Final report

Veterinary feed additive safety program for the Australian feedlot industry

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Abstract

The Australian feedlot industry relies on mineral and vitamin supplements to fill nutritional gaps in rations and provide a method of effective administration of veterinary feed additives to promote cattle production and health. A common form of supplementation utilised by the industry is molasses based liquid supplements. A survey of supplement manufacturers and feedlots in combination with supplement sampling from feedlots was undertaken to investigate how the manufacture and management during storage of these supplements may affect animal performance, welfare and food safety. A review of liquid supplement mixing systems was also undertaken and supported by a benchtop experiment.

The survey, feedlot supplement samples and benchtop sample results were assessed by basic statistical comparisons of means, standard deviations and co-efficient of variation.

Domestic and international quality assurance programs play an important role in supporting supplement manufacture. Particle size was a key requirement for low solubility and toxicity risk ingredients. Survey showed 96% of feedlots using monensin. Management of supplement storage tanks at the feedlot important for supplement integrity and consistency. Laboratory error was identified across all components analysed. Aeration should not be used as a mixing method, but re-circulation improves nutrient consistency during extended storage periods.

Feedlot industry including supplement manufacturers and supplement tank suppliers would benefit from recommendations of best practice use and management of liquid supplements. Liquid supplements should only be sent for analysis in duplicate and to labs that can achieve a minimum 90% sensitivity.

Executive summary

Background

The Australian feedlot industry relies on mineral and vitamin supplements to fill nutritional gaps in rations fed and provide a method of effective administration of veterinary feed additives to promote cattle production and health. A common form of supplementation utilised by the industry is molasses based liquid supplements. The method in which these supplements are manufactured and then managed at the feedlot can have animal welfare and food safety implications. This is particularly relevant if the supplement undergoes separation, changing the concentration of nutrients fed to cattle.

In the USA, mixing directions are provided for liquid supplements containing monensin due to large particle size (850µm). Mixing is rarely practiced by Australian feedlots as supplement manufacturers source 75µM monensin. The potential of monensin to separate within a liquid supplement during storage has direct animal welfare and health implications. Evaluating the effects of mixing liquid supplements on integrity during storage was investigated. In situations where mixing improves liquid supplement stability and consistency due to storage period and/or type of supplement used, mixing systems for storage tanks were investigated.

Objectives

The objectives of the survey include:

1) Survey 30 feedlots across Australia to identify methods by which feedlot supplements are stored, managed and used.
2) Survey supplement manufacturers of both dry and liquid supplements to assess manufacturing requirements and management of manufacture, storage and delivery.
3) Assess use and management of veterinary feed additives by supplement manufactures and feedlots.
4) Collect supplement samples from the surveyed feedlots to determine baseline variation of monensin, moisture, ash, calcium, copper, zinc and potassium.
5) Review agitation and recirculation systems for liquid supplements.

The objectives of the benchtop study include:

1) Determine by benchtop assessment the effect of mixing liquid supplements by aeration or recirculation on supplement consistency over a 12-week usage period
2) Assess effect of mixing method on consistency of monensin, ash, zinc, calcium, copper and zinc over a 12-week usage period

Methodology

A survey involving 30 feedlots, 4 liquid and 4 dry supplement manufacturers was undertaken to establish an overview of feedlot supplement manufacture as well as storage, use and management. Interpretation of survey results assessed by a frequency table and assessing data by mean, maximum/minimum values and standard deviation.

The feedlots using liquid supplement were required to sample a supplement delivery six times in duplicate. One duplicate sample was removed directly from transport tank providing a reference sample, with the remaining five duplicate samples removed directly from the storage tank representing 95%, 75%, 50%, 25% and 5% of the storage tanks capacity. All duplicate samples collected were analysed for monensin (mg/kg), moisture (%), ash (g/100g), calcium (g/kg), zinc (mg/kg), copper (mg/kg) and potassium (g/kg).

For the benchtop project, the experimental design was based on comparing three mixing treatments – control, aeration, re-circulation, and two supplement types – blended and suspension over a 12-week storage period. Each week of the 12-week storage period, the mixing treatments were applied prior to supplement removal. Samples were collected and stored from both the top and bottom of the squib on week 1, 7, 11 and 12 representing 95%, 50%, 5% and empty of squib contents. A duplicate sample from each supplement was taken before project began and used as delivery comparison.

All duplicate liquid supplement samples collected were analysed for monensin (mg/kg), moisture (%), ash (g/100g), calcium (g/kg), zinc (mg/kg), copper (mg/kg) and potassium (g/kg). Results were provided on a dry matter basis. The samples were summarised by calculating mean, standard deviation, co-efficient of variation and error evaluations.

Results/key findings

- Risk of supplement separation minimised by manufacturers sourcing ingredients with appropriate particle size and relying on QA systems
- Monensin should only be sourced and controlled by the supplement manufacturer, thereby controlling product form and ensuring particle size is no greater than 75µM.
- 96% of feedlots surveyed using monensin
- Feedlot liquid supplement tanks need to be cone based, fitted with separate delivery lines, fully emptied one tank at a time after each fill, with a turnover period of 30 days.
- Aeration should not be used to improve consistency liquid supplements

Benefits to industry

The feedlot industry can have confidence in supplement manufactures to produce products that meet their needs and are safe to use.

The integrity and consistency of liquid supplements can be improved by relatively simple management practices at the feedlot such as ensuring tanks are emptied prior to re-filling and using separate delivery lines for different liquid ingredients.

Future research and recommendations

The findings from this project identifies the following further research and recommendations:

- Supplement taken for analysis require duplicate samples and request from laboratory testing sensitivity. Require sensitivity of at least 90%.
- Provide recommendations of best practice use and management of liquid supplements to feedlot operators, supplement manufacturers and supplement tank suppliers
- Collecting retention samples of supplement from each delivery received at feedlot

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1. Background

The feedlot supplement is an important component of the ration. Its function is to fill dietary gaps derived from ration ingredients (minerals and vitamins) and enhance animal production and health with the addition of natural and veterinary feed additives. In Australia, feedlot supplements are provided in liquid form, using molasses as the carrier or in a dry form, using carriers such as cereal grain, cereal grain by-products, protein meal or limestone. Liquid supplement is the most common form utilised, providing advantages in terms of cost, manufacturing and ingredient flexibility, ease of storage and loading and greater inventory control.

Supplements provide an ideal method for the addition and distribution of low inclusion ingredients such as veterinary products. Veterinary products registered for beef production include glycophospholipids (bambermycins - flavophospholipol), ionophores (monensin, lasalocid, salinomycin, narasin) and medically important antimicrobials such as tylosin, virginiamycin, chlortetracycline and oxytetracycline. These additives can be highly concentrated (+100,000mg/kg) but are required at a low daily intake (100 to 300mg/day) relative to daily ration intake (+10kg/day). Using supplements to promote distribution throughout the ration as well as removing the responsibility of the feedlot for accurate use each day, avoids risk of loading and mixing errors. Under dosing can have negative impacts on production such as reduced performance and product efficiency, while overdosing could lead to animal toxicity and death.

In the USA, mixing directions are provided for supplements containing monensin due to the 850µM particle size of the monensin carrier. Mixing methods include re-circulation and aeration. Re-circulation is the process of running supplement out of the tank, through a pump and returning it to the top of the tank. Aeration is the process of pumping compressed air into the bottom of the tank and using the air movement through the tank for agitation. In contrast to the USA, Australian liquid supplement manufacturers source monensin with a guaranteed 75µM or less particle size, reducing requirement of mixing to stop monensin from separating. Mixing methods may compromise supplement stability. For example, re-circulation can shear the suspending agent, reducing binding capacity and aeration may generate oxidative reactions.

With the Australian feedlot industry reliant on supplements to satisfy production requirements and provide a reliable and safe method of supplying veterinary feed additives, the industry was interested in reviewing how supplements are managed during manufacture as well as management and use at the feedlot. Management may include implementation of mixing systems on existing tanks to promote stability during storage period, as well as greater feedlot quality assurance by collecting retention samples at delivery and using on-feedlot assessment methods to determine supplement integrity.

A survey of Australian feedlots and supplement manufacturers was used to identify how veterinary feed additives are delivered to feedlot cattle by assessing the methods of supplement management by both manufacturer and feedlots, along with the presence of agitation or recirculation systems. To quantify supplement consistency, selected feedlots were requested to take five samples from their storage tank (in duplicate) and sample during use from the one supplement delivery. An additional sample from the delivery tanker was taken to identify baseline variation of supplement components including veterinary additive concentration. The benchtop project was undertaken to determine the effect of using aeration or recirculation on both blended and suspension supplement on integrity and consistency during a 12-week usage period.

2. Objectives

The objectives of the supplement manufacturer and feedlot survey include:

1) Survey 30 feedlots across Australia to identify methods by which feedlot supplements are stored, managed and used.
2) Survey supplement manufacturers of both dry and liquid supplements to assess manufacturing requirements and management of manufacture, storage and delivery.
3) Assess use and management of veterinary feed additives by supplement manufactures and feedlots.
4) Collect supplement samples from the surveyed feedlots to determine baseline variation of monensin, moisture, ash, copper, zinc and potassium.
5) Review agitation and recirculation systems for liquid supplements.

The objectives of the benchtop study include:

1) Determine by benchtop assessment the effect of mixing liquid supplements by aeration or recirculation on supplement consistency over a 12-week usage period
2) Assess effect of mixing method on consistency of monensin, ash, zinc, calcium, copper and zinc over a 12-week usage period

3. Survey methodology

A survey of feedlots (30) and supplement manufacturers (4 liquid, 4 dry) was undertaken to establish an overview of feedlot supplement manufacture as well as storage, use and management. Survey questionnaires were developed for supplement manufacturers (see Appendix 8.1.1) and feedlot operators (see Appendix 8.1.2). Surveys were completed by phone or during an on-site visit.

Interpretation of survey results assessed by a frequency table and assessing data by mean, maximum/minimum values and standard deviation.

3.1 Supplement manufacturer survey

A total of eight supplement manufacturers were surveyed, including four liquid supplement manufactures supplying both suspension (with limestone) and blended (no limestone) molasses based supplements and four dry supplement manufactures supplying a combination of pellet, meal and mineral concentrates. In combination, the four liquid supplement manufactures had locations in QLD and VIC. In contrast, the four dry supplement manufactures in combination covered all feedlot surveyed states.

The supplement manufacturer survey covered quality assurance systems, provision of ingredient and nutrient specification recommendations to ensure manufacturing ease, product integrity and veterinary additive products were held by facility and their associated management (Appendix 8.1.1). Supplement manufacturers were asked about situations where supply of supplement had caused an adverse effect either during manufacture, storage, transport or when the product was fed.

3.2 Feedlot supplement survey

A total of 30 state licenced and NFAS accredited feedlots were surveyed. These feedlots were selected to represent major feedlot operating states (QLD, NSW, VIC, SA and WA) and cattle feeding capacities (Very large (VL) +25,000; Large (L) 10 to 25,000; Medium (M) 5 to 10,000; Small (S) less than 5,000 head) as described in Table 1.

Table 1. Selection of feedlots for supplement survey based on number of feedlot operations and feedlot capacity by State.

State	QLD	NSW	VIC	SA	WA
Feedlot No.	204	73	16	16	38
Head Capacity					
VL	9	3	-	-	-
L	16	2	2	2	2
M	11	10	1	2	-
S	168	56	13	12	36
Selected for survey					
VL	2	1	NA	NA	NA
L	3	2	1	1	1
M	1	3	1	1	NA
S	6	4	2	2	2
Total	12	10	4	4	3

Of the 347 feedlots available, those selected for survey represented 40% of QLD feedlots, 33% of NSW feedlots, 13% of both VIC and SA feedlots and 10% of WA feedlots.

Feedlot selection emphasised those using liquid supplements (30 feedlots using either suspension or blended) but also included three feedlots using the range of dry supplement options (pellet, meal or mineral). The dry supplement feedlots were all from WA.

The feedlot supplement survey covered supplement inclusion level and usage, infrastructure, supplement composition, method of delivery, veterinary feed additive use (included in supplement and/or added directly to ration), quality assurance and risk management (Appendix 8.1.2).

3.3 Supplement sampling & analysis

The feedlots using liquid supplement were requested to sample a supplement delivery six times in duplicate from the storage tank during its use, providing twelve samples in total. One duplicate sample was removed directly from transport tank providing a reference sample, with the remaining five duplicate samples removed directly from the storage tank representing 95%, 75%, 50%, 25% and 5% of the storage tanks capacity. The protocol for sampling is provided in Appendix 8.2.

Each feedlot was provided with a sampling kit which contained twelve 75ml plastic jars with screw top lids, twelve zip lock bags with sufficient size to hold single sample jar, sticky labels and written instructions for sampling, labelling, storage and shipping (Appendix 8.2). Feedlots were requested to use a specific labelling technique to ensure the testing laboratory could not identify sampling sequence (Appendix 8.2), removing sampling bias as well as providing blinded results. Feedlots were asked to store samples until sampling procedures were completed. Storage instructions required samples to be held in a cupboard out of direct sunlight. Before shipping, feedlots were requested to place each sample jar in a zip lock bag to protect sample integrity in case of sample jar leakage or breakage. Samples were posted to a project team member in Brisbane for collection and co-ordinating samples to be sent to Symbio Alliance for testing. The majority of samples were sent in separate duplicate groups as described by Appendix 8.3. Feedlots 10, 19, 27 and 28 had both duplicates sent to Symbio on the same day.

Of the 30 feedlots selected for survey and sampling of supplement, 19 feedlots provided supplement samples, 7 collected the supplement samples but unfortunately were unable to access postage, 2 feedlots provided no correspondence, and 2 feedlots had samples lost during postage. Of the 19 feedlots that provided samples for analysis, 13 provided the full 12 samples, 5 feedlots provided 10 samples (Feedlot ID 1 missing 5% sample, 13 missing 25% sample, 17 missing 75% sample, 19 missing 5% sample and 28 missing 25% sample) and one feedlot provided 9 samples (Feedlot 27 missing duplicate 50% sample and second duplicate of 75% sample).

All duplicate samples collected were analysed for monensin (mg/kg), moisture (%), ash (g/100g), calcium (g/kg), zinc (mg/kg), copper (mg/kg) and potassium (g/kg). Results were provided on an 'as is' or wet basis. Monensin was analysed using an in-house LC-MSMS method. The process involves extracting the analytes from the samples by sonication in an aqueous methanol solution. The extract is then centrifuged, diluted and filtered. Quantitative and confirmatory analysis is performed simultaneously by LC/MS/MS (liquid chromatography, mass spectrometry) and relying on an internal standard calibration. Moisture was analysed using a vacuum oven based on the AOAC 934.01 method. Ash was analysed using the method based on AOAC 923.03 and 920.153. Zinc, Calcium, Copper and Potassium levels within the liquid supplement samples were analysed using an inductively coupled plasma mass spectrometer.

The analysed values were converted to a dry matter basis to remove the effect of varying supplement moisture levels. Dry matter values were calculated using the corresponding sample moisture level. An example of the supplement sample analysis is provided in Appendix 8.4.

4. Results and discussion

4.1 Supplement manufacturer survey

The following frequency tables summarises the survey results of the supplement manufacturers for quality assurance and procedures (Table 2), recommendations for supplement manufacture (Table 3), ionophores/antimicrobials in use (Table 4) and whether site has had an adverse experience with supplement on site, during transport or at the feedlot (Table 5).

4.1.1 Supplement manufacturer quality assurance & procedures

Surveyed supplement manufactures have well established quality assurance (QA) systems with 100% surveyed having one or more QA systems in conjunction with Feedsafe®. The purpose of QA is to develop and implement procedures and actions within a production system which ensures the final products meet a minimum standard and in the case of feed manufacture, identify and develop procedures that manage risk. Ultimately, the objective of these QA systems is to ensure animal health and food safety for the consumer. Specific examples include meeting trade requirements for antibiotic free product, minimising the risk of toxicity, chemical residue, microbial contamination (E.coli, Salmonella) and eliminate the risk of Bovine Spongiform Encephalopathy (BSE) transfer from animal products to humans.

Surveyed supplement manufacturers are all members of the Stock Feed Manufactures Council of Australia (SFMCA). The SFMCA is the peak national body representing the Australian animal feed manufacturing industry (www.sfmca.com.au). SFMCA developed and implemented the QA program called FeedSafe® (www.feedsafe.com.au). FeedSafe® uses the quality standards of Good Manufacturing Practice (GMP) and Hazard Analysis and Critical Control Points (HACCP).

Table 2. Summary of supplement manufacture survey quality assurance (QA) responses (expressed as percentage)

	Supplement Manufacture	
Manufacture QA	**Dry**	**Liquid**
QA Systems in use	100	100
- Feedsafe®	100	100
- One or more other QA (ISO, GMP, HACCP)[1]	50	100
Formulation software in use	50	100
Batching software in use	75	50
QA Procedures		
Copy of medications / S4 antimicrobial orders	100	100
Ingredient sampling & testing[2]	100	75
Carriers tested	100	100
Ionophore testing[3]	50	100
Supplement sampling	100	100
Uses antioxidants	100	100
Use of S4 anti-microbials	100	25
Antimicrobial free manufacturing line	50	50
Flushing protocol	100	100
Assesses mixer consistency	100	100
CV target	75 CV5; 25 CV10	100 CV10

[1]International Organisation for Standardisation, Good Manufacturing Practice, Hazzard Analysis and Critical Control Points; [2]All dry manufacture sample & test ingredients for nutrients, chemical residues & heavy metals. All liquid supplement manufactures sample & test molasses daily; [3]Where no, manufacture use certified suppliers as required by FeedSafe®

The QA system used by the Australian feedlot industry, the National Feedlot Accreditation Scheme (NFAS) also recognises the value of an audible QA system, requiring manufactures to have FeedSafe® accreditation as described in Figure. 1.

ELEMENT FS4 – Additive, Premix and Liquid Supplement Manufacturer Accreditation

OUTCOME: ***Systems have been implemented to ensure the safety and integrity of products through sourcing and documenting all additives, premixes and liquid supplements from manufacturers which retain relevant industry accreditation.***

PERFORMANCE INDICATORS:

1. Premix and liquid supplements are sourced from manufacturers which retain FeedSafe, FIAAA Code of Practice/FAMI-QS Certification, APVMA GMP or equivalent accreditation
2. All additive, premix and liquid supplements feedstuffs entering the feedlot are accompanied with a valid Stock Food Supplier Declaration Form or SAFEMEAT Commodity Vendor Declaration (CVD).

Figure 1. Standard required for sourcing feed additives, premixes and supplements from accredited manufacturers (NFAS, 2022)

Manual systems for formulation and batch control remain in use in both dry and liquid manufacturers. For dry supplement manufacturers, 50% relied on manual systems for formulation, whereas all the liquid supplement manufacturers were using formulation software. Manual batching systems remained present in both dry (25%) and liquid (50%) supplement manufacturers.

The survey results for QA procedures show all manufacturers have medications and Schedule 4 (S4) antimicrobial (antimicrobials that require veterinary prescription for use) procedures. The importance of risk management of medications is highlighted by 75% of the liquid supplement manufacturers avoiding use of S4 antimicrobials and half of all manufacturers investing in antimicrobial (including ionophores) free production line.

The sampling and testing of both ingredients (carriers, ionophores) and finished products was common practice for both dry and liquid supplement manufacturers. Common carriers include molasses for liquid supplements and cereal grain or grain by-products such as millrun, protein meal or limestone for dry supplements. These ingredients are tested for nutrient content and presence of chemical residues and heavy metals. Carriers often contribute the highest inclusion ingredient within the supplement. The consistency of carriers enhances integrity of the supplement, preventing separation or sorting, reacting with other ingredients and providing an efficient method of distributing ingredients and related nutrients evenly throughout the mixed ration.

Mixer flushing procedures to avoid cross contamination between product lines and reduce accumulation of medications, antimicrobials and ionophores within the mixer were undertaken by all manufactures. Mixer function was also assessed by all manufacturers by undertaking mixing testing and assessment by co-efficient of variation (indicates level of variation around the average value). The dry manufacturers favoured a 5% co-efficient of variation target, whereas the liquid supplement manufactures used 10%.

4.1.2 Supplement manufacture recommendations

All supplement manufactures surveyed provided recommendations for formulation (ingredient and nutrient specifications) and management of supplement once delivered as described in Table 3. The recommendations for formulation relate directly to integrity and consistency of supplement during transport and storage at the feedlot and account for the manufacturer infrastructure and manufacturing process.

All liquid supplement manufactures recommended supplement (blended or suspension) be stored in a cone bottom tank. Cone bottom tanks reduce the likelihood of accumulation, particularly if separation of components occurs and allows the contents of the tank to fully empty as discharge is at the lowest point. Only one of the four liquid supplement manufactures recommended the use of aeration or re-circulation prior to supplement use.

All liquid supplement manufacturers recommended the use of suspending agents for both blend and suspension products.

Particle size of limestone, magnesium carbonate and monensin were identified by all liquid supplement manufacturers as critical to integrity and safety of supplement. All manufactures specified that monensin would not be accepted in granular form. However, only 50% of the liquid supplement manufacturers physically assesses particle size of monensin, requiring particles less than 75µM. In contrast, all manufacturers assessed particle size of limestone and magnesium carbonate. Preferred limestone particle size, no greater than 75µM. Preferred magnesium carbonate particle was 40 to 45µM. Smaller particles have greater surface area, promoting binding capacity and reduced risk of separation, particularly of insoluble ingredients such as limestone (0.0013g/100g) and magnesium carbonate (0.06g/100g).

Table 3. Summary of supplement manufacture survey recommendations responses (expressed as percentage) with formulation specifications provided as minimum (MIN) or maximum (MAX) inclusion on a percentage basis for ingredient and nutrient

	Supplement Manufacture	
Recommendations	**Dry**	**Liquid**
Formulation	100	100
Storage period	100	100
Usage rate	50	100
Storage method	100	100
Ingredient Formulation Specifications		
Ingredient & nutrient specifications provided	100	100
	Meal[1] / Pellet[2]	**Blend[3] / Suspension[4]**
Dry matter %	NA	60-75 / 65-70
MIN % Carrier[5]	20 / 50	20-35
MAX % Limestone	10 / 15	NA / 30
MAX % Urea	20 / 10	10-30
MIN % Vegetable oil	1-3	NA
MIN % Protein meal	10 / 15	NA
MAX % Magnesium carbonate	NA	3-5
MAX % Combined salts	NA	15-35 / 20-40

[1]A meal is a blended combination of dry ingredients of similar particle size (cereal grain is rolled or hammer milled) using vegetable oil to suppress dust and reduce separation. [2]A pellet is dry ingredients combined into a pellet using a pellet press. [3]A blend is a molasses based liquid supplement without limestone. [4]A suspension is a molasses based liquid supplement with limestone. [5]Carrier refers to cereal grain, grain by-products or protein meal for dry supplements and molasses for liquid supplements

4.1.3 Use of ionophores or antimicrobials

The dry supplement manufacturers had many more ionophore and antimicrobial products on hand compared to liquid supplement manufacturers as shown in Table 4. Dry supplement manufacturers make feed for multiple species (agricultural and companion), requiring a wider range of products on hand. In contrast, the liquid supplement manufacturers are concerned with ruminants, of which beef cattle feedlot and pasture supplements dominate.

Despite the availability of more than 30 registered monensin products, both dry and liquid supplement manufacturers had only between one and four products on site (Table 4). The limited number of monensin products in use is a function of logistics and application. Limiting the number of additives on hand promotes turnover, reducing wastage and risk of products reaching expiry dates as well as simplifying inventory control. In terms of application, products which vary in concentration provide flexibility for the dry supplement manufacturers, whereas for the liquid supplement manufacturers, particle size is a critical requirement to ensure supplement stability.

Table 4. Ionophore and antimicrobial products held at manufacturing site

	Supplement Manufacture	
Ionophores / Antimicrobials	**Dry**	**Liquid**
Number used	20 to 30	4
Number of monensin products	1 to 3	2 to 4

4.1.4 Adverse experience

Dry and liquid supplement manufacturers have all had some type of adverse experience during production, transport as well as at the feedlot, as described in Table 5.

During production, the dry supplement manufacturers reported physical issues including bridging in out-loading silos and pellets heating and sweating releasing ammonia and losing durability. The dry supplement manufacturers also reported mineral reactions compromising supplement integrity from heating (loss of fat soluble vitamins), binding and volatilization. No adverse effects occurred during transport of dry supplements. This is likely a function that supplements are dry, any issue with product integrity will occur during manufacture and delivery occurs using specific mill transport or product is delivered in bags (20kg or 1t bulk bags), risk of contamination is minimised. Adverse experiences on site were mainly related to sorting of the supplement for the ration. Reasons for sorting include palatability, pellet durability, volatilisation and ammonia emissions and variation in particle size and density (feeding action results in separation).

For liquid supplement manufacturers, adverse experiences during production were related to transfer errors and pH reactions. Adverse experiences during transport included direct contamination or chemical reactions caused by transport tanker holding residue from a previous delivery. At the feedlot, adverse effects were caused by limestone and monensin particle size causing separation, errors in ingredient concentration (formulation entry) and additive loading errors. Delivering supplement to the wrong tank was a common theme experienced by all manufactures and not necessarily the fault of the supplier. Feedlots may have multiple tanks containing not only a single feedlot supplement but storing multiple supplements, pasture supplements and/or molasses. Other effects related to the residue retained in storage tanks, particularly flat bottom tanks but also cone bottom tanks that have never been fully emptied or cleaned out.

Table 5. Summary of supplement manufactures adverse experiences responses occurring during production, transport and at feedlot (expressed as a percentage)

	Supplement Manufacture	
Location of adverse experience	**Dry**	**Liquid**
Production	75	75
Transport	0	75
Feedlot	50	100

4.2 Feedlot survey

The following frequency tables summarise the feedlot supplement survey results. Results reported are on 28 of the 30 feedlots surveyed using liquid supplements. Liquid supplements were provided by all four available manufacturers. Feedlot supplement survey results are provided in the following tables covering infrastructure (Table 6), feedlot liquid supplement storage, daily usage and turnover period (Table 7), summary of veterinary feed additive use (Table 8) and assessment of feedlot liquid supplement composition (Table 9).

4.2.1 Infrastructure for receival, storage & transfer of feedlot liquid supplements

Of the 28 feedlots surveyed, 93% were using suspension supplements (limestone included). Although limestone can be sourced relatively easily and cheaply when delivered in bulk, the preference to have limestone as a component of the supplement improves loading accuracy as limestone is very dense (bulk density of 2.71t/m^3) and difficult to load accurately to low inclusion levels of less (1.2 to 1.5%). Inclusion of limestone in the supplement also simplifies ingredient inventory control as it is one less ingredient for concern.

Supplement inclusion level in the finisher ration averaged 5.1% $\pm$0.69 SD. The lowest inclusion level reported was 3.5% and the highest 8.0%. Inclusion level of mineral supplements should be sufficient to achieve consistent distribution throughout the entire ration, creating a homogonous mix. The recommended minimum inclusion of mineral supplements within a ration is 5% on a dry matter basis. Minimum inclusion levels can vary depending on supplement manufacturing limitations, such as daily turn-over requirements, mixing equipment operation and variation in ration ingredient characteristics (particle size, density, moisture) (Table 3.). Ultimately, the inclusion level needs to create a homogenous mix, assessed objectively by calculating a co-efficient of variation (CV). Target CV levels for feedlot rations are 10 to 15% (Herman and Behnke, 1994).

The summary of infrastructure survey results is described in Table 6. Preference for cone bottom tank storage (93%) aligns with feedlots using suspension supplements. Cone bottom tanks are the recommended method for storing any form of liquid supplement as discussed in section 1.1.2.

Mono or helical pumps are the preferred pump type (70%), although gear pumps remain common.

Mono or helical pumps belong to the positive displacement group of pumps. They use a single helix rotor within a stator to create a continuous cavity which is moved to the discharge end of the pump. These types of pumps work well with high viscosity and dense liquid supplements. They provide smooth linear and measurable flow rates and can increase flow rate as viscosity increases. Gear pumps have been a popular method of transferring high viscosity liquids such as molasses. Gear pumps use rotating cogs or gears to develop a seal, creating suction at the inlet and enclosing the liquid within cavities of the rotating gears transferring towards discharge. Gear pumps have higher maintenance cost as gears wear more rapidly due to corrosion caused by limestone and other insoluble components contained within the supplement.

The preferred method of loading is direct from the storage tank to the mixer (74%). Of the large and very large feedlots surveyed (8 responded), 75% were using either batch box or liquid batching systems. These systems reduce time required for loading as required quantities of liquids are stored and ready for discharge as well as improving inventory control.

Although liquid supplements often represent the lowest inclusion component of the ration, they are also the most concentrated and costly of the ingredients. Despite this, 30% of feedlots surveyed transferred liquids through a shared delivery line. Other liquid ingredients used by feedlots include water, molasses, a second supplement, vegetable oil and/or glycerol. A shared delivery line can result in dilution, reduced loading accuracy of small load size and reduce inventory control as a quantity of any liquid transferred will remain in the delivery line.

Of the feedlots surveyed, 78% preferred to use a single supplement and 93% surveyed adjusted the inclusion rate of an individual supplement to suit different rations. Within the large and very large feedlots, 38% preferred to use a single supplement with only one feedlot using three supplements. A single supplement improves inventory control, ease of ordering and increases storage capacity. Multiple supplements may be a market requirement (ionophore free, Wagyu) or a specific supplement to promote health and feed intake (starter supplement).

Of the feedlots surveyed, 78% had two or more storage tanks. Of the 15 small feedlots, 60% had two storage tanks. Additional storage tanks provide feedlot ingredient security in case of delivery disruption (flooding) and reduces freight cost as larger loads can be received. The emphasis of reducing freight cost shown by 78% of feedlots have supplements delivered by B-double. In most cases (71%), delivery tankers had the sole purpose of delivering liquid supplements. Dual purpose tankers carried related liquid products such as molasses-based pasture supplement and/or raw molasses. Single function tankers reduce risk of contamination and adverse reactions associated with mixing products differing in pH.

Of the feedlots surveyed, only 18% of the liquid supplement storage tanks were set up for mixing (aeration or re-circulation), however none of these feedlots were using mixing systems. Of the 7% of feedlots that had considered mixing, no feedlot was mixing the supplement within the storage tank at the time of survey. Distances with which liquid supplements were transported from manufacturing site to feedlot varied. The average distance was 350km ±233SD, with a minimum of 30km and a maximum of 985km.

Table 6. Summary of infrastructure used by feedlots to receive, store and transfer liquid supplements (expressed as a percentage)

	Cone	Flat bottom		
Type of tank	93	7		
	Mono / Helical	Gear		
Type of pump	70	30		
	Direct to Mixer	Batch Box	Liquid Batching	
Loading method	74	11	15	
	Poly	PVC	Steel	
Plumb material	74	15	11	
	Separate	Shared		
Delivery lines	70	30		
	Single	Two or more		
Supplements in use	78	22		
	Changed for Ration	Same rate for all rations		
Inclusion level	93	7		
	1	2	3	4 or more
Tank number	22	56	7	15
	Tank set up		Feedlots considered mixing	
Storage tank mixing	18		7	
	Single	B-double	B-triple or Road train	
Delivery tanker	7	78	15	
	Specific	Dual		
Tanker function	71	29		

4.2.2 Feedlot liquid supplement storage & turnover

The diversity of feedlot operations surveyed highlight the storage capacity and turnover range described in Table 7. Total storage capacity shows a reliance on greater delivered quantities, with minimum storage capacity surveyed able to receive a B-double delivery (38 tonnes).

Table 7. Assessment of feedlot liquid supplement storage, daily usage and turnover period

	Mean	Minimum	Maximum	SD
Total storage capacity (t)	102	38	320	61.77
Daily usage (t/day)	6.5	0.8	38	8.21
Turnover for load (day)	16	2	48	12.09
Turnover for storage (day)	30	5	125	25.55

Daily usage ranged from 38t/day down to 0.8t/day with a mean of 6.5t/day. In turn, these usage levels influenced turnover period (days). The maximum number of days required to turnover the load (48 days) was less than the maximum recommended storage period for suspension supplements of 3 months or 90 days. This shows feedlots are aware of the importance of regular supplement turnover. However, the minimum turnover periods also highlight that some feedlots have less than a week total storage capacity, requiring additional management of supplements during supply interruption.

4.2.3 Feedlot liquid supplement stock-on hand management

Stock on hand is the quantity of ingredients held at the feedlot. Stock on hand is determined by undertaking a stocktake. Stocktakes generally occur monthly, although some higher turnover ingredients, such as liquid supplements may be reviewed more frequently. Stock on hand is used to identify ingredient shrink or loss and assist with inventory control (quantity of ingredient on hand and purchased compared to quantity required).

Of the feedlots surveyed, 50% completed a supplement stocktake and of those that completed a stocktake, 20% ran the stocktake more frequently than monthly (weekly or fortnightly). Although half the feedlots did not stocktake the supplement, 75% of feedlots assigned a shrink value. Shrink values averaged 0.8% $\pm$1.00SD, with a minimum of 0.2% and a maximum of 4.6%. The highest supplement shrink levels (2.0% and 4.6%) were from feedlots with shared liquid delivery lines.

The feedlots that did not undertake supplement stocktake used a pencil shrink of either 0.3 to 0.5%. Those feedlots that did stocktake the supplement had an average shrink of 1.1% $\pm$1.17SD, indicating the assigned shrink is not sufficient to cover actual supplement loss. Causes of supplement loss reported include loading error, loss (drips) from delivery system into mixer, leakage from worn pump seals and spills during unloading.

4.2.4 Additive use

The veterinary feed additives used by the feedlots surveyed are summarised in Table 8. Monensin was the dominant feed additive with 96% of feedlots using the additive and 54% using monensin as the only feed additive. 64% of feedlots preferred to rely on a single additive. The remaining 56% of feedlots used either barbermycin, lasalocid or tylosin, or a combination. Feed additives that were not used included salinomycin, narasin and oxytetracycline.

Table 8. Summary of veterinary feed additive use (expressed as a percentage)

	Feed Additive (active compound)						
	Monensin	Monensin Only	Barbermycin	Lasalocid	Tylosin	Virginiamycin	CTC[1]
Usage	96	54	14	14	4	4	21

[1]Chlorotetracycline

All feedlots sourced feed additives from licensed manufactures.

The S4 (requiring a veterinary script for use) feed additives included tylosin, virginiamycin and CTC. The S4 feed additives were used by 25% of the feedlots surveyed. In all cases, tylosin and virginiamycin were contained in the liquid supplement, whereas the CTC was directly added to the ration by the feedlot. All feedlots held and maintained veterinary script records for use of S4 additives, and these were provided with instructions for use and requirements for cleaning mixing equipment.

4.2.5 Feedlot liquid supplement composition

The composition of the liquid supplements used by feedlots surveyed are summarised in Table 9. Liquid supplement composition generally follows the recommendations provided by the manufacturers as described in Table 3, particularly for dry matter and maximum limestone inclusion.

The molasses inclusion in Table 9 describes levels for suspension supplement only. The two feedlots using a blended supplement contained a higher molasses level (72 and 77%) accounting for the lack of limestone and reduced water requirement to maintain desired supplement dry matter levels.

Bulk density averaged at 1.37kg/m^3. The bulk density of liquid supplement compared to other commonly used liquids such as water (1kg/m^3), molasses (1.2kg/m^3) and vegetable oil (0.9kg/m^3), highlights the value of using separate delivery lines to promote loading accuracy and ingredient inventory control.

Table 9. Composition of feedlot suspension supplement

	Mean	Minimum	Maximum	SD
Molasses inclusion (suspension) %	39.5	28.5	51.7	7.20
Supplement dry matter %	69.3	65.0	71.0	1.64
Supplement bulk density kg/m^3	1.37	1.20	1.45	0.07
Limestone inclusion %	25.4	12.0	30.0	4.03

4.2.6 Feedlot liquid supplement quality management

The QA applied to liquid supplements by the feedlot at receival, storage and during use was minimal.

Of the feedlots surveyed, 25% collected samples of supplement on delivery. 21% sent supplements for formal testing at a lab. All testing was done by Symbio Alliance. Of those that collected samples, 30% were collected from each load, 14% took samples monthly, 14% took samples annually and 43% took samples when there was a problem.

Of the feedlots surveyed, only 14% undertook a formal visual assessment of the liquid supplement and/or conducted testing onsite. Visual assessments included rate of loading, colour and aroma. Onsite testing involved assessing bulk density.

As described in Table 6, 78% of feedlots had two or more storage tanks and of the 15 small feedlots, 60% had two storage tanks. Despite availability of multiple tanks on site, only 11% of feedlots reported cleaning storage tanks. 39% ensured a tank emptied completely prior to re-filling. Annual cleaning and ensuring tanks empty completely between loads reduce the risk of extending the effects of a problem load over time.

Much of the emphasis for supplement quality and integrity was viewed as the responsibility of the manufacturer. Of the feedlots surveyed this assumption is supported by only 18% of feedlots reporting a supplement issue. Reported experiences were mainly concerned with limestone separation.

4.3 Baseline variation of supplement constituents

Of the 30 feedlots selected, 19 provided supplement samples. These 19 feedlots were representative of the 30 originally selected in terms of size (VL, L, M, S), supplement type (suspension, blend) and type of tank (cone bottom, flat base). In terms of feedlot size, the representation of the 30 feedlots included 10% VL, 23% L, 20% M and 47% S. For the 19 feedlots, the representation included 16% VL, 21% L, 16% M and 47% S. The 19 feedlots had the same representation as the 30 feedlots for suspension supplement use and cone bottom tank storage, both 93%.

To assist with interpretation of supplement constituent analysis, Table 10 provides a summary of the participating feedlots size, infrastructure, supplement turnover rate, inclusion and storage tank management. These factors assist in providing a production basis for the analysed values.

Table 10. Feedlot infrastructure, supplement use and storage tank management

Feedlot ID	Size	Supp Type	Tank#	Tank Type	Turnover days	Inclusion %	Tank Emptied	Tank Cleaned	Lines Separate
1	S	LSUSP	1	Cone	4	8.0	NO	NO	YES
3	S	LSUSP	2	Cone	16	5.0	YES	YES	YES
5	S	LSUSP	2	Cone	21	5.0	YES	NO	YES
8	L	LSUSP	4	Cone	4	5.0	NO	NO	YES
10	L	LSUSP	1	Cone	4	5.2	NO	NO	NO
11	VL	LSUSP	4	Cone	7	5.2	NO	NO	YES
12	L	LSUSP	2	Cone	2	5.1	YES	NO	YES
13	S	LBLD	1	Flat	40	4.5	NO	NO	NO
16	S	LSUSP	2	Cone	14	5.0	YES	NO	YES
17	M	LSUSP	3	Cone	7	5.0	YES	NO	NO
18	M	LSUSP	2	Cone	6	5.0	YES	NO	YES
19	M	LBLD	2	Flat	10	5.0	NO	NO	YES
21	L	LSUSP	2	Cone	9	5.4	NO	YES	NO
22	VL	LSUSP	4	Cone	4	5.0	YES	NO	YES
23	S	LSUSP	1	Cone	40	5.0	NO	NO	YES
24	S	LSUSP	2	Cone	18	5.0	NO	NO	NO
26	VL	LSUSP	6	Cone	3	5.0	YES	NO	YES
27	S	LSUSP	2	Cone	27	5.0	NO	NO	NO
28	S	LSUSP	1	Cone	32	5.0	NO	NO	NO

The results of all the feedlot sampled are summarised using tables. Results of individual feedlots are described in figures to highlight changes in nutrient value as the tank emptied over time. The figures describe the range of values from the delivery truck in blocks and lines describe the minimum, average and maximum values of the nutrient at each sampling period.

The five duplicate samples (10 samples in total) collected from the feedlot's storage tanks were summarised by calculating the mean, standard deviation and co-efficient of variation. The difference between each duplicate sample, including the sample taken from the delivery tanker was used to calculate average laboratory error. Laboratory error can be reviewed as samples were blinded.

An average absolute deviation with error percentage (Absolute Dev with Error%) was calculated to identify the variation between the Lab Error and the mean delivery nutrient level. The purpose of this calculation was to determine if the differences between the samples and the delivered mean were actual, in-field or due to Lab Error.

An absolute deviation percentage (Absolute Dev%) was calculated to define the differences between the average storage tank samples and the delivered sample mean. This was calculated firstly determining the average delivered nutrient level from the duplicate samples, then the absolute deviation was calculated from the delivered nutrient level at each storage tank level by averaging the nutrient level from each duplicate sample taken from the storage tank and then the variance of each average storage tank result the delivered average was calculated. The absolute deviation percentage was calculated using the variance percentage at each level in the storage tank.

Within individual feedlot analysis, methodology errors or outliers are apparent. The result tables provided have these outliers removed. These result summaries were conducted for each individual analysed component (monensin, moisture, ash, calcium, zinc, copper, potassium) described in the following tables.

4.3.1 Monensin

The monensin levels across the 19 feedlots were generally consistent, achieving close to or below the 10% coefficient of variation (CV%) target. Feedlots 5 and 8 had higher CV values of 58.2% and 19.5% respectively and absolute variation % of 85.4 and 53.2% respectively. With none of the feedlots undertaking supplement mixing, the use of the small particle size monensin was sufficient to prevent separation.

Section 4.1.2 described the supplement manufacturer's management of monensin products with half physically assessing particle size with the requirement that 90% of particles are 75µM or less. The other manufactures ensure use of only feed tech grade non-granular monensin. In contrast, the larger particle size with cereal (corn) carrier (+800µM) monensin used in the USA provides specific mixing recommendations prior to each use as described in section 4.3.

Table 11. Monensin baseline variation of liquid supplement samples

	Monensin (mg/kg DM)					
Feedlot ID	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
1	259.0	25.29	9.8	25.8	7.9	10.1
3	503.0	15.87	3.2	39.8	8.4	5.9
5	7.2	4.19	58.2	7.7	17.0	85.4
8	314.3	61.40	19.5	23.0	8.1	53.2
10	297.9	22.50	7.6	20.2	7.9	4.8
11	459.7	33.87	7.4	53.7	10.4	3.7
12	399.0	18.49	4.6	32.1	7.3	7.7
13	538.8	53.13	9.9	35.2	6.9	7.0
16	387.4	44.82	11.6	41.8	12.4	7.2
17	547.1	17.98	3.3	23.7	3.3	2.7
18	666.1	44.51	6.7	52.5	6.2	3.9
19	543.4	33.47	6.2	28.8	3.3	4.8
21	382.5	28.85	7.5	39.4	12.9	16.2
22	494.1	29.15	5.9	26.4	5.3	3.3
23	581.6	38.73	6.7	65.9	11.3	3.8
24	517.6	27.19	5.3	24.7	5.0	3.1
26	527.7	43.85	8.3	71.0	12.9	3.1
27	538.2	28.45	5.3	31.5	6.4	12.2
28	564.6	28.53	5.1	21.7	4.6	3.8

The results from feedlot 5 indicate that current flushing procedures in place for delivery tankers need reviewing by supplement manufacturers. Feedlot 5 should contain no monensin (uses a different ionophore). However, monensin was identified within the supplement. The highest levels, 51 and 47mg/kg DM for duplicate 1 and 2 samples respectively were derived from the delivery truck as described in Figure 3. Monensin levels fell once supplement was transferred to the storage tank and during use as described in Figure. 2. The reduction in monensin due to dilution with residual supplement remaining in the tank.

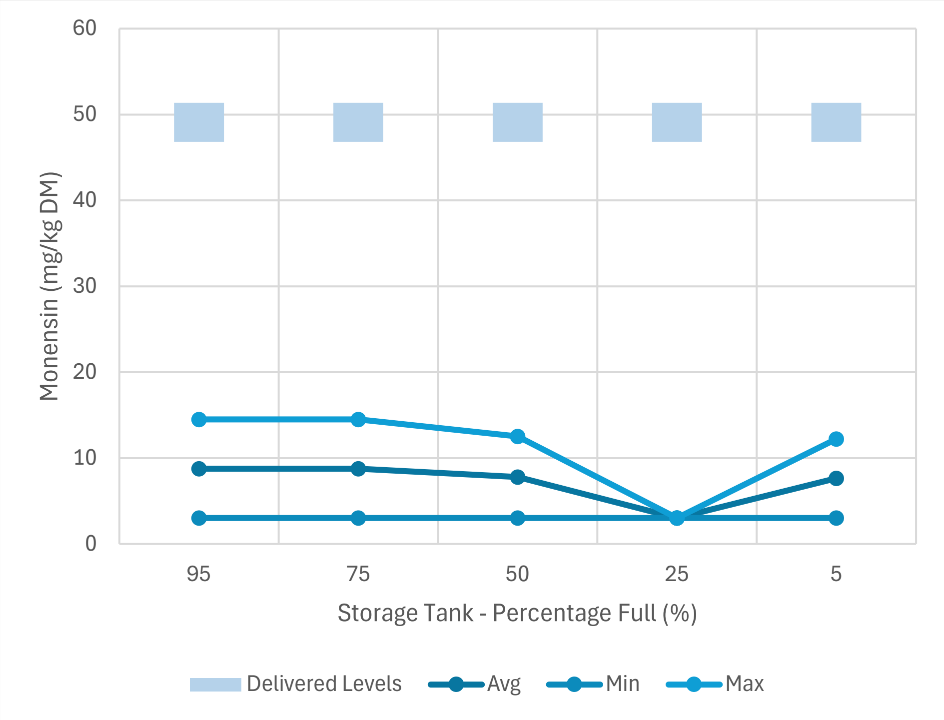


Figure 2. Feedlot 5 monensin concentration (mg/kg DM) at delivery and at storage tank sampling capacities

This is particularly relevant as an adverse experience reported by a supplement manufactures in section 4.1.4 involved contamination and/or chemical reaction within a supplement due to delivery tanker holding residue supplement from a previous delivery. In addition, supply of non-ionophore containing supplement could be compromised.

The results from feedlot 8 show an increase in monensin concentration compared to delivery and 95% tank capacity level as shown in Figure. 3. Although feedlot 8 has 4 storage tanks and only a 4 day load turnover, it not only does not empty the tanks before been refilled, but all the tanks are open and thereby contents of all tanks are affecting nutrient concentration. The increase in monensin concentration is not due to separation as levels did not continue to increase but potentially derived from residue accumulating in other tanks and been drawn out during transfer.

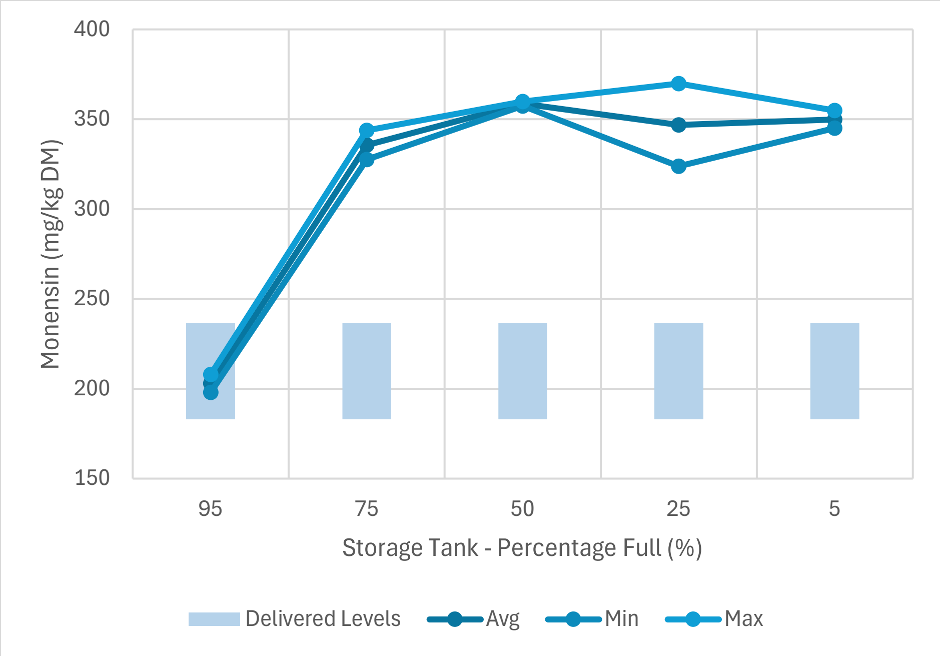


Figure 3. Feedlot 8 monensin concentration (mg/kg DM) at delivery and at storage tank sampling capacities

Rate at which supplement is turned over increases consistency of monensin level as the tank empties as described in Figure. 4. Feedlots with 18 days or less to turn over tank had monensin concentrations from the tank similar to the concentration from the delivery tank. This was the case irrespective of whether the feedlot emptied the storage tank completely or not or used separate or shared delivery lines.

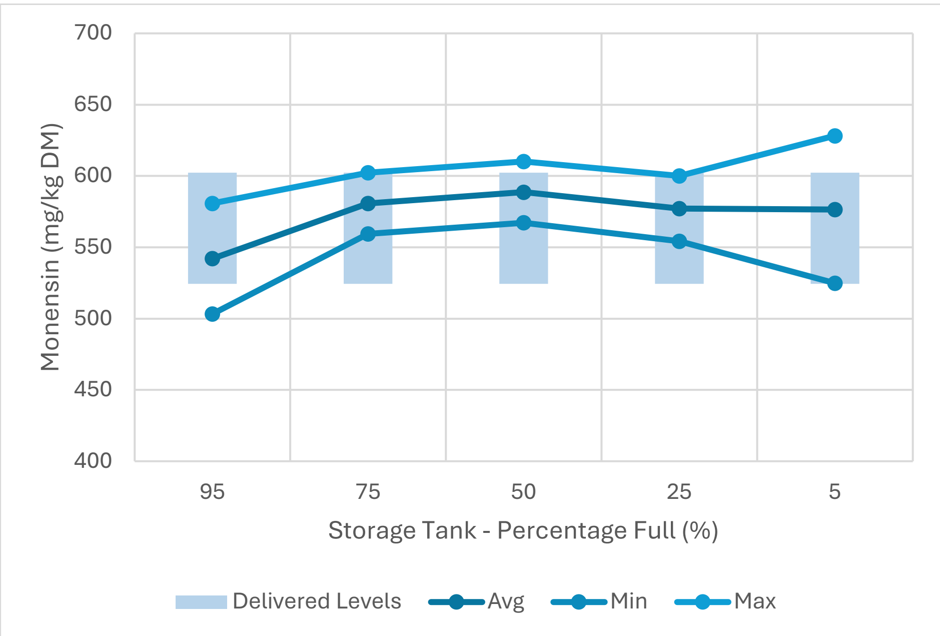


Figure 4. Monensin (mg/kg DM) concentration of feedlots with 18 days or less turnover supplement

Feedlots which required 27 days or greater to empty tank observed a lower monensin concentration on re-filling as described by Figure 5, suggesting a dilution of supplement residue which may remain in tank. This potentially leads to increasing concentration of monensin as the tank empties. Changes in monensin concentration may not be large enough to cause an animal response (<50mg/kg DM) however, feedlots with single tanks which don't empty out and have extended turnover may benefit from re-circulation.

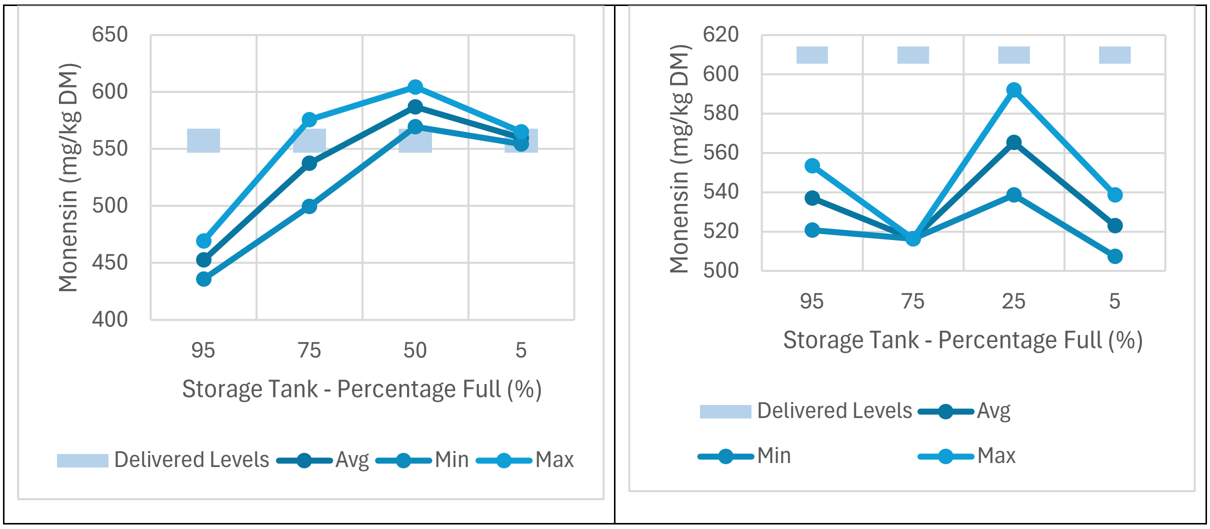


Figure 5. Monensin (mg/kg DM) concentration of feedlots (13 and 27) with 27 days or more to turnover supplement

4.3.2 Moisture

The moisture levels across the 19 feedlots were also consistent with all feedlots close to or below the 10% CV target as described in Table 12. All liquid supplements require the addition of water during manufacture to achieve desired dry matter targets provided by the manufacturers as described in Table3. Recommended dry matter range for blended supplements is 65 to 75% and for suspension supplements 65 to 70%. The mean moisture levels averaged 31.1% $\underline{+}$2.32SD.

Feedlots 8 and 22 had higher absolute deviations as shown in Table 12. In both cases, it appears the greater variation is caused by sample preparation error. For feedlot 8 the Delivery and 95% tank samples of duplicate 1 were 17.3 and 16.6% moisture respectively compared to duplicate 2 with both 22.1% for the same samples. These values appear unrealistic when compared with the preferred supplement target dry matter values provided in Table 3.

For feedlot 22 the higher absolute variation % including laboratory error was 22.8%. This was due obvious differences in moisture values between the samples of duplicate 1 and 2. For duplicate 1, the supplement moisture averaged 31.0% and for duplicate 2, the supplement moisture averaged 24.7%. The duplicate samples from feedlot 22 were sent for analysis on different dates. Duplicate 1 was sent on 25th March 2024 and duplicate 2 on 4th April 2024. When the Lab Error was removed, the absolute deviation level reduced to 2.8%.

Table 12. Moisture baseline variation of liquid supplement samples

				Moisture %		
Feedlot ID	Mean	SD	CV%	Lab Error Ave Ave	Absolute Deviation% with error	Absolute Deviation%
1	32.9	0.62	1.9	0.7	2.3	2.6
3	31.0	0.54	1.7	1.0	3.1	0.9
5	35.0	0.66	1.9	0.8	2.3	3.5
8	26.5	2.51	9.5	3.1	14.2	28.1
10	33.3	0.43	1.3	0.4	1.3	1.0
11	29.8	1.50	5.0	2.5	8.8	1.3
12	27.6	0.86	3.1	3.6	13.5	11.4
13	32.0	3.98	12.4	0.6	1.9	9.7
16	29.6	3.03	10.2	2.3	7.9	7.5
17	31.8	1.43	4.5	1.8	4.8	2.3
18	33.5	2.99	8.9	4.6	14.1	1.8
19	31.6	0.63	2.0	0.9	2.3	1.3
21	30.2	3.25	10.8	2.8	9.3	6.2
22	27.9	1.77	6.3	6.3	22.8	2.8
23	32.3	1.96	6.1	3.8	10.5	3.0
24	33.8	1.84	5.4	2.6	8.9	6.4
26	29.2	0.98	3.3	1.6	5.6	2.7
27	32.8	0.58	1.8	0.5	2.1	0.9
28	29.6	0.67	2.3	0.7	1.5	1.6

4.3.3 Ash

The ash concentrations across the 19 feedlots were consistent with all feedlots close to or below the 10% CV target as described in Table 13. Ash describes the inorganic or mineral component of a feedstuff. It is calculated by completely combusting all organic matter in a furnace at 700°C. The weight difference of the cooled sample is the ash content.

Feedlots worth investigating further include 8, 13 and 18. Feedlot 8, again had discrepancies between delivered duplicate samples. Duplicate 1 had an ash concentration of 75g/100g DM compared to duplicate 2 with 59g/100g DM. The discrepancy is unlikely caused by a difference in sample preparation as both duplicate samples were analysed on the same day. The consistency of this variation between the delivered samples could indicate mis-labelling of the samples at the feedlot.

Feedlot 13 with a coefficient of variation of 10.5% exhibited a decrease in ash content at 95% tank capacity followed by an increase in ash content with each subsequent sample as described in Figure. 6. Feedlot 13 supplement storage is two flat bottom tanks that are not emptied or cleaned and require 40 day turnover per load. Supplement deliveries are pumped into the base of tanks, causing displacement of residue supplement remaining in the bottom of the tank. If the remaining residue has a lower concentration compared to the delivery, then the initial supplement transferred from the tank will be diluted, reducing concentration fed.

This is a risk of using flat bottom tanks, although cheaper to install, they fail to empty evenly due to larger surface area. Supplement closer to outlet is removed first, with supplement on the far side remaining. A flat bottom tank can also never empty completely as the outlet is an approximately

150mm higher than the base of the tank. Supplement remaining below this point will remain in the tank. Industry is aware of the disadvantage of using flat bottom tanks. Of the feedlots surveyed, 93% were storing supplement in a cone bottom tank.

Table 13. Ash baseline variation of liquid supplement samples

			Ash g/100g DM			
Feedlot ID	Mean	SD	CV%	Lab Error Ave	Abs Dev with error%	Absolute Deviation%
1	39.7	0.73	1.8	0.9	2.4	0.9
3	13.3	1.02	7.7	1.9	12.4	9.4
5	23.4	0.81	3.4	2.9	7.8	15.8
8	46.9	6.20	13.2	4.6	3.2	28.2
10	49.2	1.09	2.2	1.1	2.8	1.0
11	48.8	1.44	3.0	1.8	3.8	1.7
12	49.7	1.49	3.0	1.6	3.6	4.5
13	8.4	0.89	10.5	0.3	2.6	8.5
16	46.9	2.14	4.6	1.4	2.9	3.4
17	18.5	1.79	9.7	2.1	12.6	10.1
18	23.8	4.16	17.5	7.2	30.6	2.4
19	14.5	0.99	6.8	0.4	3.2	5.0
21	53.8	1.45	2.7	1.9	4.4	8.4
22	20.1	1.86	9.3	4.3	21.5	7.1
23	21.6	1.33	6.1	1.8	8.1	3.7
24	23.3	2.37	10.2	2.7	14.7	9.2
26	49.6	0.90	1.8	0.9	1.9	1.4
27	22.8	1.49	6.5	2.9	12.5	7.0
28	19.4	0.67	2.3	0.5	1.6	2.7

Feedlot 18 had a coefficient of variation of 17.5% and an absolute variation % including laboratory error of 30.6%. This was due again to differences in ash concentrations between the samples of duplicate 1 and 2. For duplicate 1, the supplement ash concentration averaged 27.4% and for duplicate 2, the supplement ash concentration averaged 20.3%. The duplicate samples from feedlot 18 were sent for analysis on different dates. Duplicate 1 was sent on 25th March 2024 and duplicate 2 on 4th April 2024. When the Lab Error was removed, the absolute deviation level reduced to 2.4%.

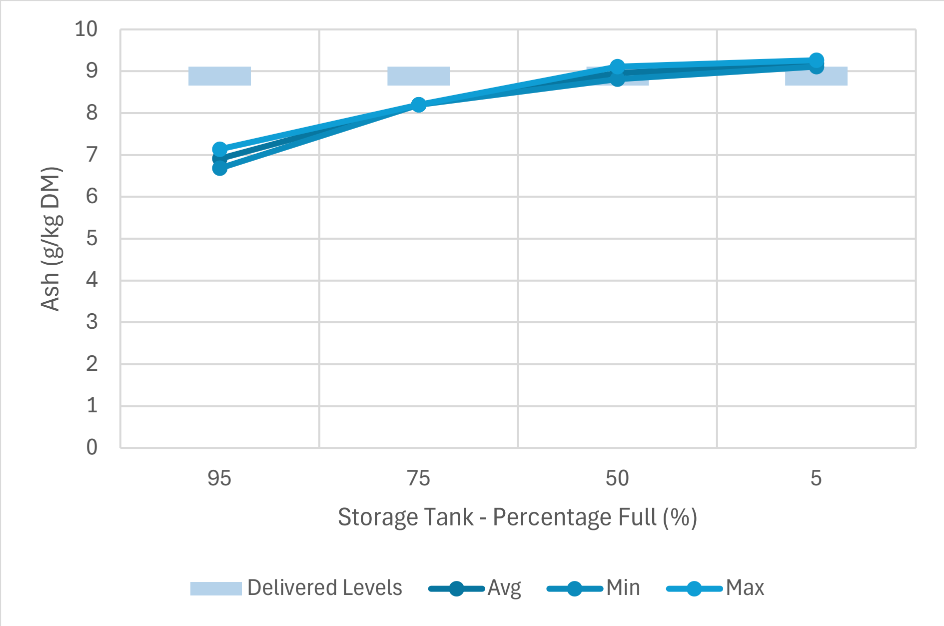


Figure 6. Feedlot 13 ash concentration (g/kg DM) at delivery and at storage tank sampling capacities

4.3.4 Zinc

The zinc concentrations across the 19 feedlots were very consistent with majority of feedlots below the 10% CV target and some under 5% as described in Table 14. Zinc concentration varied across the 19 feedlots with a range from 435.6mg/kg DM to 2756.1mg/kg DM. The higher zinc levels are due to use of additional organic zinc source inclusion to the inorganic zinc (zinc oxide or zinc sulphate) contained in the trace mineral premix.

The feedlots which did not achieve the target coefficient of variation (CV%) included 1, 10, 11 and 21. These were all examples of sample preparation error between duplicate 1 and 2 analysis as defined by the laboratory average error value described in Table 14. Feedlots 1, 10, 11 and 21 had zinc concentration error values of 312.3, 618.8, 477.3 and 666.7mg/kg DM respectively. The likely cause for the difference is failure to homogenise the sample prior to analysis.

Feedlots 10, 11 and 21 had the duplicates analysed on different days. In all cases, duplicate 1 samples were analysed on the 16th November 2023 and duplicate 2 samples analysed on the 23rd November 2023. Of concern is feedlot 10, which had both duplicates analysed on the same day of the 23rd November 2023, but for whatever reason a different preparation method was employed.

These results highlight the importance and value of always sending duplicate samples to any laboratory. Sending blind samples enables the feedlot to determine if differences are due to feedlot operation or laboratory error.

Table 14. Zinc baseline variation of liquid supplement samples

			Zinc (mg/kg)			
Feedlot ID	Mean	SD	CV%	Lab Error Ave	Abs Dev with error%	Absolute Deviation%
1	1017.2	84.99	8.4	312.3	30.3	5.2
3	827.5	41.67	5.0	71.6	9.9	6.3
5	798.0	42.84	5.4	56.4	7.4	3.2
8	1496.9	78.96	5.3	83.0	5.1	11.8
10	2576.3	320.10	12.4	618.8	22.9	1.9
11	1624.0	250.72	15.4	477.3	29.4	1.4
12	1166.7	32.36	2.8	60.7	4.9	3.6
13	882.0	80.54	9.1	42.5	5.6	6.8
16	1889.7	131.18	6.9	108.0	6.4	4.6
17	435.6	27.37	6.3	36.2	9.0	2.8
18	1307.3	72.76	5.6	96.2	6.9	2.6
19	445.6	12.41	2.8	16.5	3.6	1.3
21	2262.8	361.21	16.0	666.7	29.0	2.9
22	803.2	42.00	5.2	61.8	8.0	17.5
23	876.1	48.17	5.5	79.4	7.5	3.0
24	952.9	54.47	5.7	62.1	6.9	7.7
26	2756.1	47.42	1.7	53.6	2.2	3.1
27	1419.9	64.22	4.5	82.7	4.7	3.8
28	758.0	31.13	4.3	36.6	5.1	3.5

4.3.5 Calcium

The calcium concentration across the 19 sampled feedlots were variable as described in Table 15. Only five feedlots, 1, 8, 10, 23 and 26 had coefficient of variations less than 10%. The main cause for variation was Lab Error (difference between duplicate concentrations). This was the case for feedlots 11, 12, 17, 18, 21, 22, 27 and 28. Feedlots 1 and 3 contained outlier values and feedlot 5 contained Lab Error and outlier values.

An example of Lab Error is described in Figure. 7. for feedlot 17 compared with Figure. 8. describing feedlot 10 with values in line with delivered concentration levels.

The level of Lab Error may be elevated with calcium due to its insolubility requiring greater and more regular homogenising during the analysis process. As a broader consideration of this variation, calcium may not provide a good indicator for assessing mixing efficiency.

Table 15. Calcium baseline variation of liquid supplement samples

	Calcium (g/kg DM)					
Feedlot ID	Mean	SD	CV%	Lab Error Ave	Abs Dev with error%	Absolute Deviation%
1	118.2	9.80	8.3	27.1	29.1	3.4
3	33.3	11.50	34.6	16.5	41.5	20.6
5	63.1	18.40	29.2	35.4	55.6	13.8
8	120.1	3.24	2.7	5.4	3.6	19.5
10	149.5	10.91	7.3	9.2	6.2	6.0
11	126.5	18.30	14.5	21.5	18.4	10.7
12	105.4	14.09	13.4	17.3	22.5	14.6
13	6.2	0.93	15.0	0.3	4.9	13.6
16	133.5	16.00	12.0	19.3	13.6	8.9
17	47.0	18.67	39.7	23.5	51.6	18.5
18	66.9	14.10	21.1	28.2	41.4	8.2
19	30.3	3.55	11.7	4.3	14.2	8.1
21	129.4	34.79	26.9	44.4	43.6	14.8
22	54.4	13.41	24.7	25.8	46.8	14.1
23	65.6	2.76	4.2	4.1	6.7	5.0
24	76.8	11.73	15.3	11.3	17.0	9.3
26	119.3	8.93	7.5	10.0	9.7	2.1
27	57.4	13.73	23.9	20.5	26.8	14.4
28	48.2	9.30	19.3	12.4	30.9	17.5

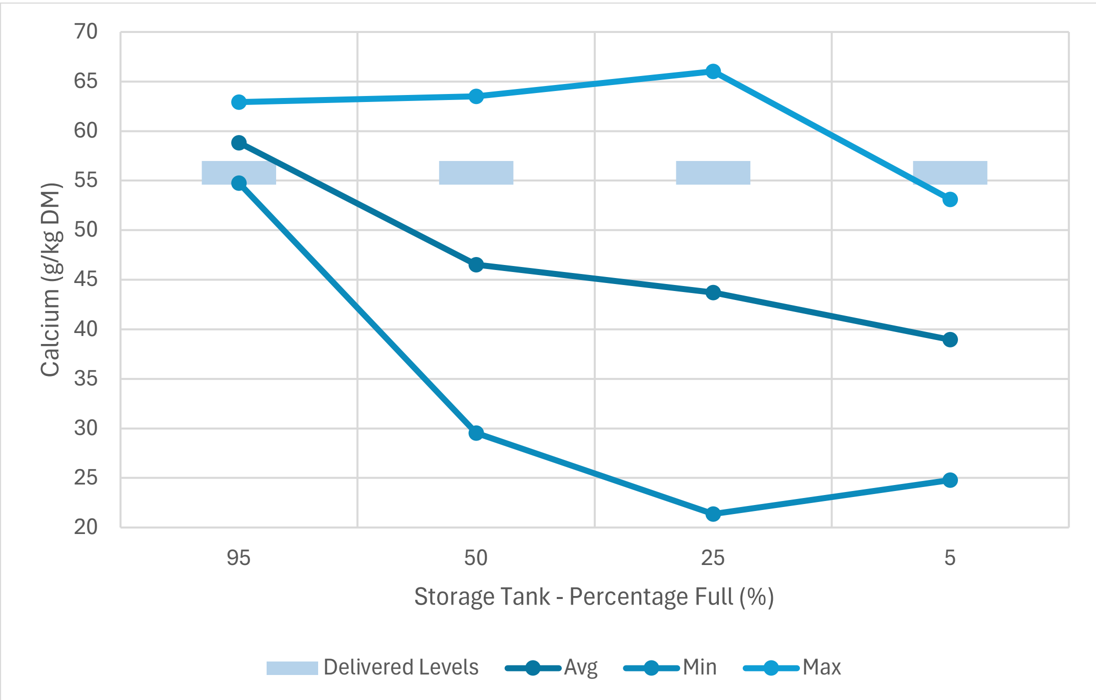


Figure 7. Feedlot 17 calcium concentration (g/kg DM) at delivery and at storage tank sampling capacities

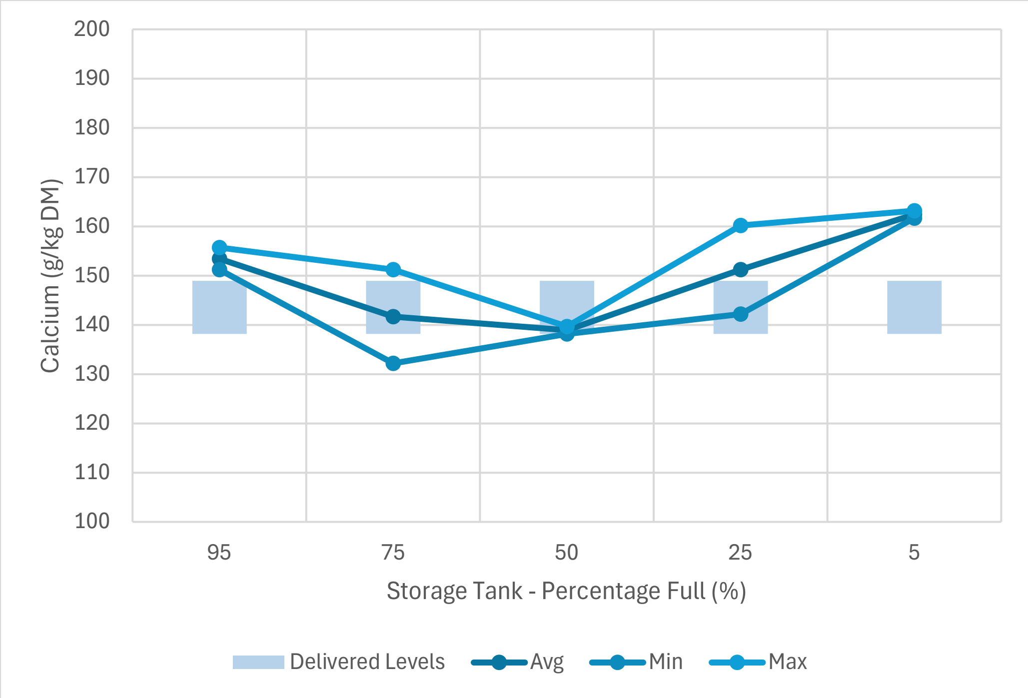


Figure 8. Feedlot 10 calcium concentration (g/kg DM) at delivery and at storage tank sampling capacities

4.3.6 Copper

The copper concentration across the 19 sampled feedlots is described in Table 16. Main reasons for variation in samples and failure to achieve target coefficient of variation levels due to outliers within the sample set. Examples include feedlots 1, 19, 22 and 27. Laboratory error caused variation in feedlots 5 and 13. Copper concentrations taken from the storage tank were similar to delivery concentrations as defined by absolute deviation.

4.3.7 Potassium

The potassium concentration across the 19 sampled feedlots is described in Table 17. Variation between delivered and storage tank concentrations were low, with 13 of the 19 feedlots achieving a coefficient of variation of less than 5% and 17 of the 19 feedlots reaching the target coefficient of variation of 10%. The consistency of potassium is related to molasses providing the carrier source. Molasses contains a natural potassium level of around 4% DM (40g/kg DM). As described in Table 3, the recommended minimum inclusion of molasses in blended and suspension supplements is 20 and 35% respectively. Within the sample group there were three feedlots which exhibited greater variation. Feedlot 12 was affected by laboratory error and feedlot 22 had an outlier value.

Feedlot 13 exhibited a decrease in potassium level compared to the delivered concentration at the 95% tank capacity. As the tank emptied, the concentration increased to levels similar to the delivered concentration as described in Figure. 8. Feedlot 13 relies on a flat bottom tank that never empties between deliveries. When a new delivery is transferred into the tank, it is diluted by the residue remaining, reducing the initial concentration. As the tank is used, the residue settles with potassium concentration levels increasing.

Table 16. Copper baseline variation of liquid supplement samples

	Copper (mg/kg DM)					
Feedlot ID	Mean	SD	CV%	Lab Error Ave	Abs Dev with error%	Absolute Deviation%
1	112.5	16.01	14.2	12.3	11.6	5.5
3	90.4	6.71	7.4	13.0	13.0	4.7
5	26.8	16.97	69.4	26.8	181.3	77.6
8	153.2	1.25	0.8	7.7	3.2	11.2
10	176.0	2.75	1.6	11.7	2.1	11.2
11	180.0	6.29	3.5	5.5	3.0	2.3
12	255.7	5.83	2.3	8.7	3.0	0.9
13	100.2	18.92	18.9	26.7	35.0	11.0
16	189.1	11.21	5.9	5.3	3.3	3.7
17	133.5	14.49	10.9	19.1	14.3	5.2
18	103.0	16.71	16.2	17.8	18.4	11.0
19	90.6	25.11	27.7	26.7	31.6	15.9
21	172.2	16.45	9.6	10.7	6.1	13.8
22	182.2	45.98	25.2	28.5	20.3	18.5
23	95.2	8.40	8.8	13.5	11.4	13.3
24	97.6	8.22	8.4	11.2	11.5	11.7
26	178.1	4.71	2.6	5.8	3.5	6.7
27	100.3	21.96	21.9	27.9	22.2	19.4
28	63.7	11.04	17.3	11.3	11.0	16.8

Table 17. Potassium baseline variation of liquid supplement samples

	Potassium (g/kg DM)					
Feedlot ID	Mean	SD	CV%	Lab Error Ave	Abs Dev with error%	Absolute Deviation%
1	25.4	0.92	3.6	0.9	4.2	3.6
3	11.2	0.33	2.9	0.5	4.9	2.9
5	10.8	0.31	2.9	0.6	4.7	3.1
8	24.7	1.66	6.7	1.0	4.9	14.4
10	24.0	0.65	2.7	0.8	3.7	2.1
11	20.6	0.85	4.1	1.3	7.4	2.9
12	18.7	2.55	13.7	2.7	16.8	7.1
13	16.6	1.87	11.3	1.3	6.9	9.1
16	20.1	1.31	6.5	1.2	6.7	6.4
17	9.7	0.32	3.3	0.5	4.6	2.4
18	18.1	1.15	6.4	1.4	7.1	3.9
19	11.1	0.27	2.5	0.2	1.8	2.8
21	43.5	1.94	4.5	2.5	5.7	2.0
22	6.5	0.22	3.4	0.3	5.2	12.8
23	22.7	0.53	2.3	1.0	2.6	1.5
24	11.6	0.16	1.4	0.2	1.6	5.2
26	25.3	1.37	5.4	2.2	8.9	1.4
27	25.4	0.75	3.0	0.9	3.3	1.7
28	17.3	0.41	2.4	0.7	4.4	0.5

Feedlot 9 potassium concentration had a similar pattern to monensin described in Figure. 3. Figure. 10. describes the effect of increase in potassium concentration when residue remaining in other tanks are combined with the tank been used.

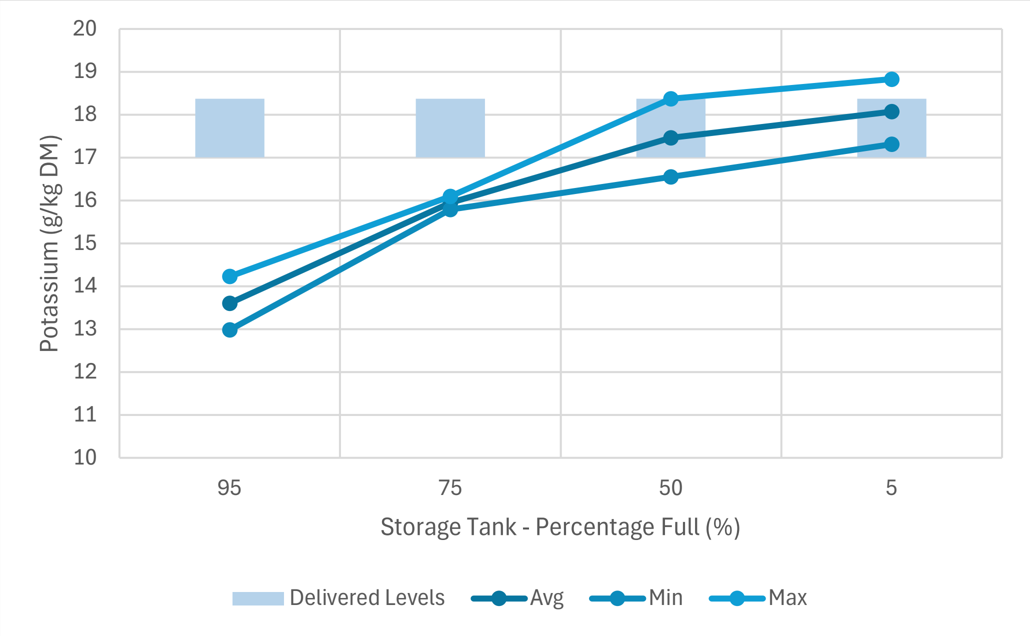


Figure 9. Feedlot 13 potassium concentration (g/kg DM) at delivery and at storage tank sampling capacities

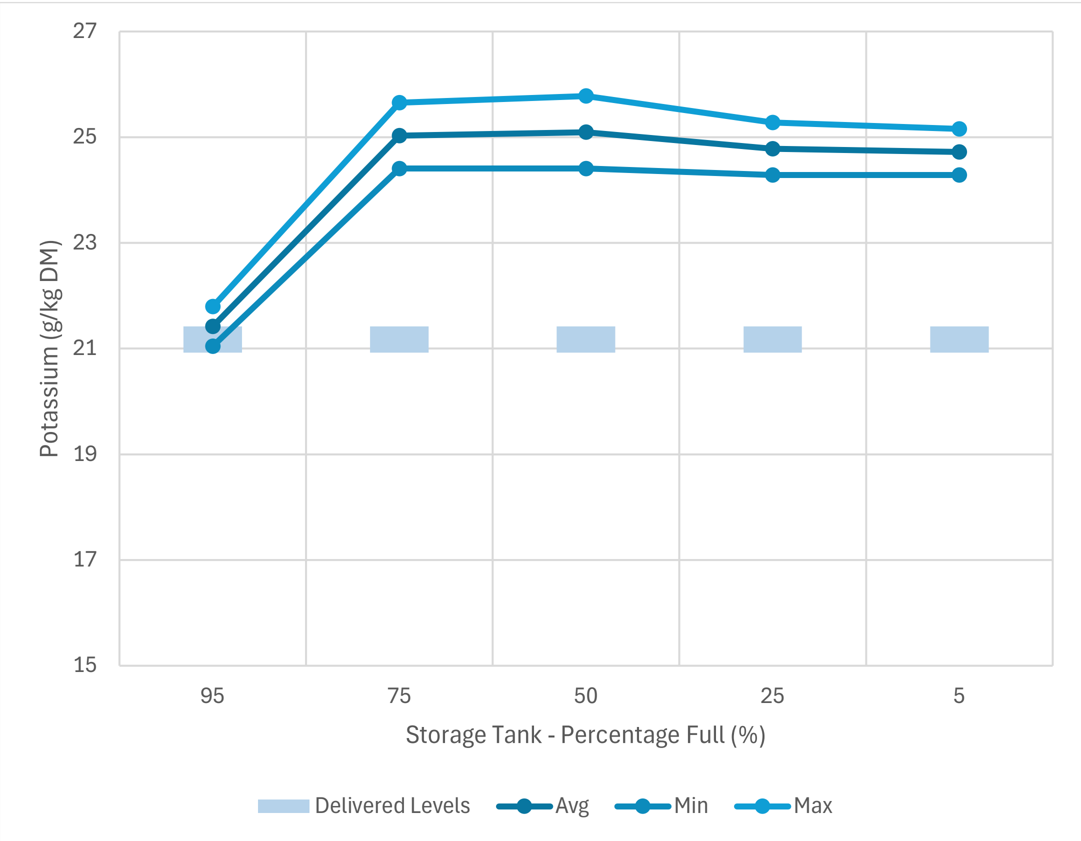


Figure 10. Feedlot 8 potassium concentration (g/kg DM) at delivery and at storage tank sampling capacities

5. Review agitation & reticulation systems for liquid supplements

The ingredients included in the liquid concentrate contain a combination of essential nutrients and additives to supplement the diet of the cattle. Common ingredients used in liquid concentrates in Australia include molasses, vegetable oil, non-protein nitrogen (such as urea, ammonium sulphate), trace minerals (cobalt, copper, iodine, iron, manganese, zinc, selenium), macro minerals (limestone, magnesium, salt, potassium, sulphur)), vitamins (A, , E) and other additives to enhance animal performance (such as ionophores – monensin, lasalocid; and probiotics). The liquid concentrate provides a convenient way to combine many ingredients into one combined single ingredient which can easily be handled and added to the other ration ingredients to provide a nutritionally balanced ration to meet animal requirements and enhance performance.

Manufactures of liquid concentrates invest in specialised equipment, quality systems (i.e. FeedSafe, ISO) and rely on specific manufacturing expertise to make sure liquid concentrates are made with the correct nutrient levels, stable in solution or suspension and also mixed homogenously so that the ingredients are evenly distributed throughout the concentrate. Ingredients are also added to the liquid concentrate (i.e. water, suspending agents, clays, emulsifiers) to ensure the integrity of the concentrate by avoiding ingredient separation during transporting to the feedlot and\or during storage at the feedlot. Through quality systems (i.e. Feedsafe, ISO) at manufacturing sites and routine monitoring, a well-mixed liquid concentrate is delivered to storage tanks at the feedlot.

Feedlots have noted the utilisation of air agitation as a possible method to maintain supplement homogeneity. The air agitation would require an air compressor to supply compressed air delivered into storage tanks. Additional requirements include pipes and plumbing to transfer air into storage tanks. The quoted cost of a suitable sized air compressor is $2,000 (3HP with 100L storage) and associated pipes and plumbing would be $1,000 per tank, for a total price of $3,000 for a feedlot with one storage tank.

Westway Feeds (major North America liquid feed manufacturer) has a concern with the effectiveness of air agitation as a method to homogenise liquid concentrate. Air tends to concentrate within a central area within tank creating a void as the air makes its way to the top of the tank. In addition, air can promote oxidative reactions, potentially degrading the bonding and increasing separation risk once agitation cycle is completed.

Homogenising the stored liquid concentrate prior to transfer to mixing equipment for mixing with other ration ingredients could promote integrity of liquid concentrate in storage. Homogenising methods include:

- Recirculation – using a pump to move liquid concentrate from base of tank to top
- Air agitation – using compressed air to circulate liquid concentrate within the storage tank

These methods offer the simplest systems for application to existing tanks. In contrast, physical agitation using equipment to stir/mix liquid concentrate within the storage tank would require extensive retrofitting with mechanical equipment and may not be suitable for existing tank structures.

Only one of the four current Australian manufacturers provide mixing instructions when using liquid concentrates in feedlot rations. In contrast, the USA have specific recommendations relating to mixing liquid concentrates with ionophores such as monensin prior to use. The USA legislation requires liquid concentrates to have mixing instructions on the label as described by Figure. 11.

CAUTION: USE ONLY AS DIRECTED.
Use only as recommended on this tag.
See **MIXING DIRECTIONS**

****MIXING DIRECTIONS****

A. For liquid Type B feeds stored in re-circulating tank systems: Re-circulate or agitate immediately prior to use for not less than 10 minutes, moving not less than 1% of the tank contents per minute from the bottom of the tank to the top or creating a turbulence at the bottom of the tank that is visible at the top. **Re-circulate or agitate daily, as described above, even when not in use.**

B. For liquid Type B feeds stored in mechanical, air or other agitation-type tank systems: Agitate immediately prior to use for not less than 10 minutes, creating a turbulence at the bottom of the tank that is visible at the top. **Agitate daily, as described even when supplement is not used.**

Do not feed undiluted. Feeding undiluted or mixing errors resulting in high concentrations of monensin has been fatal to cattle and could be fatal to goats.

Figure 11. Mixing instruction label provided by Westaway Liquid Feed Supplement

Paul Mostyn and Wes Klett from Westway Feed USA were identified as subject matter experts, and they were interviewed on the use of monensin in liquid supplements and the current recommendations relating to mixing liquid concentrates in storage tanks. The current recommendations for Westway Feed customers are to recirculate the liquid concentrate in the storage tank (i.e. using a pump) for more than 10 minutes prior to adding to the feedlot ration. The recommendation from Westway Feed is for 400kg of liquid concentrate to be pumped per minute, which equates to 4,000kg per 10 minutes.

Historically, the recirculation recommendation has been on the product label of Rumensin from Elanco Animal Health for 30 plus years as a standard recommendation however, objective measurements assessing the effectiveness of these recommendations in relation to improving homogeneity is unknown. It is also unknown the number of USA feedlots which comply with these recommendations.

As part of the interview with Westway Feed, it was noted that the USA currently use monensin contained within a cereal grain (corn) carrier which tends to float when included in liquid concentrates. This has been identified as a major issue and a potential risk for toxicity to occur especially when the level of the liquid concentrate in the storage tank becomes low and the floating monensin begins to concentrate, causing a higher level of monensin to be delivered into the feedlot ration than expected level. Westaway Feed were particularly interested that in Australia, a technical grade of monensin is used in liquid supplements which uses a limestone carrier. Further to this, in Australia the particle size specification of the technical grade monensin is 75 microns (i.e. 90% or less) as compared to the particle size of the US monensin is 840 microns. Granular or dust free monensin is not recommended to be used in liquid concentrates. It would seem the technical grade monensin should be the only form of monensin to be used in liquid concentrates. This could also provide guidance for other ionophores and other additives in liquid concentrates.

An additional difference is use by dates of liquid concentrates between the two countries. In the US, it is 8 weeks as compared to up to 6 months in Australia (blended concentrate).

Currently, Australian feedlots don't have sufficient capacity pumps to achieve transfer rates recommended by the Westaway team (400kg per minute) for recirculation or would require investment in air compressor with pipes and plumbing to achieve adequate air distribution within storage tank for air-agitation.

The recirculation option would require an additional pump as it is most likely the current pump will be under specification for the required turnover rate of 400kg of concentrate per minute. The quoted cost of a suitable pump is $7,500 (roto pump to deliver 400kg per min @ 20 M total head, 2 bar) and the associated pipe work is $1,000 per tank, for a total price of $8,500 for a feedlot with one storage tank. As per the recommendation from Westway Feeds, this is their preferred method of homogenising the liquid concentrate in storage tanks at the feedlot.

Homogenising liquid concentrate in feedlot storage tanks is one suggestion to help reduce adverse effects of liquid concentrates on animal welfare and food safety. At this stage, there is no objective measurements to use to valid the need to homogenise liquid concentrates prior to using in feedlot rations, apart for submission of supplement samples as the tank empties, and comparison against delivered nutrient composition. A concern for daily re-circulation is the risk of shearing suspending agent due to multiple times the concentrate passes through the pump. Loss of binding capacity of suspending agent risk limestone fallout and compromising concentrate quality.

6. Benchtop methodology

6.1 Experimental design

Experimental design for the benchtop comparison was based on three mixing treatments (control, aeration, re-circulation) and two supplement types (blended, suspension). Each treatment was duplicated providing a total of 6 batches for each supplement type as described in Table 18.

Table 18. Experimental design of liquid supplement batches for benchtop assessment

Supplements		Blended			Suspension	
Treatment	Control	Aeration	Re-circ	Control	Aeration	Re-circ
Duplicate1	1	1	1	1	1	1
Duplicate2	1	1	1	1	1	1
Totals						
Duplicates	2	2	2	2	2	2
Treatments		6			6	

The batches were held in glass 2 Litre squibs as shown in Figure 12. Each squib had a plastic stop cock for ease of sampling. Squibs were held in elevated metal stands to allow ease of sample recovery.

Figure 12. Glass squib used to hold liquid supplement treatments

Treatments and duplicate identification were blinded to avoid potential bias when samples were analysed. Identification method described in Table 19.

Table 19. Sample identification system of supplement type, treatment and duplication removed from bottom of squib

Suppl Type	Dingo (Blend)			Devil (Suspension)		
Mixing Method	Goanna	Frill Neck	Blue Tongue	Goanna	Frill Neck	Blue Tongue
	(Control)	(Re-Circ)	(Air)	(Control)	(Re-Circ)	(Air)
Duplicate	Gallah	Rosella		Gallah	Rosella	
	(1)	(2)		(1)	(2)	
Samples	Wallaby		(Delivery)	Wallaby		(Delivery)
	Wombat		(95%)	Wombat		(95%)
	Koala		(50%)	Koala		(50%)
	Possum		(5%)	Possum		(5%)
Repeat only	Catfish		(0%)	Catfish		(0%)

The experimental design was repeated using a different analytical lab for sample analysis to assist with identifying differences between mixing methods for each supplement and sampling period. The repeated experiment is referred to as repeat. The repeated project used the same formulated blend and suspension supplements. Sampling was also the same, except an additional sample was taken collecting the last of the supplement remaining in the squib (Catfish) as described in Table 19 as well as samples removed from the top of the squib as described in Table 20.

Table 20. Sample identification system of supplement type, treatment and duplication removed from top of squib for repeat benchtop

Suppl Type	Dingo - Wedgie (Blend)			Devil - Wedgie (Suspension)		
Mixing Method	Goanna	Frill Neck	Blue Tongue	Goanna	Frill Neck	Blue Tongue
	(Control)	(Re-Circ)	(Air)	(Control)	(Re-Circ)	(Air)
Duplicate	Gallah	Rosella		Gallah	Rosella	
	(1)	(2)		(1)	(2)	
Samples	Wombat		(95%)	Wombat		(95%)
	Koala		(50%)	Koala		(50%)
	Possum		(5%)	Possum		(5%)

The first benchtop study began with collection of the suspension supplement on 9th September 2024 and the blended supplement on 11th September 2024. The experiment began on the 29th September 2024. The repeat benchtop study began with collection of suspension and blended supplements on the 3rd March 2025. The repeat experiment began on the same day. In both experiments, each supplement type was stirred using an electric drill fitted stirrer prior to transfer into squibs. All squibs were filled to the 2L level. The first project had squibs set up on two tables, as shown in Figure 13. The repeat project had squibs held within a wooden cradle as shown in Figure 14 to improve stability and avoid squib loss.

Figure 13. Arrangement of squibs filled with blended and suspension supplements

Figure 14. Arrangement of squibs filled with blended and suspension supplements in wooden cradle

The darker brown coloured supplement was the blend. The light brown coloured supplement was the suspension supplement. Table 21 describes the characteristics of each supplement type.

Table 21. Liquid supplement characteristics with theoretical nutrient values

	Blend	Suspension
Manufacturing site	Kingstown, QLD	Dalby, QLD
Dry matter %	66.5	70.8
Ash % DM	22.93	42.1
Calcium % DM	0.7	14.8
Potassium % DM	2.83	1.9
Monensin mg/kg DM	461.2	452.2
DM inclusion %	3.3	5.3

The mixing treatments were designed to meet the directions provided by Elanco for supplements containing monensin. Mixing directions recommend mixing to occur immediately prior to use for not less than 10 minutes creating a turbulence at the bottom of the tank that is visible at the top. The aeration treatment used fish tank aerators placed within the four squibs (2 duplicates of blend and suspension supplements) and fitted to a four-outlet air pump. The re-circulation treatment was implemented by decanting the complete contents held by the squib into a plastic tub which was then poured back into the top of the squib. A cooking spatula was used to remove any remaining supplement from the tub to avoid excessive supplement loss. The control treatment was not changed once filling of squibs with the appropriate supplement was completed. Figure 15 shows operating aerator and a cleaned plastic tub used for re-circulation.

Figure 15. Aerators operating and a cleaned plastic tub

6.2 Sampling & analysis

Sampling from the first benchtop experiment began on the 29th August 2024. The repeat benchtop sampling began on 10th March 2025. At the time of the first experiment, weather conditions were cool (daytime temperatures low 20°C, nighttime temperatures low 10°C). To avoid temperature interfering with mixing methods, heat pads set at 25°C were wrapped around all squibs until daytime temperatures increased. Duplicate samples from each supplement type (blend, suspension) were removed directly from the delivery tub. Each week for the 12week duration of the project, all treatments had approximately 167ml of supplement removed from each squib.

Sample quantities removed from the squibs were retained only during week 1, 6 and 11 with the final quantity remaining in the squib retained on week 12 for the repeat. These sample periods attempted to represent capacity of squibs at 95, 50 and 5% full. All samples were placed in 150ml plastic jar with screw top lid. All other quantities removed during non-sampling weeks were discarded.

The aeration treatments for both experiments had air pumped into squibs for approximately 45 to 50minutes each week for the first 6-week period which was then reduced to 20 to 25minutes for the final 6-week period to reflect reduced quantity held within squibs and reflect reduced time required for re-circulation.

During the same time, the re-circulation treatments were run. The blended supplements emptied at a faster rate than the suspension supplements reflecting differences in viscosity of the two supplements. Each week for the first 6-week period, blended supplements required 15 to 20 minutes to empty, whereas the suspension supplements required 35 to 45minutes. During the final 6-week period, each week the blended supplement required 12 to 15minutes to empty, whereas the suspension supplement required 20 to 25 minutes. Time differences between the first and final 6 weeks periods again reflecting reduction in volume of supplement held in squibs. During the first experiment, sampling period, two squibs failed for the first sampling period. Both were blended supplements. One was an aeration treatment (duplicate 1) discovered on the 26/08/2024. Contents of squib were salvaged for continuing sampling. The other loss occurred 08/10/2024. This was a control treatment (duplicate 2). Contents of squib could not be recovered missing the 5% sample. During the repeat experiment, no squibs were lost.

Samples were retained with squibs out of direct sunlight until end of 12week period. Duplicate sample jars were placed in a zip lock bag to protect sample integrity in case of sample jar leakage or breakage. Samples were delivered directly to project team member in Brisbane for collecting and co-ordinating postage of samples. The first benchtop project samples were sent to Symbio Alliance for testing. The repeat samples were sent to AsureQuality in New Zealand.

All duplicate samples collected were analysed for monensin (mg/kg), moisture (%), ash (g/100g), calcium (g/kg), zinc (mg/kg), copper (mg/kg) and potassium (g/kg). Results were provided on a dry matter basis. Monensin was analysed using an in-house LC-MSMS method. The process involves extracting the analytes from the samples by sonication in an aqueous methanol solution. The extract is then centrifuged, diluted and filtered. Quantitative and confirmatory analysis is performed simultaneously by LC/MS/MS (liquid chromatography, mass spectrometry) and relying on an internal standard calibration. Moisture was analysed using a vacuum oven based on the AOAC 934.01 method. Ash was analysed using the method based on AOAC 923.03 and 920.153. Zinc, calcium, copper and potassium levels within the liquid supplement samples were analysed using an inductively coupled plasma mass spectrometer.

The duplicate samples collected during both projects were summarised by calculating the mean, standard deviation and co-efficient of variation.

The difference between each duplicate sample, including the sample taken from the delivery tanker was used to calculate average laboratory error. Laboratory error can be reviewed as samples were blinded.

An average absolute deviation with error percentage (Absolute Dev with Error%) was calculated to identify the variation between the Lab Error and the mean delivery nutrient level. The purpose of this calculation was to determine if the differences between the samples and the delivered mean were actual or in-field or due to Lab Error.

An absolute deviation percentage (Absolute Dev%) was calculated to define the differences between the average storage tank samples and the delivered sample mean. This was calculated firstly determining the average delivered nutrient level from the duplicate samples. Then the absolute deviation was calculated from the delivered nutrient level at each squib level by averaging the nutrient level from each duplicate sample taken from the squib and then the variance of each average squib result. Then finally the delivered average was calculated. The absolute deviation percentage was calculated using the variance percentage at each level in the squib.

7. Benchtop results & discussion

7.1 Data corrections

For samples from the first experiment, methodology errors or outliers were apparent. The result tables provided have these outliers removed. The result summaries were conducted for each individual analysed component (monensin, moisture, ash, calcium, zinc, copper, potassium) described in the following tables.

Laboratory error occurred with analysis of samples from the first experiment. Methodology of sample preparation for analysis appeared to be problematic with variation in sample homogeneity. In addition to sampling method, an apparent oversight occurred on reporting of the results with failure to account for the dilution of the liquid supplement as part of the sample preparation process (samples diluted by 50% with water). Differences between reported results and expected levels, as well as between duplicate 1 and 2 samples highlighted reported nutrient levels at half the concentration. Results from the first experiment are not presented where errors were above acceptable levels identified by CV values or in comparison to theoretical values as described in Table 22.

Table 22. Nutrients from the first experiment not included in results due to excessive co-efficient of variation (CV%), lab error and absolute deviation values

Monensin mg/kg DM	Control	Re-circulation	Aeration
CV%	28.1	19.8	14.5
Lab error	187.8	193.3	72.9
Absolute Dev%	21.7	22.1	24.0
Zinc mg/kg DM			
CV%	11.2	8.3	15.1
Lab error	80.6	140.6	256.8
Absolute Dev%	17.9	12.4	10.7
Copper mg/kg DM			
CV%	22.1	14.6	20.9
Lab error	35.5	106,2	118.2
Absolute Dev%	31.5	16.3	20.0

In contrast, the repeat benchtop experiment provided acceptable laboratory error and nutrient values that compared well across sampling periods and with expected concentrations.

7.2 Blended supplement

7.2.1 Monensin

The monensin analysis from the repeated experiment sampled from both the base and top of the squib were very consistent as described by Tables 23 and 24 respectively. Monensin analysis achieved less than 5% CV% for both base and top sampling, on average less than 20mg/kg difference between the duplicate samples and difference to the delivered value of 5% as described by the absolute deviation.

Table 23. Effect of mixing treatment of repeated experiment on blended supplement monensin concentration sampled from base

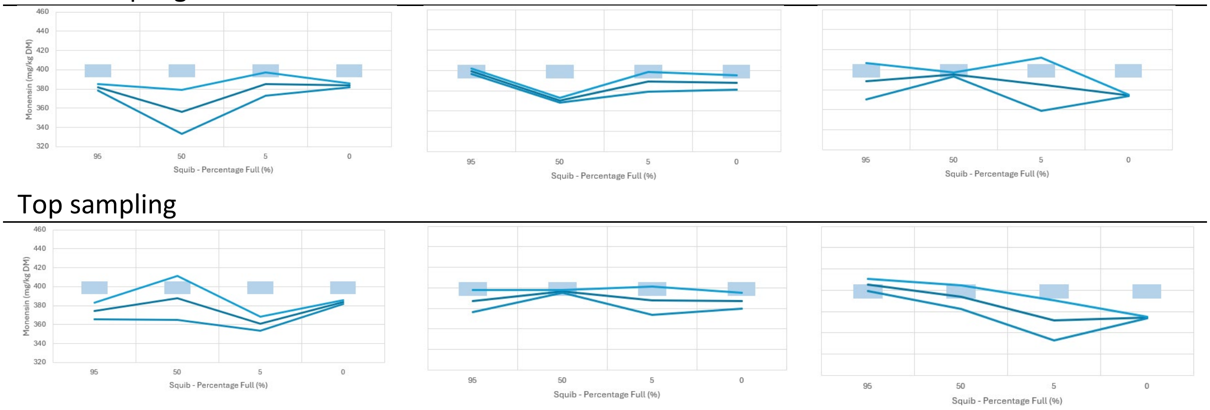
	Monensin (mg/kg DM)					
Blend Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	381	19.5	5.1	18.9	5.1	4.8
Re-Circulation	389	12.9	3.3	11.4	2.8	3.1
Aeration	388	18.0	4.6	21.6	5.9	2.6

Table 24. Effect of mixing treatment of repeated experiment on blended supplement monensin concentration sampled from top

	Monensin (mg/kg DM)					
Blend Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	381	18.4	4.8	19.1	5.2	5.3
Re-Circulation	391	10.9	2.8	16.0	4.2	1.6
Aeration	389	17.8	4.6	17.0	4.5	4.3

Changes to monensin concentration during the sampling period from both base and top of squib is described in Figure 16.

Base sampling



a) Control b) Re-circulation c) Aeration

Figure 16. Effect of mixing treatments of repeated experiment on monensin concentration over a 12week sampling period of blended supplement sampled from the base and top of squib

The control and re-circulation treatments exhibited a similar trend in concentration over the sampling period. For these treatments, monensin concentration decreased at the 50% sampling period base and increased at the same sampling period when supplement was removed from the top. This may indicate that in blended supplements monensin can gravitate to the top, but the changes remain within safe usage levels when using the very small particle size monensin. The remaining sampling periods held consistent if not slightly below the concentration of the delivered sample.

In contrast, the aeration treatment exhibited decreasing monensin concentrations across all sampling periods for both base and top sampling. The monensin concentration appeared slightly elevated for the top sample compared to the delivered level and decreased with sampling periods. It appears the addition of air is affecting monensin distribution differently in that both base and top samples values are similar. A characteristic of aeration is formed air bubbles travel through channels, even though the air is transferred into a tank through a distributor or manifold. The method of aeration used in this study relied on a folded aeration stick where air passed through multiple locations into the squib. However, when observing the air bubbles forming at the top of the squib, bubbles appeared to be concentrated in only one or two areas, indicating that even in a smaller volume container, air channels were formed. Of the mixing methods, aeration is less efficient at creating a homogenised sample.

7.2.2 Moisture

The moisture analysis from the repeated experiment sampled from both the base and top of the squib were very consistent as described by Tables 25 and 26 respectively. Variation and error assessments of treatment samples were minimal.

Table 25. Effect of mixing treatment of repeated experiment on blended supplement moisture sampled from base

	Moisture (%)					
Blend Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	33	0.2	0.5	0.2	0.5	0.4
Re-Circulation	33	0.4	1.1	0.7	2.1	0.5
Aeration	33	0.2	0.7	0.3	1.0	0.6

Table 26. Effect of mixing treatment of repeated experiment on blended supplement moisture sampled from top

	Moisture (%)					
Blend Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	33	0.4	1.2	0.5	1.8	0.8
Re-Circulation	33	0.3	0.9	0.2	0.6	1.6
Air	33	0.3	0.9	0.4	1.4	0.7

7.2.3 Ash

The ash analysis from the repeated experiment sampled from both the base and top of the squib were very consistent as described by Tables 27 and 28 respectively. Variation and error assessments of treatment samples were minimal.

Table 27. Effect of mixing treatment of repeated experiment on blended supplement ash sampled from base

	Ash (% DM)					
Blend Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	23	1.6	7.1	1.0	2.7	7.9
Re-Circulation	23	1.3	5.6	1.3	4.8	8.4
Aeration	22	0.9	3.9	1.2	3.6	1.9

Table 28. Effect of mixing treatment of repeated experiment on blended supplement ash sampled from top

	Ash (% DM)					
Blend Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	23	1.3	5.7	1.2	3.8	5.9
Re-Circulation	23	0.8	5.0	0.9	4.9	5.6
Aeration	22	0.8	3.4	0.9	2.1	2.2

7.2.4 Zinc

The zinc analysis from the repeated experiment sampled from both the base and top of the squib were very consistent as described by Tables 29 and 30 respectively. Variation and error assessments of treatment samples were minimal.

Table 29. Effect of mixing treatment of repeated experiment on blended supplement zinc concentration sampled from base

	Zinc (mg/kg DM)					
Blend Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	1449	32.0	2.2	51.5	3.1	0.4
Re-Circulation	1465	30.0	2.1	30.7	1.9	1.9
Aeration	1463	32.3	2.2	33.5	1.5	1.5

Table 30. Effect of mixing treatment of repeated experiment on blended supplement zinc concentration sampled from top

	Zinc (mg/kg DM)					
Blend Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	1461	36.3	2.5	54.2	3.3	1.6
Re-Circulation	1461	33.7	2.3	57.4	3.6	1.5
Aeration	1467	70.0	4.8	81.7	5.7	2.6

7.2.5 Copper

The copper analysis from the repeated experiment sampled from both the base and top of the squib were very consistent as described by Tables 31 and 32 respectively. Variation and error assessments of treatment samples were minimal.

Table 31. Effect of mixing treatment of repeated experiment on blended supplement copper concentration sampled from base

			Copper (mg/kg DM)			
Blend Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	348	14.5	4.2	17.3	5.3	3.5
Re-Circulation	336	7.9	2.3	6.4	2.1	2.2
Aeration	334	7.5	2.2	8.8	2.2	1.2

Table 32. Effect of mixing treatment of repeated experiment on blended supplement copper concentration sampled from top

			Copper (mg/kg DM)			
Blend Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	337	7.1	2.1	14.7	4.5	2.5
Re-Circulation	335	8.3	2.5	15.4	4.7	0.8
Aeration	339	10.7	3.2	14.6	4.4	3.1

7.2.6 Potassium

The potassium analysis from the repeated experiment sampled from both the base and top of the squib were consistent as described by Tables 33 and 34 respectively. Variation and error assessments of treatment samples across the sampling periods were minimal.

Table 33. Effect of mixing treatment of repeated experiment on blended supplement potassium concentration sampled from base

			Potassium (g/kg DM)			
Blend Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	27	0.5	1.8	0.4	1.5	0.8
Re-Circulation	27	0.5	1.7	0.3	2.1	0.9
Aeration	27	0.5	1.9	0.4	1.9	1.0

Table 34. Effect of mixing treatment of repeated experiment on blended supplement potassium concentration sampled from top

			Potassium (g/kg DM)			
Blend Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	26	0.6	2.4	0.6	2.8	2.4
Re-Circulation	27	0.5	1.8	0.3	1.4	1.4
Aeration	26	0.5	1.8	0.5	2.1	0.8

7.3 Suspension supplement

7.3.1 Monensin

Monensin concentration across the three treatments for suspension supplement sampled during the first experiment all achieved desired CV target of less than 10% with low error values as described in Table 35.

Table 35. Effect of mixing treatment on suspension supplement monensin concentration

	Monensin (mg/kg DM)					
Suspension Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	466	28.7	6.2	22.3	3.7	7.4
Re-circulation	485	27.0	5.6	39.3	8.3	2.3
Aeration	474	24.2	5.1	22.8	3.8	5.2

The first experiment treatment values corresponded well with the repeated analysis sampled from the base as described in Table 36.

Table 36. Effect of mixing treatment of repeated experiment on suspension supplement monensin concentration sampled from base

	Monensin (mg/kg DM)					
Suspension Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	466	19.9	4.3	25.3	5.5	4.0
Re-Circulation	479	22.2	4.6	29.7	6.5	5.2
Aeration	491	26.6	5.4	18.4	3.6	8.1

Table 37 describes the effect of mixing treatment of repeated experiment on monensin concentration sampled from the top of the squib.

Table 37. Effect of mixing treatment of repeated experiment on suspension supplement monensin concentration sampled from top

	Monensin (mg/kg DM)					
Suspension Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	459	21.9	4.8	23.8	5.1	4.2
Re-Circulation	482	25.2	5.2	29.5	6.6	6.7
Aeration	487	22.1	4.5	14.9	2.6	7.2

Changes to monensin concentration during the sampling period of the suspension supplement mixing treatments from both base and top of squib is described in Figure 17. Both mixing treatments had higher monensin concentrations for base and top sampling compared to the delivery concentration. The aeration treatment monensin concentrations increased at the 5% and 0% sample. This contrasts with the blended supplement where monensin concentration decreased with sampling period described in Figure 16. This suggests that suspension supplements may respond differently to aeration with the addition of air creating separation of ingredients. These results also suggest whether any mixing method is required for suspension supplements.

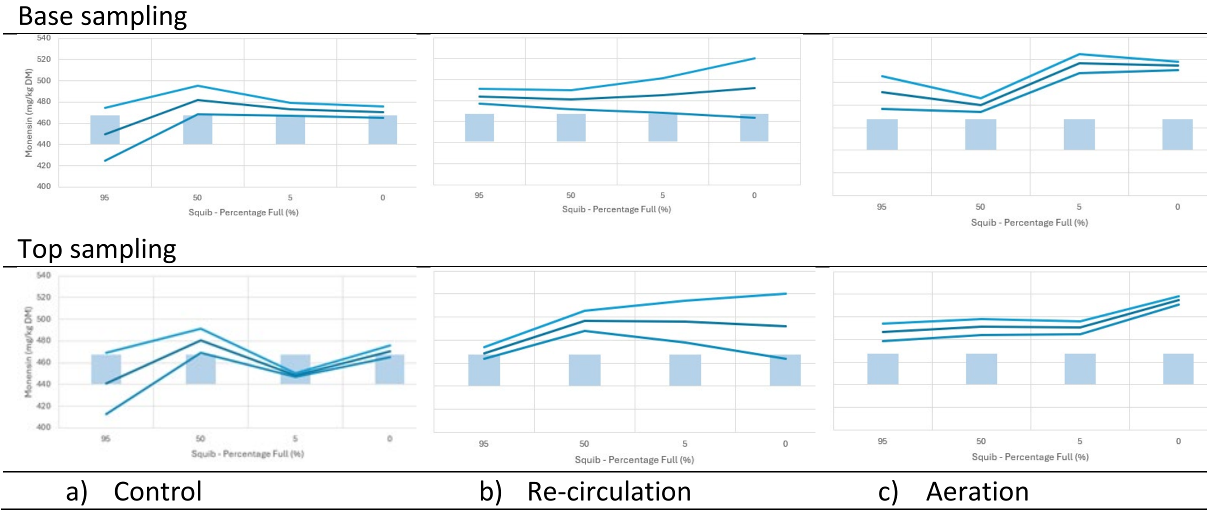


Figure 17. Effect of mixing treatments of repeated experiment on monensin concentration over a 12week sampling period of suspension supplement sampled from the base and top of squib

7.3.2 Moisture

The moisture analysis from the repeated experiment sampled from both the base and top of the squib were very consistent as described by Tables 38 and 39 respectively. Variation and error assessments of treatment samples were minimal.

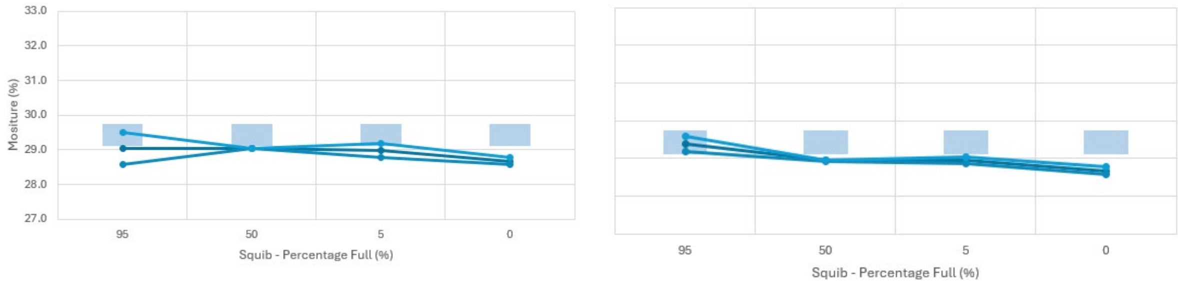
Table 38. Effect of mixing treatment of repeated experiment on suspension supplement moisture sampled from base

	Moisture (% DM)					
Suspension Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	29.4	0.4	1.3	0.4	1.2	0.9
Re-Circulation	29.0	0.5	1.7	0.8	2.7	0.7
Aeration	29.0	0.3	1.1	0.4	1.3	0.5

Table 39. Effect of mixing treatment of repeated experiment on suspension supplement moisture sampled from top

	Moisture (% DM)					
Suspension Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	29.0	0.5	1.9	0.5	1.6	1.3
Re-Circulation	29.1	0.6	1.9	0.9	3.3	0.9
Aeration	29.0	0.3	1.1	0.3	0.7	1.1

The aeration treatment of the repeated experiment shows a decline in moisture over the sampling periods as described in Figure 18. The decline in moisture was not observed in the control or re-circulation treatments. The decrease in moisture or alternatively increase in dry matter may suggest separation occurring within the aeration treatment.



a) Sample at base b) Sample at top

Figure 18. Effect of aeration of repeated experiment on moisture content of suspension supplement aeration treatment over a 12week sampling period

7.3.3 Ash

Ash concentration across the three treatments for suspension supplement sampled during the repeated experiment for base and top samples are described in Table 40 and 41. Variation and error assessments of treatment samples were minimal.

Table 40. Effect of mixing treatment of repeated experiment on suspension supplement ash sampled from base

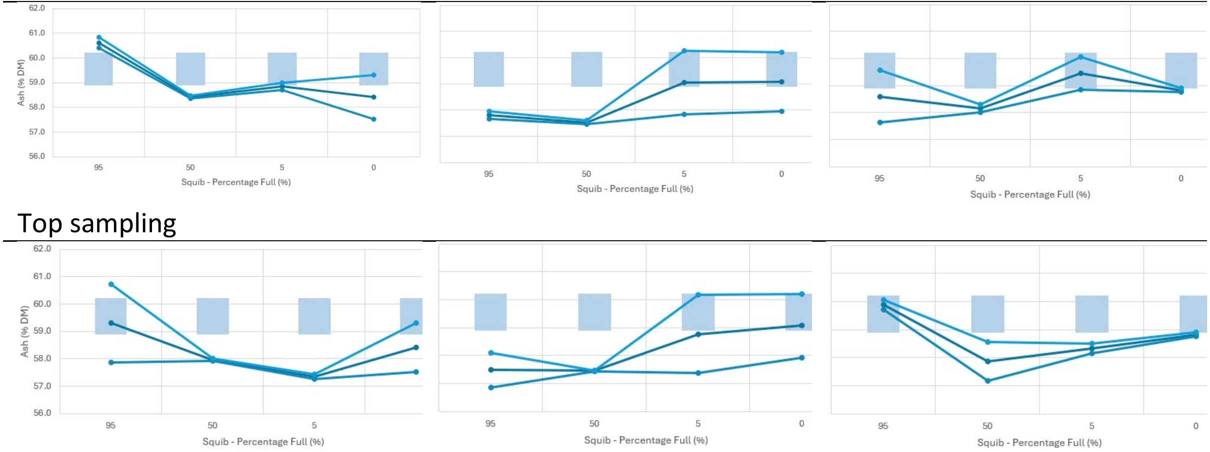
	Ash (% DM)					
Suspension Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	59.0	1.0	1.7	0.8	1.1	1.9
Re-Circulation	58.5	1.1	1.9	1.3	2.2	1.7
Aeration	59.0	0.3	1.1	0.3	0.7	1.1

Table 41. Effect of mixing treatment of repeated experiment on suspension supplement ash sampled from top

	Ash (% DM)					
Suspension Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	59.0	1.0	1.7	0.8	1.1	1.9
Re-Circulation	58.3	1.2	2.1	1.5	2.7	2.0
Aeration	58.8	0.9	1.4	0.7	0.9	1.5

Changes to ash levels during the sampling period of the suspension supplement mixing treatments from both base and top of squib is described in Figure 19. The changes appear similar between base and top samples for control and re-circulation but vary between the mixing systems. The control ash level is elevated at 95%, then decreases at 50% and remains relatively stable to 0%. The re-circulation ash level is less than the delivery ash content at 95% and 50% but then increases at 5% and 0%. The aeration ash content is relatively stable during the base sampling but for top sampling decreases at 50% and increases slightly at 5% and 0%. It is difficult to determine what is causing this variation other than the mixing methods of re-circulation and aeration effect suspension supplement consistency and effect differs between the mixing methods.

Base sampling



a) Control b) Re-circulation c) Aeration

Figure 19. Effect of mixing treatments of repeated experiment on ash concentration over a 12week sampling period of suspension supplement sampled from the base and top of squib

7.3.4 Zinc

Zinc concentration across the three treatments for suspension supplement sampled during the repeated experiment for base and top samples are described in Table 42 and 43. Variation and error assessments of treatment samples were minimal.

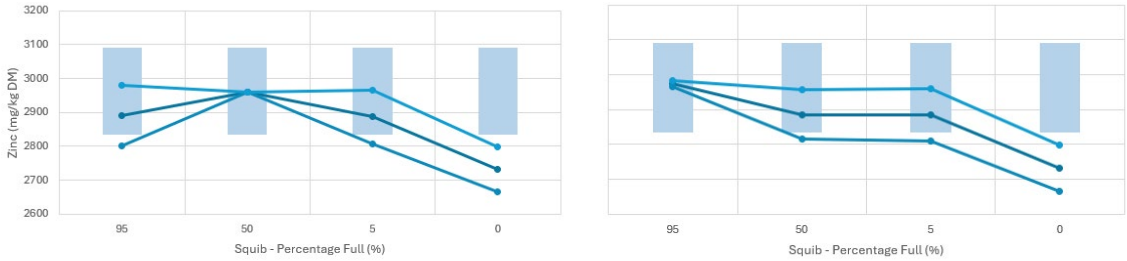
Table 42. Effect of mixing treatment of repeated experiment on suspension supplement zinc sampled from base

	Zinc (mg/kg DM)					
Suspension Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	2930	93.6	3.2	90.7	1.7	1.8
Re-Circulation	2889	123.7	4.3	202.9	6.4	2.6
Aeration	2886	124.1	4.3	144.8	4.0	2.6

Table 43. Effect of mixing treatment of repeated experiment on suspension supplement zinc sampled from top

	Zinc (mg/kg DM)					
Suspension Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	2943	84.0	2.9	94.8	1.8	1.1
Re-Circulation	2891	99.1	3.4	139.0	3.7	2.6
Aeration	2888	122.9	4.3	139.1	3.7	3.4

The aeration treatment of the repeated experiment shows a decline in zinc over the sampling periods as described in Figure 20. The decline in zinc was not observed in the control or re-circulation treatments. Similarly to the effect observed with moisture in Figure 18, aeration may create separation that is only evident when the tank is nearly empty.



a) Sample at base b) Sample at top

Figure 20. Effect of aeration of repeated experiment on zinc content of suspension supplement aeration treatment over a 12week sampling period

7.3.5 Calcium

Calcium concentration across the three treatments for suspension supplement sampled during the repeated experiment for base and top samples are described in Table 44 and 45. Variation and error assessments of treatment samples were minimal.

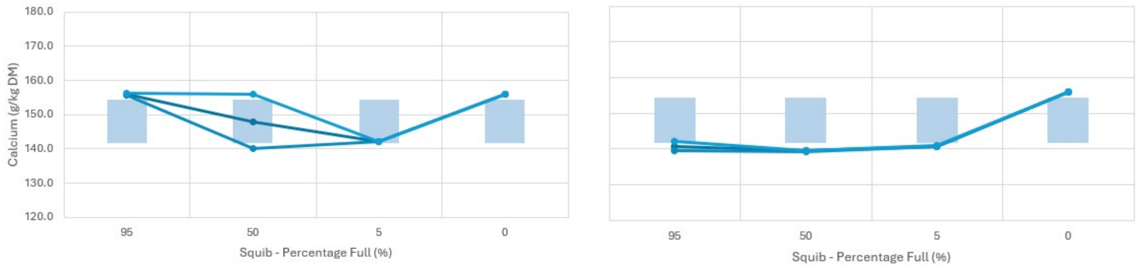
Table 44. Effect of mixing treatment of repeated experiment on suspension supplement calcium sampled from base

	Calcium (g/kg DM)					
Suspension Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	150	7.4	4.9	6.0	2.9	2.4
Re-Circulation	148	17.1	7.2	29.9	12.8	1.7
Aeration	148	7.8	5.2	8.6	5.1	2.7

Table 45. Effect of mixing treatment of repeated experiment on suspension supplement calcium sampled from top

	Calcium (g/kg DM)					
Suspension Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	145	7.3	5.0	3.3	0.6	6.6
Re-Circulation	144	6.2	4.3	6.9	3.7	3.5
Aeration	146	7.7	5.2	5.8	2.8	5.2

The concentration levels of calcium across the sampling periods were consistent for re-circulation and aeration. For the control, there was a slight increase in concentration at the 0% sampling period, indicating the final supplement remaining in the squib after 12weeks had accumulated calcium. The relative quantity is small at 10 to 15g/kg and the 12week period represents twice the time of the longest storage period of a surveyed feedlot at 40days.



a) Sample at base b) Sample at top

Figure 21. Effect of repeated experiment on calcium content of suspension supplement control treatment over a 12week sampling period

7.3.6 Copper

Copper concentration across the three treatments for suspension supplement sampled during the repeated experiment for base and top samples are described in Table 46 and 47. Variation and error assessments of treatment samples were minimal for control and aeration. The re-circulation treatment for the base sampling had a greater lab error of 29.9mg/kg increasing the absolute deviation with error above 10% as described in Table 46. The difference between the delivered copper level and the treatment level remained very low at 1.7%. A similar effect occurred for the top sampling, but absolute deviation error was under 10% as described in Table 47.

Table 46. Effect of mixing treatment of repeated experiment on suspension supplement copper sampled from base

			Copper (mg/kg DM)			
Suspension Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	249	12.6	5.1	11.8	3.4	3.3
Re-Circulation	237	17.1	7.2	29.9	12.8	1.7
Aeration	239	13.7	5.7	17.2	6.3	3.1

Table 47. Effect of mixing treatment of repeated experiment on suspension supplement copper sampled from top

			Copper (mg/kg DM)			
Suspension Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	237	15.7	6.6	12.1	3.6	4.9
Re-Circulation	236	14.4	6.1	21.9	8.7	2.4
Aeration	241	12.6	5.2	14.0	4.5	5.4

7.3.7 Potassium

Potassium concentration across the three treatments for suspension supplement sampled during the repeated experiment for base and top samples are described in Table 48 and 49. Variation and error assessments of treatment samples were minimal for all treatments.

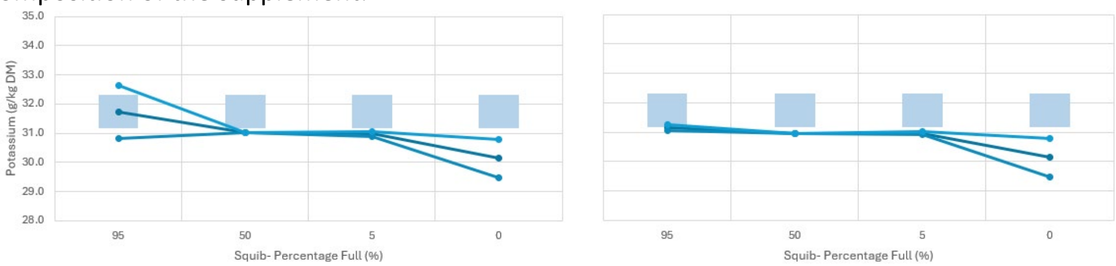
Table 48. Effect of mixing treatment of repeated experiment on suspension supplement potassium sampled from base

	Potassium (g/kg DM)					
Blend Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	32	0.8	2.5	1.5	4.9	0.4
Re-Circulation	31	0.8	2.7	1.1	3.5	1.7
Aeration	31	0.9	2.8	0.9	2.6	2.4

Table 49. Effect of mixing treatment of repeated experiment on suspension supplement potassium sampled from top

	Potassium (g/kg DM)					
Blend Treatments	Mean	SD	CV%	Lab Error Ave	Absolute Dev with Error%	Absolute Dev%
Control	31	0.8	2.6	1.0	2.9	1.6
Re-Circulation	31	0.9	2.8	1.0	3.2	2.2
Aeration	31	0.7	2.2	0.5	1.3	2.5

The aeration treatment of the repeated experiment shows a decline in potassium at the 0% sampling period as described in Figure 22. The decline in potassium was not observed in the control or re-circulation treatments. The potassium level also tended to remain below the delivery concentration. This also was not observed in the other treatments. It appears that aeration changes the composition of the supplement.



a) Sample at base b) Sample at top

Figure 22. Effect of aeration of repeated experiment on potassium content of suspension supplement aeration treatment over a 12week sampling period

8. Conclusion

Conclusions from B.FLT.4022 Evaluation of liquid supplement storage tank agitation methods and Australian feedlot supplement manufacture and on-site feedlot management including assessment of supplement composition during storage and use include:

- Quality assurance programs play an important role supporting supplement manufacturing process. All supplement manufacturers surveyed had one or more QA programs in place. This QA commitment by supplement manufacturers is supported by the Australian feedlot industry QA program, National Feedlot Accreditation Scheme (NFAS). Feedlots rely on manufacturers QA systems as few apply quality assessments to supplements at receival, storage or during use.
- Liquid supplement manufacturers define and assess maximum particle size of ingredients with low solubility including limestone (75µM) and magnesium carbonate (45µm) and ingredients with toxicity risk such as monensin (75µM). Restricting the use of ingredients based on particle size promotes confidence in liquid supplement integrity as particle size has been identified as a major cause of separation. The baseline variation highlighted the consistency of monensin

concentration during storage and use with all but two feedlots achieving a co-efficient of variation of 10% or less. 93% of feedlots surveyed used suspension supplements (limestone included), ensuring limestone meets required particle size is critical. Toxicity risk ingredients such as monensin should only be sourced and controlled by the supplement manufacturer.
- Only one of the four liquid supplement manufacturers surveyed recommended agitating supplement during storage. The recommendation is in line with mixing instructions provided by Elanco for monensin in the USA. In the USA, monensin has a much larger particle size (840µM) due to its corn carrier compared to Australia's monensin which is held in a limestone carrier (75µM). Agitation methods include re-circulation or air agitation using compressed air that is pumped into the storage tank. None of the feedlots surveyed were agitating liquid supplement in storage. The risk of agitation is loss of binding capacity of suspending agent by shearing during pumping and air promoting oxidation and altering chemical reactions. Further research is required to confirm these effects.
- All manufacturers undertake extensive sampling and testing of carrier composition, specifically the presence of chemical residues, heavy metals and nutrient content.
- Supplement manufacturers promote integrity and consistency of products by providing ingredient and nutrient formulation recommendations. The composition of feedlot supplements tends to follow these recommendations. Baseline composition values for moisture/dry matter were similar to recommended levels for both blended and suspension supplements.
- Despite the availability of more than 30 registered monensin products, both dry and liquid supplement manufacturers had only 1 to 4 products on hand. Liquid supplement manufacturers require monensin to have a particle size no greater than 75µM.
- Many of the adverse experiences reported by supplement manufacturers would be alleviated by following formulation recommendations, ingredient particle size requirements and the feedlot providing clear labelling of which tanks hold what products.

Production based conclusions relating to supplement management at the feedlot include:

- The preferred loading method is direct from the supplement storage tank to the mixer with larger feedlots investing in batch box or liquid batching systems.
- Feedlots using shared delivery lines (plumbing systems connected to other liquid storage such as water, vegetable oil, molasses) causes dilution of supplement and increases shrink. Sharing lines and pulling supplement from multiple tanks at the same time changes the composition of the supplement by pulling residue remaining in storage tanks.
- Feedlots tend to use single supplement with changes to inclusion to suit different rations. Single supplement inclusion improves inventory control, ease of ordering and increases storage capacity.
- Most feedlots had 2 or more tanks, providing ingredient security and reducing freight by accepting larger loads. Supplement storage tank hygiene was low with only 11% reporting tanks cleaned and 39% ensuring the tank is emptied before re-filling. Emptying the tank completely before re-filling reduces dilution of any supplement residue remaining, particularly when supplement turnover is 27 days or more.
- Flat bottom tanks increase the opportunity for residue to accumulate over time, altering the composition of the supplement when re-filled.
- Pencil shrinks of 0.3 or 0.5% are not sufficient to cover actual shrink values of liquid supplements of 1.1 to 1.2%.
- Monensin remains the most commonly used additive with half the feedlots surveyed only using monensin. Two thirds of feedlots preferred to use a single additive. Other additives in use include barbermycin, lasalocid, tylosin and virginiamycin. The S4 additives virginiamycin and tylosin were included in the supplement at a small proportion of feedlots (4%). CTC was directly added to the ration by some feedlots. All feedlots held and maintained veterinary script records for use of S4 additives.

- All feedlots were turning the liquid supplement over quicker than the maximum turnover period that was recommended by manufacturer. Turnover period effects the composition consistency of supplement during use. Monensin levels from the storage tank remained similar to levels reported from the delivery tank when supplement turnover was 18 days or less.

Conclusions from B.FLT. 4022 benchtop evaluation of mixing methods of feedlot blended and suspension liquid supplements include:

- The benchtop study extended more than twice as long (84 days) compared to the longest storage period recorded of the surveyed feedlots (40 days)
- The suspension supplement appeared to remain more consistent over the sampling period compared to the blended supplement
- Mixing methods of re-circulation or aeration did not improve nutrient consistency over the sampling period compared to the control for the suspension supplement.
- Using aeration as a mixing method is not recommended for either blended or suspension supplements.

8.1 Key findings

- Supplement manufacturers have in place QA and operational systems which promote supplement integrity and reliability. Feedlots place a high reliance on supplement manufactures as few apply regular QA to received supplements, unless there is an identified problem.
- Liquid supplement manufacturers focus on particle size of ingredients with low solubility that can compromise supplement integrity (limestone, magnesium carbonate) and monensin, a toxicity risk ingredient.
- Monensin should only be sourced and controlled by the supplement manufacturer, thereby controlling product form and ensuring particle size is no greater than 75µM.
- Review of protocols for cleaning delivery tanks are required to prevent contamination risk between loads.
- None of the feedlots surveyed were mixing liquid supplement in storage.
- Types of antimicrobial products in use by supplement manufacturers and feedlots was limited with 96% of feedlots using monensin and 64% relying on a single additive.
- Pencil shrink values for liquid supplement (0.3 to 0.5%) were insufficient to account for true shrink values.
- Supplement protocols for duplicate sampling both delivery and storage tanks were successful in picking up differences in liquid supplement during usage.
- Storage systems require separate delivery lines for all liquid ingredients. Sharing delivery lines increases the liquid supplement shrink and dilutes supplement concentration.
- Supplement storage tanks should be managed the same way as commodity shed bays (first in, first out). Use one tank at a time and allow tanks to empty before re-filling.
- Turnover periods of 18 days or less improves supplement consistency, irrespective of tank management (shared lines, not emptied). Ensuring supplement tanks are empty before re-filling improves supplement consistency when turnover is 27 days or more.
- Cone bottom tanks provide best method of storing liquid supplements.
- Install sampling tap on each tank directly after the outlet valve to ensure representative samples can be taken.
- Aeration should not be used to improve consistency of high concentration liquid supplements.
- Storage periods need to be considered when determining liquid supplement inclusion level and resulting ingredient and nutrient concentration
- Feedlots requiring extended storage of liquid supplements should consider alternative supplement options (pellets or meal)

- Methodology for analysing liquid supplement nutrients requires review as high and unacceptable analytical variations were common during this project.

8.2 Benefits to industry

The feedlot industry can have confidence in the supplement manufactures to produce products that meet their needs and are safe to use.

The integrity and consistency of liquid supplements can be improved by relatively simple management practices at the feedlot such as ensuring tanks are emptied prior to re-filling and using separate delivery lines for different liquid ingredients.

Pencil shrink values applied to liquid supplements require review. Flat bottom tanks are inadequate storage method for liquid supplements.

Liquid supplement samples should be sent in duplicate and only to laboratories that can consistently achieve a 90% sensitivity testing level. Duplicate samples allow the feedlot to identify laboratory error by calculating the difference between the duplicate values.

Current flushing protocols of delivery tank may be insufficient to adequately remove monensin. Supplement manufactures supplying antibiotic free products may require the use of designated delivery tanks to avoid risk of contamination and residue detection.

Feedlots should consider undertaking supplement sampling and analysis during use from storage tanks to identify changes in nutrient concentration.

Mixing methods of aeration or re-circulation do not improve consistency of liquid supplements during storage period.

9. Future research and recommendations

The findings from this project identifies the following further research and recommendations:

- Only use laboratories that can consistently achieve testing sensitivity of 90%
- Considerations for feedlot industry QA systems:
  - Single dedicated lines for supplements with veterinary feed additives are utilised when adding to batching systems or mixers.
  - Maintaining proper inventory control for liquid supplements on an all-in-all out basis and are not refilling partially empty tanks.
  - Maintaining a liquid supplement testing program in consultation nutritionist.
- Review flushing protocols of liquid supplement delivery tanks.

10. References

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Herman, T. and Behnke, K. (1994). Testing mixer performance. MF-1172. Kansas State University Agricultural Experiment Station and Cooperative Extension Service Bulletin

NFAS (2022). National Feedlot Accreditation Scheme. Rules and standards of accreditation. September Edition. AUS-MEAT.

11. Appendix

11.1 Supplement manufacturer survey questionnaire

SUPPLEMENT MANUFACTURE SURVEY June2023 v5

Supplement Manufacturer Information			
Manufacturer Name:			
Head Office Address:			
Technical / Sales Manager:			
Contact Details:	Mobile:		
	Email:		
Survey Date:			
Completion Method:	☐ By Phone	☐ On-Site	
Number of Sites & Location			
Type of Manufacture: Liquid, Dry, Combination, Multi-Spp			
Supplement Manufacturer QA			
QA Programs:			
Formulation Software:			
Batching Software:			
Carrier Testing: Nutrients, heavy metals, physical characteristics (particle size, Brix, density)			
Modification of Carrier: Particle size, adjust Brix, adjust density			
Copy of Orders for Medicated Feed & S4 Antimicrobials		Yes ☐	No ☐
Ingredient Sampling / Testing	Yes ☐	No ☐	
Frequency of Sampling / Testing			
Testing of ionophores / medications	Yes ☐	No ☐	
Frequency of Testing ionoph/med			
Access of Testing Data*	Yes ☐	No ☐	
Storage Duration of Samples			
Use of antioxidants	Yes ☐	No ☐	
Use of anti-microbials	Yes ☐	No ☐	

Antibiotic Free Manufacturing Line:	Yes	☐	No	☐
Flushing Protocol:	Yes	☐	No	☐
Assess Mixing Variation:	Yes	☐	No	☐
CV Target %:				

Supplement Manufacturing Recommendations				
Formulation:	Yes	☐	No	☐
Storage Period:	Yes	☐	No	☐
Usage Rate:	Yes	☐	No	☐
Storage System:	Yes	☐	No	☐

Supplement Manufacturing Recommendations Dry		
	Pellet	Meal
Dry Matter Range %		
Limestone % Max		
Carrier % Min		
Carrier Preference		
Urea % Max		
Combined Urea & Am Sulphate Max %		
Fat % Min / Max		
Protein Meal % Min / Max		
Other		
Other		
Supplement Manufacturing Recommendations Molasses		
	Blend	Suspension
Dry Matter Range %		
Limestone Max %		
Carrier % Min		
Urea % Max		
Crude Protein % Max		
Combined Salt % Max		
Magnesium Carbonate % Max		
Suspending Agent Yes/No		
Particle Size of Medications		
Particle Size of Insoluble Raw Materials		
Total % of Insoluble Raw Materials		

Supplement Manufacturer Additives & Adverse Experience	
Number of Different Additives on Site	
Number of Monensin Products on Site	
Preparation of Additives (suspend monensin)	
Has Manufacturer Experienced Adverse Supplement Issue at Site:	Yes ☐ No ☐
Has Manufacture Experienced Adverse Supplement Issue During Transport	Yes ☐ No ☐
Has Manufacture Experienced Adverse Supplement Issue at Feedlot	Yes ☐ No ☐

11.2 Feedlot supplement survey questionnaire

FEEDLOT SUPPLEMENT SURVEY **May2023 v5**

Feedlot & Supplement Supplier Information	
Feedlot Name:	
Feedlot Address:	
Owner / Manager:	
Contact Details:	Mobile:
	Email:
Survey Date:	
Completion Method:	☐ By Phone ☐ On-Site
Permission for Photos:	☐ Yes ☐ No
Supplement Supplier:	
Manufacturing Site:	
Supplier Contact Details:	Mobile:
	Email:

Supplement Type	
Dry Concentrates	**Liquid**
Meal ☐ Pellet ☐	Suspension ☐ Solution ☐
Carrier Type:	Limestone inclusion %
Carrier Processed:	
Carrier Bulk Density (kg/m^3):	

Bulk ☐ Bags ☐	
Load Size Received (t):	Load Size Received (t):
Daily Usage (t):	Daily Usage (t):

Supplement Infrastructure	
Storage Method: Bulk in commodity shed, bags, tank	
Method of Loading into Mixer: Bucket, bag, conveyor, auger, pump	
Type of pump:	
Loading System:	Direct to Mixer ☐ Batch Box ☐ Batching ☐
Plumbing Material: Poly, flexible hose, steel piping	
Plumbing lines:	Shared ☐ Seperate ☐

Supplement Usage	
Inclusion Level % in Finisher:	
Supplements in Use:	Single ☐ Multiple ☐
Supplement Inclusion Changed for Different Diets:	Yes ☐ No ☐
Usage (t/day):	
Storage Capacity (t):	
Number of Tanks / Storages:	
Turn-over Rate (days):	
Conduct Monthly Stocktake:	Yes ☐ No ☐
Supplement Shrink %:	
Cause of Shrink: Dust, loading error, leaks, spills, drips	

Feed Additives in Use	Liquid supplement prepared by licensed feed manufacturer	Dry Supplement prepared by a licensed feed manufacturer	Direct addition by vet sale of concentrated S4 medicine into feed on feedlot (specify route e.g., top dress vs. mixer addition vs. micromachine)	S4 medicine from vet sale extended on-site in carrier on feedlot prior to feed addition (specify route top dress vs. mixer addition).
1. Barbermycin				
2. Monensin				
3. Lasalocid				
4. Salinomycin				
5. Narasin				
6. Tylosin				

7. Virginiamycin				
8. Chlortetracycline				
9. Oxytetracycline				
10.				
11.				

Do you receive prepared supplements with S4 antimicrobials from a licensed feed manufacturer (e.g., dry or liquid supplement manufacturers)	Yes ☐	No ☐
a. If Yes, is a copy of veterinary prescription supplied provided by the Vet to the Feedlot (Note whilst it is a requirement of the vet to sign and keep a copy of the script and provide the original prescription to the licensed feed manufacturer , the requirement to provide to the feedlot varies between states)	Yes ☐	No ☐
b. If Yes, are Instructions provided regarding how to measure and combine an S4 medicine with food to administer to the animals (Y/N)	Yes ☐	No ☐
c. If Yes, are Instructions to clean any residue from the medicine or feed from any equipment used to administer it to the animals.	Yes ☐	No ☐
Do you receive S4 medicine to be mixed with feed on farm e.g., CTC/OTC (direct supply of medicine to farm by vet).	Yes ☐	No ☐
a. If Yes, is a copy of veterinary prescription supplied provided by the Vet to the Feedlot (Y/N) (Note whilst it is a requirement of the vet to sign and keep the original of the script, the requirement to provide a copy to the feedlot varies between states)	Yes ☐	No ☐
b. If Yes, are instructions provided regarding how to measure and combine an S4 medicine with food to administer to the animals	Yes ☐	No ☐
c. If Yes, are instructions to clean any residue from the medicine or feed from any equipment used to administer it to the animals.	Yes ☐	No ☐

Liquid Supplement Composition

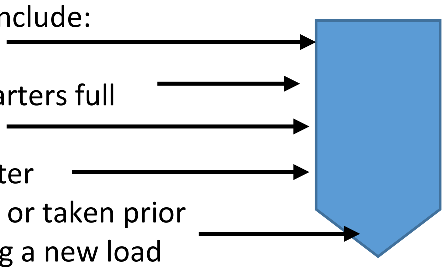
Carrier %:	
Dry Matter %:	
Bulk Density kg/m^3:	
Ingredients to Promote Ease of Use: Vegetable Oil/Surfactant/Emulsifier	

Liquid Supplement Delivery	
Distance from Supplier (km):	
Transport Contract:	Specific ☐ Multi Use ☐
Transport Capacity (t)	
Transport Configuration	Single ☐ B-Double ☐
	B-Triple ☐ Road Train: ☐
	Full Load ☐ Split Load ☐

Liquid Supplement Storage	
Number of Storage Tanks:	
Storage Type: IBC, flat bottom tank, cone bottom tank, re-purposed storage	
Construction Material: Poly, fiberglass, steel	
Volume (m^3)	
Storage Capacity (t):	
Fitted with Mixing System:	Yes ☐ No ☐
Considered Mixing System:	Yes ☐ No ☐
Method of Mixing: Air, Pump, Mechanical mixer, transfer	
Hours of Mixing:	
Frequency of Operation per Day/Week	
Supplement QA & Risk Management	
Samples Collected on Delivery	Yes ☐ No ☐
Frequency of Sampling:	
Storage Duration of Samples (mths):	
Sample Storage Method:	
Formal Assessment: Colour, aroma, density	
Formal Testing of Supplement:	Yes ☐ No ☐
Physical & Nutrient Components Tested: Dry matter, minerals, additives, ash	
Analytical Lab Used:	
Storage Cleaned	Yes ☐ No ☐
Frequency of Cleaning	
Method of Cleaning	
Sufficient Storage for Complete Emptying Before Re-Filling:	
Has Feedlot Experienced an Issue with Supplement?	Yes ☐ No ☐
Description & solution Issue:	

11.3 Feedlot supplement sampling protocol & labelling instructions

LIQUID SUPPLEMENT SAMPLING PROTOCOL ***v4 July2023***

OBJECTIVE:	To assess liquid supplement composition changes within the supplement storage tank over the time it takes to use a newly received delivery.
SAMPLING SITE:	Where possible, samples to be taken from the tank at ground level, at delivery point to tank (inlet pipe, hose). As required, use a small clean bucket to capture supplement from pipe. Collect 75ml sample from bucket. Avoid taking samples at height. DO NOT enter the mixer to retrieve sample.
SAMPLING FREQUENCY:	After designated tank receives new load of supplement, take one sample directly from delivery truck. Subsequent samples taken at five (5) intervals representing tank capacity during usage. Tank capacity intervals include: - 95% or full - 75% or three quarters full - 50% or half - 25% or one quarter - 5% nearly empty or taken prior to receiving a new load 
SAMPLING QUANTITY:	Require 75ml in duplicate (2 samples from each interval including two samples from delivery truck) placed in plastic jars with screw top lids. Fill jars to top of neck. When sampling is completed, feedlot will have 12 jars (2 from delivery tank, 10 from tank usage intervals).
SAMPLE LABELING:	Wipe clean and dry plastic jars after collection / filling. Use sticky labels provided. Record date at which samples were collected on the label. Stick appropriate label on jars following the legend provided. - Delivery = Wallaby - 95% = Wombat - 75% = Koala - 50% = Possum - 25% = Echidna - 5% = Platypus Duplicate 1 = Galah = first sample at interval, Duplicate 2 = Rosella = second sample at interval. Coding for samples required to ensure laboratory conducting the testing does not know order of sampling.
SAMPLE STORAGE:	Store samples in cupboard. Out of direct sunlight. Hold samples onsite until all 12 samples are collected.
SAMPLE POSTAGE:	Place each sample in a zip lock bag to collect any spillage that may occur during postage. Pack samples in box and post them to: **Mr. Ivan Ward**

	PO Box 2090 **Graceville East, Qld 4075** Please notify Robert Lawrence by email (rlawrence@iapnutrition.com) when samples posted

SAMPLE LABELS

Feedlot Name: Sample Date: Sample ID: WALLABY / Galah	Feedlot Name: Sample Date: Sample ID: WALLABY / Rosella
Feedlot Name: Sample Date: Sample ID: WOMBAT / Galah	Feedlot Name: Sample Date: Sample ID: WOMBAT / Rosella
Feedlot Name: Sample Date: Sample ID: KOALA / Galah	Feedlot Name: Sample Date: Sample ID: KOALA / Rosella
Feedlot Name: Sample Date: Sample ID: POSSUM / Galah	Feedlot Name: Sample Date: Sample ID: POSSUM / Rosella
Feedlot Name: Sample Date: Sample ID: ECHIDNA / Galah	Feedlot Name: Sample Date: Sample ID: ECHIDNA / Rosella
Feedlot Name: Sample Date: Sample ID: PLATYPUS / Galah	Feedlot Name: Sample Date: Sample ID: PLATYPUS / Rosella

11.4 Result summary from supplement analysis

FEEDLOT SUPPLEMENT ASSESSMENT		
Feedlot #	10	
Feedlot Name		
State	QLD	
Size Category	L	
Reviewer	JD	
Supplement Supplier	BM	
Survey Complete		
Samples received - PO Box	20-11-2023	20-11-2023
Samples Collected		
Samples Received	24-11-2023	24-11-2023
Results Issued	30-11-2023	30-11-2023
Certificate #	B1426647-A [R00] B1426647-B [R00]	B1426665-A [R00] B1426665-B [R00]

Monensin	Galah	Rosella
Delivery (Wallaby)	198	195
95 (Wombat)	202	209
75 (Koala)	205	218
50 (Possum)	208	166
25 (Echidna)	196	181
5 (Platypus)	202	203

Analysis Moisture (%)		
Delivery (Wallaby)	33.3	33.1
95 (Wombat)	33.5	33.4
75 (Koala)	33.8	33.2
50 (Possum)	33.9	33.3
25 (Echidna)	33.0	32.8
5 (Platypus)	33.1	32.5

Analysis Ash (g/100g)		
Delivery (Wallaby)	32.5	32.9
95 (Wombat)	32.2	32.8
75 (Koala)	32.4	32.9
50 (Possum)	32.3	33.0
25 (Echidna)	32.1	33.1
5 (Platypus)	33.2	34.6

Analysis Zinc (mg/kg)		
Delivery (Wallaby)	1990	1520
95 (Wombat)	1970	1510
75 (Koala)	1960	1510
50 (Possum)	1890	1550
25 (Echidna)	1900	1510
5 (Platypus)	1890	1520

Analysis Calcium (g/kg)		
Delivery (Wallaby)	99.5	92.3
95 (Wombat)	104.0	101.0
75 (Koala)	88.3	101.0
50 (Possum)	92.3	93.3
25 (Echidna)	95.0	107.0
5 (Platypus)	109.0	108.0

Analysis Copper (mg/kg)		
Delivery (Wallaby)	149	116
95 (Wombat)	119	118
75 (Koala)	119	116
50 (Possum)	117	118
25 (Echidna)	114	119
5 (Platypus)	116	120

Analysis Potassium (g/kg)		
Delivery (Wallaby)	16.6	16.2
95 (Wombat)	16.7	15.3
75 (Koala)	16.7	15.8
50 (Possum)	16.0	16.0
25 (Echidna)	16.0	15.8
5 (Platypus)	15.9	16.4