. All possible measures were taken during planning to avoid injuries or fatalities related to chamber malfunctions or gas accumulation. Great care was taken to ensure that the animal will be as comfortable as possible during its stay in the chamber. During measurements the animal can regularly be checked without disturbance by means of video surveillance (infra-red at night). A control monitor also gives information on the status of the chamber and the building (Plate 6).

Supply of fresh air and gas concentrations in the chamber are closely monitored to avoid suffocation of the animal. For safety reasons concentrations of CO_2 and CH_4 are monitored in duplicate. NH_3 is monitored to ensure explosive mixtures of these gases do not eventuate.

The emergency windows are held in place by strong electromagnets. If an alarm is triggered these release and the gas rams are able to overcome pressure difference between the inside and the outside of the chamber and open the windows. The emergency windows open at the following events:

- Power failure
- Temperature exceeds set threshold
- Humidity exceeds set threshold
- Air pressure exceeds set threshold
- Air flow exceeds set threshold (measured within the chamber (air conditioning unit) and by the data acquisition system. Either of them can trigger the opening of the emergency windows)
- Failure of ventilation of the facility (supply of fresh air)
- CO_2, CH_4, NH_3 (CO_2, and CH_4 measured within the chamber and by the data acquisition system) can trigger the opening of the emergency windows.

Events other than emergency openings that will trigger an alarm, and the system for alerting personnel (SMS-Message, cellular telephone) has not been determined but will be standardised and tested by the start of the first measurements.

No special measures for operator safety were taken but the following points will help to reduce the risk of accidents and improve easy handling:

- Good light in the chamber.
- Clear markings of steps to prevent stumbling.
- Strong stanchion with a frame to transfer forces; neither the stanchion nor chamber can be moved in case an animal behaves badly.
- Normal operation of the chamber can be done by one person.

During the first test of the new respiration chambers the main focus will be on animal welfare and safety as well as on handling and operator safety. Necessary adaptations will be made.

6.10 Weaknesses of the system

The main weakness of the system, that is, the metabolic centre itself, is its location in the outbuilding. The design of the chambers had to be made according to space limitations and all chambers had to be put in one room, and separation of the large chambers reduced visual contact between the animals. Restrictions by the Commission for Animal Welfare do not allow use of the large and medium chambers simultaneously. However, this situation will improve

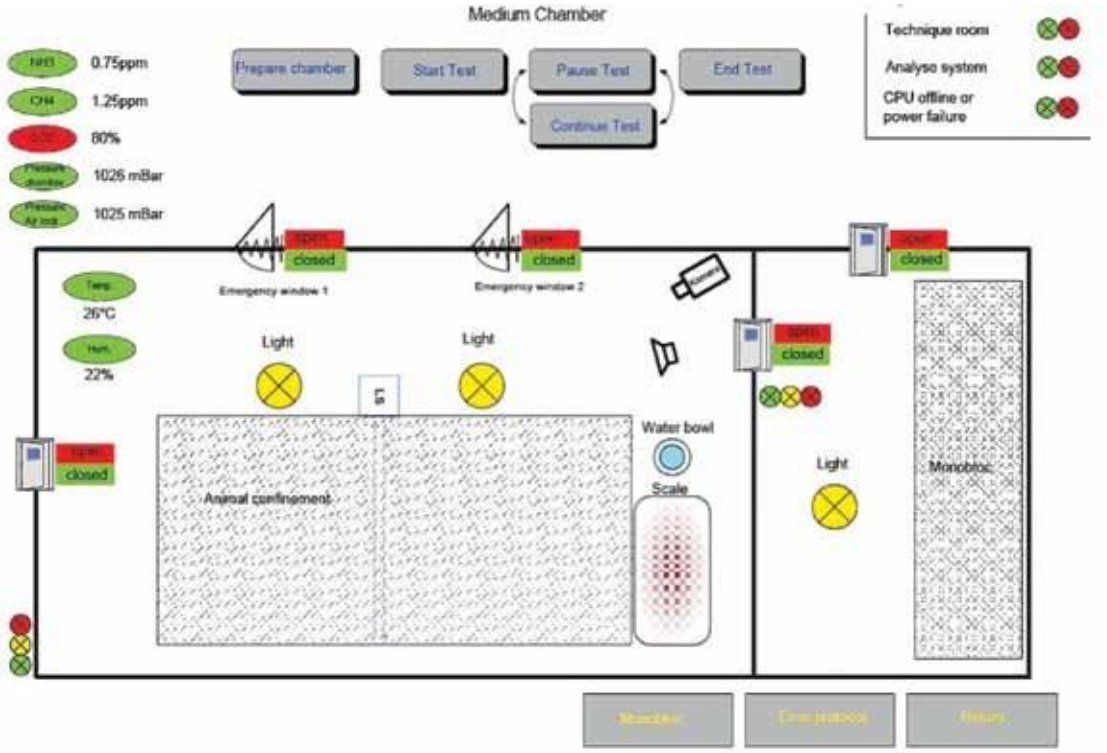


Plate 6: Project draft of how the control screen for the chambers may look like.

Plate 7: The completed chambers.

after moving to a final location where separate rooms with sufficient height for each size of chamber are planned.

Comments on the technical weaknesses or weaknesses in handling cannot be made at this time, as the system is not yet in use. In June 2011, a prototype each of the large and medium chambers is in place and ready for measurements. These prototypes will be tested and optimized before installing the second chamber (scheduled for April 2012). As many weaknesses as possible will be eliminated before the actual start of the metabolic measurements.

6.11 Description of components and equipment suppliers

Components without additional comment are used in/for the large chamber as well as in/for the medium chamber.

Chamber structure

- The sandwich panels and doors are (from the inside to the outside) stainless steel, 100mm polyurethane foam and PVDF coil coated galvanised sheet: Romakowski GmbH & Co. KG, Buttenwiesen, Germany
- Windows: Fech-Fenstertechnik GmbH & Co. KG, Buttenwiesen, Germany
- Springs and electromagnets for the emergency windows: Febrotec GmbH, Halver, Germany
- The whole chamber casing including windows and the technique for the emergency windows was assembled by Romakowski GmbH & Co. KG

Stanchion and feeder

- Huber Metallbau und Stalleinrichtungen AG, Buttisholz, Switzerland

Air conditioning

- Troges Lüftungstechnik, Vienna, Austria

Tubing to the chamber and from the chamber to the flow generator

- Hürner Kunststofftechnik, Umwelttechnik, Tagelswangen, Switzerland

Sensors in the chamber

- Temperature: Jumo Mess- und Regeltechnik AG, Stäfa, Switzerland
- Humidity: Rotronic AG Schweiz, Bassersdorf, Switzerland
- Gases: MSR electronic GmbH, Pocking, Germany

Control software

- SAIA-Burgess Controls, Murten, Switzerland

Pumps, analysers, sampling

- Promethion Metabolic Screening System by Sable Systems Europe, GmbH, Berlin, Germany. The Promethion system consists of the following items:

Large chamber

- Bev-A-Line tubing to connect the flow generator and the analyser

- Main filter (5 µm) (Solberg, provided by SableSystems)
- Flow generator and mass flow meter with integrated sub-sampling (FG-1000)
- Four-gas analyser with subsampling (GA-4)
- 8 channel flow and calibration multiplexer (CM-8)
- Interface (ACI-12)

Medium chamber

- Bev-A-Line tubing to connect the flow generator and the analyser
- Main filter (5 µm) (Solberg, provided by SableSystems)
- Flow generator and mass flow meter with integrated sub-sampling (FG-250)
- Four-gas analyser with subsampling (GA-4)
- 8 channel flow and calibration multiplexer (CM-8)
- Interface (ACI-12)

Data acquisition and logging

- Standard PC, tower case: supplied by SableSystems Europe, GmbH, Berlin, Germany
- MetaPro acquisition software: SableSystems Europe, GmbH, Berlin, Germany
- ExpeData Professional (analysis software): SableSystems Europe, GmbH, Berlin, Germany

Gas drying chemicals

- Ascarite (NaOH-coated silica), 20-30 mesh, CAS 81133-20-2: Sigma-Aldrich
- Anhydrous magnesium perchlorate (Anhydrone), granular, CAS 10034-81-8: VWR

Plate 7a: The completed chambers.

6.12 Costs of the facility

For the complete metabolic centre (2 large, 2 medium, 2 small chambers) and the ancillary equipment required, including material and work, excluding VAT.

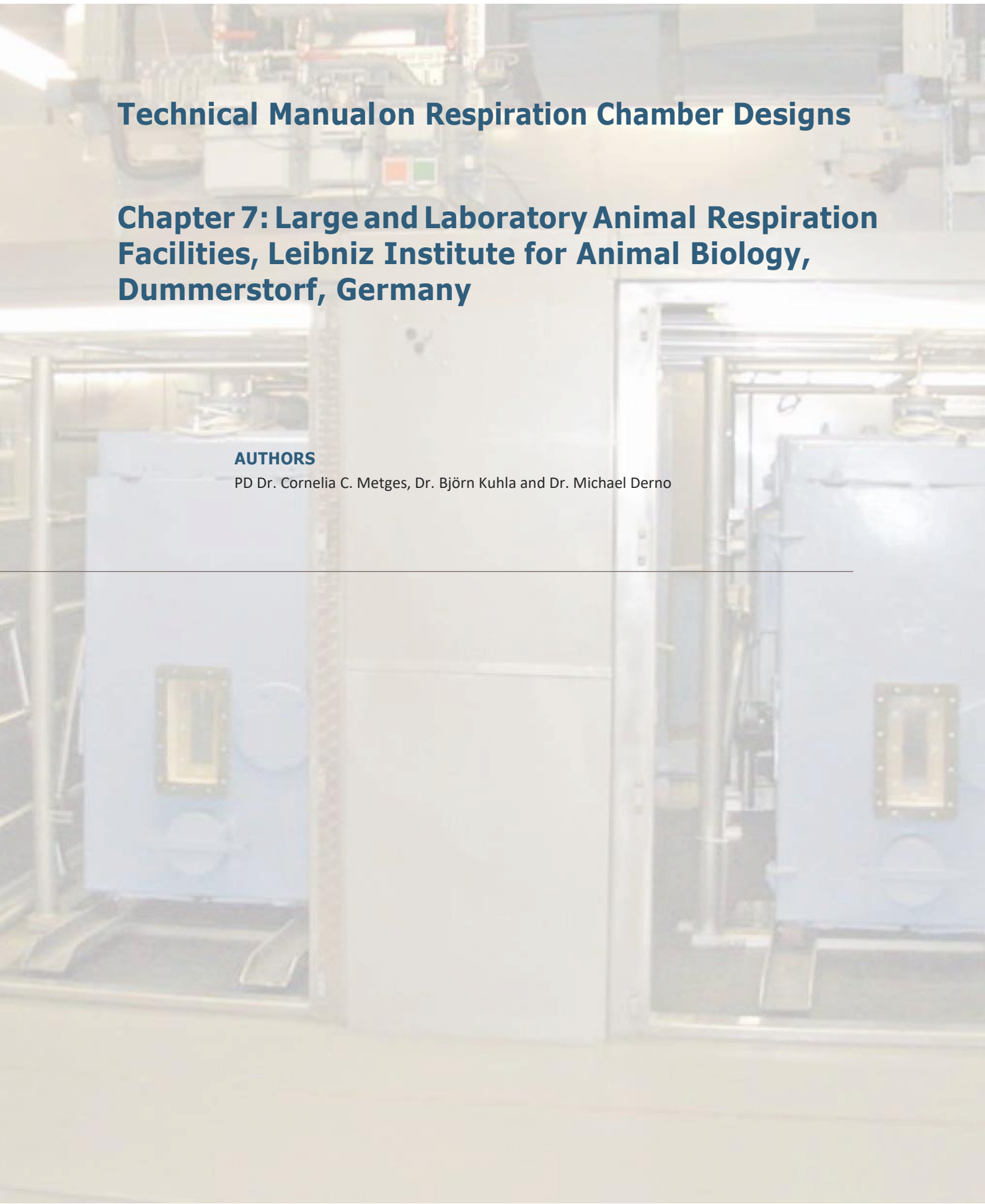
ITEMS	CHF	US$[1]
CHAMBER		
Casing including doors and windows	414,000	496,800
Animal compartment (stanchion, feeder)	52,000	62,400
Air conditioning in the chambers	453,000	543,600
Electricity, video, radio	85,000	102,000
Systems for measurement and control	215,000	258,000
Cooling (heat exchange and free cooling)[2]	195,000	234,000
Cold and warm water for the air conditioning[2]	165,000	198,000
Vapour and pressurized air system[2]	91,000	109,200
SABLESYSTEM[3]		
Gas analyser	105,000	126,000
Flow generator/mass flow meter	41,000	49,200
Sample switching system	60,000	72,000
Data acquisition and analysis	17,000	20,400
Setup and training	22,000	26,400
GAS RECOVERY[4]		
Scale	4,500	5,400
Torch	250	300
Netbook	500	600
CALIBRATION		
Calibration gases	1,300	1,560
Tubing from the gas bottles to the analysers	10,000	12,000
DRYING CHEMICALS		
Ascarite (500 g)	350	420
Mg-Perchlorate (500 g)	500	600
TOTAL COST	**1,932,400**	**2,318,880**

[1] 1 CHF = 1.20 USD.

[2] 'First' part of the air conditioning, used for the whole facility and the chambers.

[3] Values transferred from EUR to CHF, 1 EUR = 1.25 CHF; offer includes filters and computers.

[4] Only purchased once; used for all chambers.



Technical Manualon Respiration Chamber Designs

Chapter 7: Large and Laboratory Animal Respiration Facilities, Leibniz Institute for Animal Biology, Dummerstorf, Germany

AUTHORS

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Contents

Chapter 7: Large and Laboratory Animal Respiration Facilities, Leibniz Institute for Animal Biology, Dummerstorf, Germany

7.1 Summary

The open-circuit indirect calorimetry system at the Leibniz Institute for Farm Animal Biology consists of 4 respiration chambers for cattle or sheep, 4 chambers for pigs and 6 chambers for mice. All chambers as well as the gas analyzing and data acquisition system are located in a dedicated facility. Chambers can be light-cycle and climate controlled in the temperature and relative humidity range from 0°C to 35°C and 50% to 70%, respectively. The air is sucked through the chambers by rotary vane vacuum pumps having a capacity of 40 m^3/h. The airflow through the chambers can be controlled by means of a bypass. CH_4, CO_2 and O_2 concentrations are measured using specific gas analysers. Continuous monitoring CO_2 production and O_2 consumption, CH_4 emission, feed and water intake, and physical activity is possible. The facility has been designed to achieve a high standard of animal welfare.

7.2 Location of the facility

The facility is located at the campus of the Leibniz Institute for Farm Animal Biology in Dummerstorf, a village near the city of Rostock in the North of Germany at the Baltic Sea (2 hours drive to Berlin and Hamburg). The Institute is a premier European Institute for interdisciplinary basic as well as applied research on farm animals relating to resource efficient, sustainable animal production and the health and welfare of animals. The Institute employs over 300 members of staff and is a member of the Leibniz Scientific Community in Germany.

The physical address of the facility is:

Leibniz Institute for Farm Animal Biology
Research Unit Nutritional Physiology “Oskar Kellner”
Wilhelm-Stahl-Allee 2
18196 Dummerstorf
Germany
http://nutrition.fbn-dummerstorf.de/

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The facility houses all cattle and pig chambers in one room, and rodent chambers in another room. In front of the cattle chambers a balance is located. Instrumentation for calibration, gas analysis, data acquisition and data handling is located in a separate dedicated room. Adjacent rooms, equipped with temperature and humidity control, provide tie-stall housing for a limited number of cattle as well as pens and metabolic cages for pigs and sheep so that animals have short-way access to the chamber area. A video monitored surgery room with treatment stanchions for endoscopy and ultrasound, an anaesthesia box, as well as rooms for feed storage, feed preparation, milling and drying are also nearby.

7.3 Description of the chambers structure

Cattle chamber

The general outline of the system is shown in Plate 1. The open-circuit indirect calorimetry system consists of four chambers built in a pair-wise manner. Each chamber pair is constructed of stainless steel (frame, 3 side walls and roof) and separated by a divider wall constructed of acrylic glass enabling visual contact between animals in adjacent chambers (Plate 2). The entire chamber dimension is 4 m x 2 m x 2 m with a chamber volume of approximately 15.5 m^3. Inside the chamber a 2.5 m x 1.5 m stanchion is fixed which allows the individual animal to stand or lie down. The stanchion is designed for keeping animals up to 850 kg of body mass in tie stall (Plate 3). Space between the chamber walls and the stanchion allows the staff to walk around the stanchion. The chamber floor is completely covered with a rubber mat that can easily be cleaned. Feces, urine and water used for cleaning can leave the stand through openings in the back of the stanchion and pass into airtight tanks located in a cellar underneath the floor of the chambers. Each chamber is equipped with a feed bin (1 m wide, 1 m high and 0.5 m deep) that can hold 40 kg of feed (Plate 3). Feed disappearance is assessed automatically by a scale connected to an electronic registration device (PAARI, Erfurt, Germany). Water intake is registered by water flow meters equipped with electromechanical registration (Elster Messtechnik, Lampertheim, Germany). The chamber has a front door through which the animal or the smaller pig chamber (see below) can be brought in. On the rear site, the chamber can be entered through an air lock permanently rinsed with chamber air (Plate 2). To minimize disturbances of the air composition by staff entering the chamber in cases of longer-lasting feeding, milking, or sampling procedures, a facemask connected to the ambient air via flexible tubing can be put on (Plate 6). The milking device can be connected to two vacuum and one milk tubing from inside of the chamber to which a milking machine from outside can be connected (Plate 6).

Plate 1: Outline of the calorimetric system for large animals.

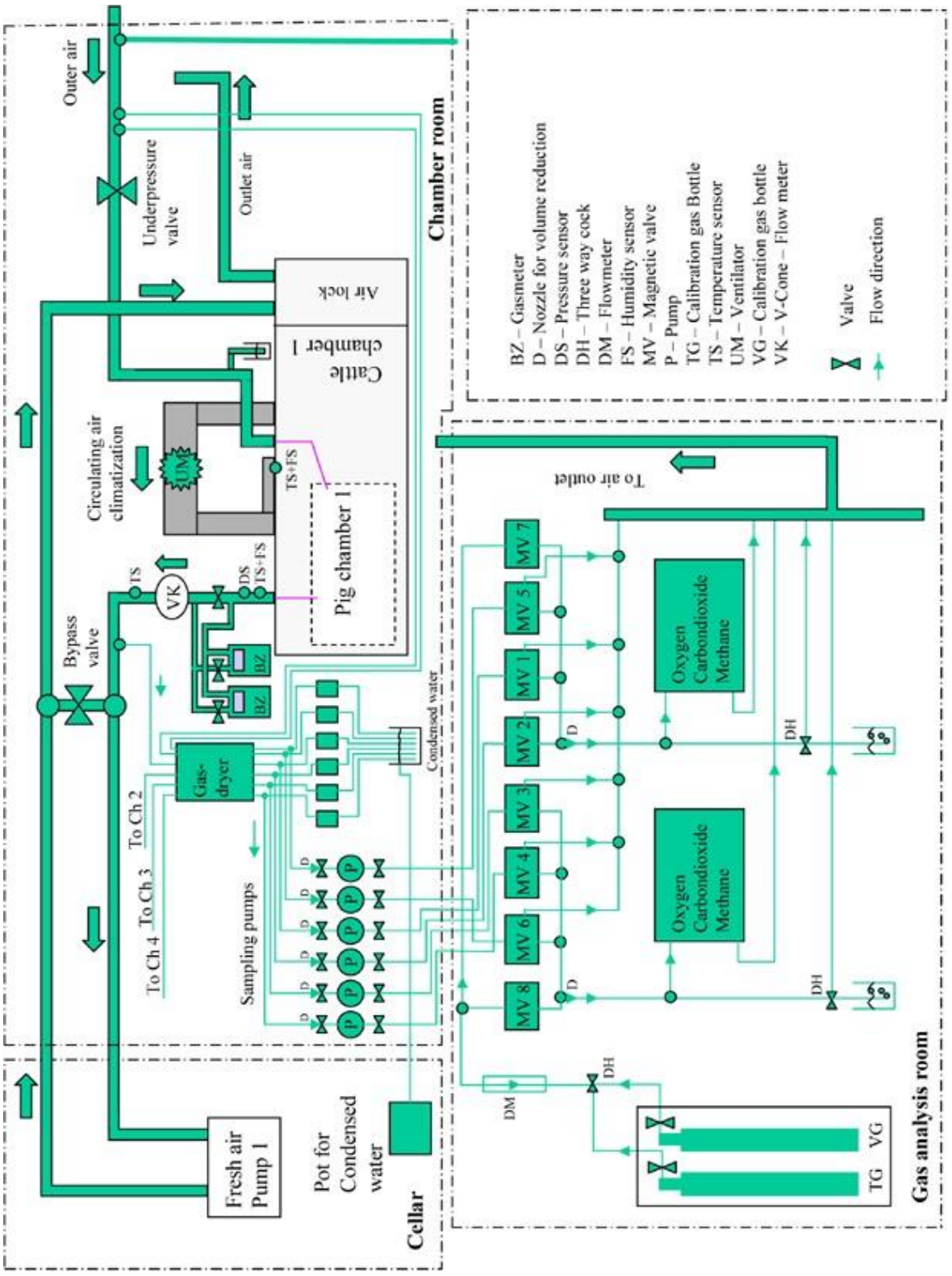
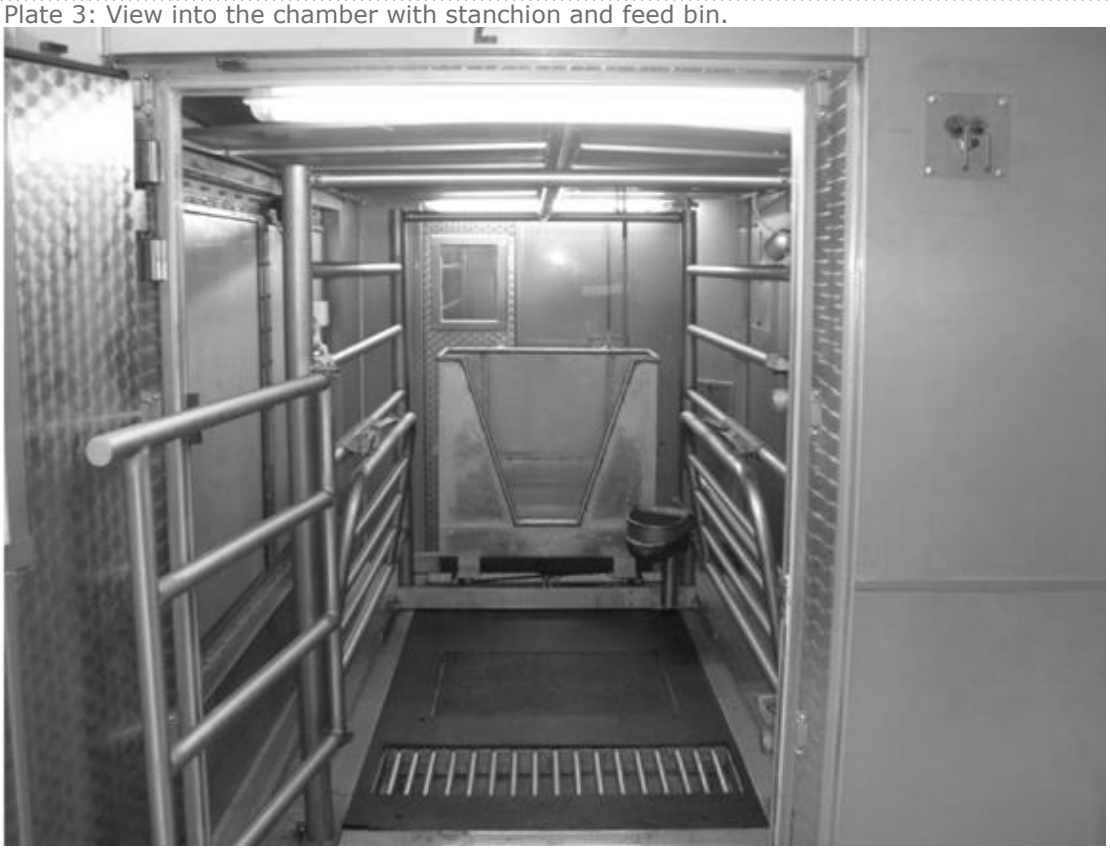


Plate 2: Respiration chamber for cattle with opened door to the airlock.

Plate 3: View into the chamber with stanchion and feed bin.



Moreover, blood can be withdrawn from catheterized animals, via a special catheter extension (4 m, inner diameter 2 mm, Perfusor, BRAUN, Melsungen, Germany) that is guided by rolls and ends outside the chamber in the air lock (Plate 4).

A communication set consisting of a microphone and a speaker inside the chamber and a monitor and a speaker outside the chamber (Orchid MD-502, Orchid Electronics, Langnau am Albis, Switzerland) allows for communication between persons in and outside the chamber. Standing and lying of the animals is registered by a photoelectric switch (SA1E, idec Elektrotechnik GmbH, Hamburg, Germany). Other physical activity is detected by a modified infrared-based motion detector (IS 120, STEINEL, Herzebrock – Klarholz, Germany) converting movements of the animal to impulses. In order to monitor the behavior of the animals, each chamber is equipped with an infrared reflector and a camera plugged into a video computer. The latter is connected via a virtual private network (VPN) to home computers of staff members allowing the animals to be observed from outside the experimental station.

The chambers are light and climate-controlled and designed to regulate the circadian ambient temperature and relative humidity in a range from 0°C to 35°C and 50% to 70%, respectively. The airflow through the cattle chamber can be set from 0–30 m^3/h. The chambers and the operating system were designed by staff members of the FBN and assembled by a contractor (LANTEC, Steinhagen, Germany). The description of the chambers is published (Derno *et al.,* Technical note: a new facility for continuous respiration measurements in lactating cows. *J Dairy Sci*. 2009; 92:2804-8).

Plate 4: Cow lying in the chamber with jugular catheter extension.

Plate 5: Chambers with attached mobile milking devices.

Plate 6: Chambers with attached mobile milking devices.

Pig chambers

For investigation on pigs, smaller steel chambers on wheels (height x width x length: 1.2 x 0.7 x 1.7 m; 1.5 m^3) can be introduced into the larger cattle chambers (Plates 7 and 8). They can be easily connected by tubes to the airflow system (airflow for pigs: 6 – 12 m^3/h) of the cattle chamber. The pig chambers are equipped with an infrared motion detector. The animals are fed through an opening at one end of the chamber, which can be closed airtight by a cover placed in a water groove. Temperature and light control is performed by the climatization system of the cattle chambers.

Plate 7: Pig chambers standing on rails in cattle chambers.

Plate 8: Opened pig chamber with ramp for driving the animals into the chamber.

Chambers for rodents

Six chambers for mice are available, which consist of transparent plastic cylinders (Ø 10 cm, 10 or 13 cm; ~ 0.8 L or 1 L) with wire mesh bottoms. Each chamber is equipped with a water flask and a hanging basket for feed pellets (Plate 9). An infrared based motion sensor mounted on the chamber cover registers physical activity of the mice. Airflow through the chamber can be regulated up to 60 l/h. Gas exchange is measured continuously in 6 to 21 min intervals, by infrared absorption based CO_2 and paramagnetic based O_2 gas analyzers (Maihak AG, Hamburg, Germany), respectively. The respiration chambers are placed inside a temperature controlled closet where temperature can be regulated from 0 to 40 °C and the light-dark cycle can be programmed (Plate 10).

Plate 9: Mice chambers equipped with water flasks and urine collecting bottles.

Plate 10: Mice chambers in the climatization closet (left) and gas analyzers (middle) and airflow meters (right).

7.4 Animal holding, feeding and cleaning

Before measurement, animals are adapted to the cambers at least 3 times for 4 h each. The criteria for successful adaptation are the voluntary entrance of an animal into the chamber, calm behavior, feed and water intake, and the lying down of an animal. Animals are moved into the chambers prior to the evening feeding and remain resident within the chambers over night before the measurement starts the next morning. This allows for priming the chamber air and the adaptation of the animal to the default ambient temperature. Depending on the specific experiment, animals remain in the chamber for two to three 24-h periods.

Feeding of cattle is performed by a staff member entering the chamber via the air-lock. Feed is normally offered as mixed ration to prevent feed selection. Lactating cows are milked at times of feeding to reduce disturbances. For daily cleaning, feed residues are removed, mats and grills are scrapped and the tanks located in a cellar beneath the chambers are emptied every morning. Once an animal has been removed, the chamber is intensively cleaned with water which drains away to the tanks located in the cellar.

7.5 Chamber airflow piping and measurement

The air is sucked through the chambers from outside of the building by rotary vane vacuum pumps having a capacity of 40 m^3/h (VT 4.40, Fuergut, Aichstetten, Germany). By means of a bypass, the airflow through the cattle chamber can be set from 0 up to 30 m^3/h, through the pig chamber from 0 to 24 m^3/h and the mice chamber from 0 to 60 L/h. A differential pressure type V-cone flow meter (McCrometer, Hemet, CA, USA) is used to measure airflow rate in cattle and pig chambers. The airflow through the mice chambers is measured by mass flow meters (Hastings Instruments, Hampton, Virginia, USA). In all cases the airflow is measured after passing the chamber. All types of gas meters are calibrated by means of wet gas meters.

7.6 Sampling, sample conditioning and analysis

The air sample for the analysis of gas composition is drawn by membrane pumps (80 l/h) (KNF Neuburger Laboport, Freiburg, Germany) located 10 cm behind the flow meters. It is then passed through infrared absorption-based analyzers for the determination of the CO_2 and CH_4 content, respectively, and through a paramagnetic analyzer for measurement of the O_2 content (SIDOR, SICK MAIHAK, Reute, Germany).

Because two sets of analyzers are available, it is possible to switch the gas sampling between two chambers. The controlling software allows the free selection of the time between switching in accordance with the flushing time of the sampling system. In each cycle, the measurement of the gas concentration of the outer air is included in order to detect any drift of the analyzers. In common experiments with large animals (cattle and pigs) the time of measuring intervals is 6 min (2 min flushing time for each unit). For special purposes, it is possible to run only one chamber with cycle times as short as 10 s. With all four cattle chambers working in parallel, data sets for airflow and gas concentrations are measured and stored every 6 minutes. It is also possible to operate chambers for cattle and mice in parallel with a maximum of 10 chambers at the same time. Barometric pressure, air temperature and relative humidity of each chamber and of the exhaust line are measured 3 times per second, averaged over 6-min intervals, and stored for further calculations.

The measured variables (gas concentrations of O_2, CO_2, and CH_4, air flow rate, feed disappearance from the feed bin, water consumption, air temperature and relative humidity in and behind the chamber, standing time, standing or lying position, activity counts, barometric pressure) are transferred to an acquisition system (Simatic, Siemens, München, Germany)

and collected by purpose-adapted software (WinCC, Version 5.1, SP 2, Siemens, München, Germany). A DELPHI-based (Delphi 2007, San Francisco, CA, USA) software (Copyright H. Scholze, FBN Dummerstorf, Germany)specifically adapted for the automatic calculation of heat production (HP) according to BROUWER (1965) [HP (kJ) = 16.18 O_2 (L) + 5.02 CO_2 (L) – 2.17 CH_4 (L) – 5.99 N (g)] collects all continuously measured data in EXCEL (Microsoft Office) files.

Collection of milk, feed, feces, and urine and subsequent analysis for carbon, nitrogen and gross energy, energy and nitrogen balance of individual animals can be calculated. Upon analyses of plasma samples, interrelations between plasma metabolites and hormone concentrations and variables obtained from gas exchange measurement or activity indices can be calculated.

7.7 Gas recovery test

The whole system can be calibrated by introduction of defined volumes of chemically pure CO_2 from a gas cylinder. The mean recovery rate is 99.9%. The analyzers are calibrated by calibration gases of known composition (zero: pure N_2, endpoint: CO_2 – 1.0 Vol%, O_2: 19.9 Vol%, CH_4: 0.1 Vol%).

7.8 Emissions calculation

The measured airflow is corrected to standard conditions and water vapour pressure. The CH_4 emission is calculated by multiplying the corrected airflow with the mean CH_4 concentration in the measuring interval.

7.9 Animal welfare and operators' safety

An alarm system that is activated by the failure of the pumps delivering fresh air to the animals is installed (comline 2016, TELENOT ELECTRONIC, Aalen, Germany) in the chambers. It responds to an increase of pressure in the chambers normally working under low pressure conditions and operates a telephone that automatically calls staff members responsible for the experiment. In case of emergency, doors of the chambers have to be opened within one hour time.

7.10 Weaknesses of the system

One weakness of the system - as compared to others - is the lack of an automatic door opening system activated in case of emergency. Thus, rapid action of the staff is required. However, animal welfare has never been compromised in case of serious risk.

Currently, it is not possible to regulate humidity in the chamber. The cooling device in the air conditioning system removes water vapour from the chamber air establishing a mean level of 50 to 70 % relative humidity in the chamber. A humidity control system will be implemented allowing the adjustment to a desired humidity value.

7.11 Description of components and equipment suppliers

- The sandwich panels and doors: stainless steel; Windows: acrylic glass; LANTEC, Steinhagen, Germany
- Stanchion: LANTEC, Steinhagen, Germany
- Feeding bin and electronic registration device: PAARI, Erfurt, Germany

- Drinking trough with electromechanical registration: Elster Messtechnik, Lampertheim, Germany
- Communication set: Orchid MD-502, Orchid Electronics, Langnau am Albis, Switzerland
- Photoelectric switch: SA1E, idec Elektrotechnik GmbH, Hamburg, Germany
- Infrared-based motion detector: IS 120, STEINEL, Herzebrock – Klarholz, Germany
- Air conditioning: provided by Hildebrandt und Kindt, Rostock, Germany
- Temperature Sensor: E + E Elektronik, Engerwizdorf, Austriaw
- O_2 gas analyzer: SIDOR, SICK MAIHAK, Reute, Germany
- CO_2 gas analyzer: SIDOR, SICK MAIHAK, Reute, Germany
- CH_4 gas analyzer: SIDOR, SICK MAIHAK, Reute, Germany
- Rotary vane vacuum pumps: Fuergut, Aichstetten, Germany
- Membrane pumps: KNF Neuburger Laboport, Freiburg, Germany
- Flow meter: McCrometer, Hemet, CA, USA
- Data acquisition: Standard PC, Software; Simatic, Siemens, München, Germany (WinCC, Version 5.1, SP 2, Siemens, München, Germany)
- Data handling: DELPHI-based (Delphi 2007, San Francisco, CA, USA) tailored software (Copyright H. Scholze, Leibniz Institute for Farm Animal Biology, Dummerstorf, Germany)

7.12 Costs of the facility

Carcass, stanchion, feed bins (including labour)	200,000 €
Flow meters, Temperature - humidity sensors, sample pumps, tubes, wires	60,000 €
Climatization	50,000 €
Feed bin balances, electronic registration	7,000 €
Milking system	6000€
Gas analyzers	24,000 €
Data acquisition and control unit	15,000 €

Design and part of construction was made by staff members of the Leibniz Institute.

Technical Manualon Respiration Chamber Designs

Chapter 8: Small Ruminant Chambers of Universiti Putra Malaysia

AUTHORS
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Contents

Chapter 8: Small Ruminant Chambers of Universiti Putra Malaysia

8.1 Summary

Five open circuit respiration chambers have been constructed in the year 2016 to quantify methane production from small ruminants (goats and sheep). Each chamber is of approximately 2.0 m^3 internal volume and consists of a clear polycarbonate shell affixed to a galvanized iron frame so the animals can see each other. The chambers are located in a well-ventilated air-conditioned room. Flow rate of the chamber is measured manually at the exhaust pipe located at the top rear end of the chamber every 30 minutes using an anemometer (SDL300 Extech Instruments, USA). A sample of gas is drawn from the exhaust and ambient air using an aquarium air-pump through a polyethylene gas sampling tube into 1L gas bags. The concentration of methane in the gas bags is determined using gas chromatography. Methane recovery through the chambers is determined by injecting a known flow of methane approximately two days prior to each experimental run.

The above chambers were designed and constructed with limited budget and could serve as an example of low-cost respiration chambers for use in developing countries. At this stage none of the measurements require expensive or complicated measuring equipment except for a gas chromatograph which is available in most laboratories. Because of the limitation of the lower detection limit for gas chromatography we describe some upgrades to this basic design. The first upgrade is the use of a portable gas analyzer (Horiba PG-300, Japan) to determine concentrations of methane and carbon dioxide in real time. This method increases the accuracy of the measurements. However, this requires the use of a data logger and additional measurements of barometric pressure, humidity and temperature in the chamber and the ambient air to calculate a standard gas flow. A second upgrade is the use of a Venturi flow meter which measures the air flow through the chambers also in real time.

8.2 Location of the facility

The physical address of the facility is:

Institute of Tropical Agriculture and Food Security

Universiti Putra Malaysia

Serdang 43400

Malaysia

Contact persons:

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The chambers are located in a well-ventilated room at the Animal Research Farm of the Institute of Tropical Agriculture and Food Security (ITAFoS), Universiti Putra Malaysia, Serdang, Malaysia. The university campus is located about 25 km south of Kuala Lumpur. The ITAFoS's animal research farm is divided into two sections; one section consists of facilities for poultry research and another section for ruminant research. The latter consists of two wings; the cattle wing which has 20 units of individual- and two units of group pens and the other wing has 40 individual- and three group-pens for small ruminants (goats and sheep) (Plate 1). The small ruminant wing also equipped with 12 units of digestibility crates. The respiration chambers room is located approximately 60 meters from the small ruminant barn making transferring of experimental animals from the feeding pens to the respiration chambers very easy and with minimal stress to the animals. The cattle and small ruminant wings are connected by a central feed preparation and storing facility.

Plate 1: Ruminant Research Facilities at the Universiti Putra Malaysia

8.3 Description of the chambers structure

The five chambers are arranged in a row in a well-ventilated air-conditioned room with the front doors of the chambers facing one side of the room and the rear doors facing the entrance of the room to facilitate easy transferring of animals in and out of the chambers (Plate 2). The room housing the five chambers is 13 m (L), 6.5 m (W) and 3 m (H) in size and has air-conditioners installed to provide an average room temperature of between 22 to 25^{O}C and relative humidity of below 50%. Currently, the five chambers only occupy about half of the room with the remaining area being used for preparation and to keep several research facilities, including a refrigerator and a freezer.

The interior volume of each chamber is approximately 2.0m^3 [1.90m (L) x 0.85m (W) x 1.25m (H)] and made of a clear polycarbonate (4 mm) shell glued to a galvanized iron frame (2.5 cm x 2.5 cm). Four heavy-duty roller-wheels are attached to the four corners at the bottom of each chamber to allow it to be moved when necessary. The whole front and rear of the chambers are doors. The doors are latched with a manually operated latch and a rubber seal ensures that the doors are relatively air tight. When opened, the front door allows free accessibility to

the feed and water troughs which are attached to the crate holding the animal inside the chamber (Plate 3), while the rear door allows for transferring of an animal in and out of the chamber as well as removing and cleaning of urine and dung collected in a tray below the floor of the crate (Plate 4). The chamber has a small inlet opening 20cm diameter at the front end on top of the chamber for ambient air to come in. The size of the opening can be controlled by an “open and shut” polycarbonate sheet to control the amount of ambient air to be drawn into the chamber and keep a negative pressure within the chamber.
The crate holding the animal inside the chamber is approximately 1.43m (L), 0.64m (W) and 0.97m (H), and made of galvanized iron frame (2.5 cm x 2.5 cm). Similar to the chamber, the whole front and rear of the crate are doors. When latched, feed and water troughs are attached to the front door of the crate while the rear door is used for transferring of the animal in and out of the crate. The crate is seated on four heavy-duty wheels at the corners to allow the crate to be rolled in and out of the chamber (Plate 5).

Plate 2: The five open circuit respiration chambers arranged in a row (top) and an on-going experiment measuring methane production from goats (bottom)

Plate 3: Front end of the chamber showing when door is opened, operator has free accessibility to feed trough and also shows strip of black fabric-lining to ensure no leakage of chamber air when operates under low flow rate.

Plate 4: Views from rear end of the chamber showing the waste collection tray, small window at the lower section of the rear door which when opened allows accessibility to the waste collection try without need to open the whole rear door (left). Roller-wheels are attached to the chamber and animal crate to allow them to be moved easily when necessary.

Plate 5: View from the rear end of the animal crate in the chamber. Note the Extended Metal floor and the location of the waste collection try below it for complete collection of urine and feces voided.

8.4 Animal holding, feeding and cleaning

Experimental animals are kept individually in crates which can be rolled in and out of the chambers (Plate 2). Each crate [1.4 m (L), 0.65 m (W) and 0.90 m (H)] is made of galvanized iron bars (2.5cm x 2.5cm) and consisted of front and rear doors plus a waste collection tray below the heavy duty metal floor (Plate 4). Feed and water troughs are attached to the front end of the crate and feed and drinking water are offered through the front door (Plate 3) while urine and feces collected in a tray can be removed from a narrow window at the lower end of the rear chamber door (Plate 4) and these feeding and cleaning activities are done each morning before gas measurements start.

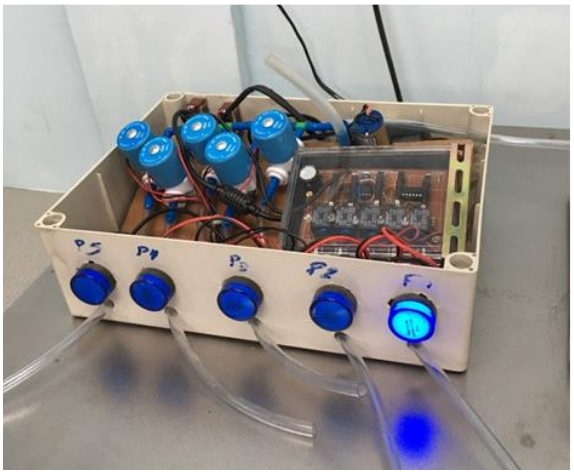
8.5 Chamber airflow piping and measurement

The flow rate of the chambers can vary between 205 and 230 L/min and is set depending on the size of the animals to be tested. The lower limit for the flow is the flow where a minimum of 2kPa pressure difference between the outside and the chamber inside is maintained. The highest level is determined by the exhaust fan used.

The air outlet is a 10 cm (4 inch) diameter standard polyethylene exhaust pipe located at the top rear end of the chamber (Plate 6). The other end of the exhaust pipes of the five chambers are joined to a common exhaust fan (60cm or 24 inch Wing Ton “Var” Series Exhaust Fans, Malaysia), located above the ceiling of the room, which draws the air from the chambers. The capacity of the exhaust fan is 133 m3 /min. The flow rate of the air out of the chamber is measured manually by inserting an anemometer (SDL300 Extech Instruments, USA) into the airstream of the exhaust pipe, sealing the port and letting the readings stabilize.

Plate 6: Air outlet exhaust pipe located at the top rare end of the chamber. The five exhaust fan are joined to a common exhaust pump above the ceiling

Plate 7: Sample of gas is draw from the exhaust and ambient air using an aquarium air-pump through a polyethylene gas sampling tube into gas bags (left) and the concentration of methane determined using Gas Chromatography or the gas is drawn into the gas analyzer (right).



8.6 Sampling, sample conditioning and analysis

A sample of gas is drawn from the exhaust and ambient air using an aquarium air-pump through a polyethylene gas sampling tube into 1L gas bags (Plate 7 left). The concentration of methane in the gas bags is determined using Gas Chromatography. At this stage the system is all manual and no computer or data logging interface is required for operation of the system.

Recently an upgrade for methane measurements was performed. Instead of using a gas chromatograph the samples are collected automatically from the exhaust pipes using a portable gas analyzer. A sample of gas (1L/min) is drawn from the exhaust and ambient air through a gas sampling tube by an inbuilt pump into the gas analyzer (Horiba PG-300, Japan) to determine concentrations of methane and carbon dioxide (Plate 7 right). The gases are dried in the analyzer before being measured by an infrared cell. To automate the sample collection, a 5-port sampler controlled by an inbuilt timer to switch the gas flow between the chambers and ambient air was constructed by UPM and added to the system (Plate 7c). Gas concentrations and chamber air flow rate measurements are stored in Excel spreadsheet for each chamber allowing methane measurements every 30 minutes.

8.7 Gas recovery test

Recovery tests are carried out by injecting a constant stream of methane into the chamber from a pure methane gas bottle. Gas concentrations are measured over a period of several hours. A steady state using the current settings is reached after approximately 90 min. The gas flow is measured using a Cole-Parmer flow meter (Model # 32908-63, Cole-Parmer Instrument Company, Illinois, USA) and the recovery of methane is calculated from the measured over the calculated methane concentration. Various concentrations of methane flows are tested in different chambers.

8.8 Emissions calculation

The emissions from the chambers are calculated from the dry gas flow and the proportion of methane recorded by the gas analyzer. The total air flow is calculated form the average of the daily flow measurements.

Calculation of dry gas flow

DGF = WF*(100-((a + b*T + c*T2 + d*T3 + e*T4 + f*T5 + g*T6)*RH/P)/100)

Calculation of standard gas flow

SGF = DGF * P/(T + 273.15) * ST/SP

Calculation of methane production

CH4 [g/d] = ((MC – MCb) / 106) / 22.414*16.043 *SGF *1440¥

¥ * 1440 min/day

Terms

Standard temperature (ST) = 273.15 K

Standard pressure (SP) = 1013.25 mbar

Molar weight CH4 = 16.043 g/mol

Gas constant = 22.414 L/mol

MC = methane concentration [ppm]

MCb = Background methane concentration [ppm]

WGF = wet gas flow [L/min]

DGF = dry gas flow [L/min]

SGF = standard gas flow [L/min]

T =Temperature [°C]

RH = relative humidity [%]

P = pressure [mbar]

Coefficients	Water Vapour
a	6.1117675
b	0.443986062
c	0.01430533
d	0.000265027
e	0.000003022469940
f	0.000000020388631
g	0.000000000063878

8.9 Animal welfare and operators' safety

The system ensures that all environmental conditions in the chambers are within the thermo-neutral zone for the animals, and there is minimal exposure to stress or risk (Plate 2 bottom). To achieve the above, flow rate, temperature, relative humidity and ambient pressure and concentrations of carbon dioxide are recorded and monitored every 30 min. At the moment, the chambers do not have auto-safety doors that automatically open when there is a power failure. In the event of a power failure, the front and rear doors of the chambers can be opened manually. Thus, the presence of a human operator is necessary throughout the duration of any experiment. Since, several parameters (flow rate, temperature, RH and carbon dioxide concentration in the chambers) are manually recorded every 30 min per chamber, the presence of human operator is always required in the chamber room and is available to open the chamber doors in case of any emergency including power failure.

8.10 Weaknesses of the system

The system was built with a limited budget. However, it has proven to be reliable and with a good recovery rate of the targeted gas (methane) and thus provides a good example for other researchers, particularly those from developing countries who are interested in building a similar facility. The main weakness of the system is its discontinuous measurements for gas flows and concentrations. This has partly been overcome by automating the methane measurements but is still the case for the flow measurements. Currently the system does not have the auto-open chamber doors in the event of power failure and thus the presence of human operator at all time near to the chamber room is essential. In addition, several parameters (flow rate, temperature, RH and carbon dioxide concentration in the chambers) are manually recorded every 30 min, thus the presence of human operator is always required. Currently the flow readings are manually collected which could be improved using a more reliable auto-measurement system.

8.11 Description of components and equipment suppliers

Chamber design and construction: Selves with local contractor
Crates design and construction: Selves with local contractor
Air ducting system and fans: Selves with local contractor
Anemometer, flow meter, Data logger and Gas analyzer: Local scientific equipment suppliers

8.12 Costing of the facility complete system with ancillary equipment

ITEMS	US$
Labor	
Design of the system	In-house
Building of chamber (5 units)	5,000
Animal crate c/w feed trough (5 units)	4,000
Piping air circulation and sampling lines	500
Electrical wiring and air-conditioners	2,000
Equipment	
Anemometer 1 unit (for flow rate measurement)	800
Data logger 5 units (for temperature, relative humidity and pressure)	1,000
Cole-Parmer flow meter	3,000
Gas analyser (for methane and carbon dioxide)	25,000
Calibration gases	1,000
Computer and data acquisition system	1,000
Total Cost	43,300