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Full Research Project Final Report

- This report must be a stand-alone report, *i.e.*, must be complete in and of itself. Scientific articles or other publications cannot be substituted for the report.
- One electronic copy and one signed original copy are to be forwarded to the lead funding agency on or before the due date as per the investment agreement.
- A detailed, signed income and expenditure statement incurred during the entire funding period of the project must be submitted along with this report. Revenues should be identified by funder, if applicable. Expenditures should be classified into the following categories: personnel; travel; capital assets; supplies; communication, dissemination and linkage; and overhead (if applicable).
- For any questions regarding the preparation and submission of this report, please contact ACIDF

Section A: Project overview

1. Project number: **2015C028R**
2. Project title: **Development and commercialization of aerobic digestion of poultry manure to produce bio-active fertilizers.**
3. Research team leader: **Marc Legault**
4. Research team leader's organisation: **Alberta Agriculture and Forestry**
5. Project start date: **2015 September 1**
6. Project completion date: **2017 December 1**
7. Project final report date: **2018 January 31**

Section B: Non-technical summary (max 1 page)

Provide a summary of the project results which could be used by the funders for communication to industry stakeholders (e.g., producers, processors, retailers, extension personnel, etc.) and/or the general public. This summary should give a brief background as to why the project was carried out, what were the principal outcomes and key messages, how these outcomes and key messages will advance the agricultural sector, how they will impact industry stakeholders and/or consumers, and what are the economic benefits for the industry.

Alberta Poultry producers contribute to the Albertan economy by creating employment and revenue. However, inherent to poultry production is manure by-product that is viewed as a low value liability. This project provided an opportunity to support Alberta Agriculture and Forestry's commitment to develop technologies to transform agricultural wastes into value-added products. The fermentation of poultry manure to produce non-pathogenic biologically active plant nutrient solutions not only met this

challenge but it also demonstrated how to use these value-added process solutions in a greenhouse setting while recycling all water. Poultry manure was chosen due its high nitrogen to carbon ratio and it typically contains less fibrous bedding materials than other manures. Bedding materials were problematic for initial efforts however as the project progressed these seemingly problematic materials were used to produce an additional value-added solids product.

The goal of this project was to enhance the value of poultry manure by producing biologically active fertilizers through aerobic digestion. A core accomplishment was the successful development of a fermentation process to produce non-pathogenic biologically active nutrient solutions; which could be applied to other agri-food organic wastes. The project successfully developed greenhouse methods for using these biologically active nutrient-rich solutions to safely grow food. Plant biomass and produce were observed to be healthy, particularly for heavy feeders such as the *Brassica* family, tomatoes and squash.

The hypothesized mechanisms associated with biologically active nutrient solutions are as follows. Microbial biomass acts as a slow-release source of nutrients that complement the manure-derived nutrients. Microorganism activity can suppress or counter soil pathogens. These solutions can improve soil fertility, thus leading to healthier soils which in turn promotes healthy plants.

The project furthered a sustainability goal by demonstrating waste is actually a resource. Future efforts could involve the co-fermentation of liquid and solid organic wastes. The goal is to destroy pathogens and increase the non-pathogenic bacteria cell count; the greater the cell counts of non-pathogenic microorganisms, the higher the nutrient content of the manure-derived solutions.

With regard to soluble carbon, the fermentation broth's soluble carbon significantly declines within days. Increasing the soluble carbon by adding methanol, increased the bacteria cell count a thousand fold. Therefore, co-fermenting manure with a soluble carbon waste stream would produce an ideal microorganism rich product while processing or disposing two waste streams.

Organic certification of the process would be a strong economic incentive for industry adoption. However, accredited organic authorities need to vet the process with regard to organic certification.

An exploratory economic analysis **[not including facility, overhead and utilities costs]** at the 1,000 liter scale suggests it is economically feasible to pursue this technology (or more prudently seek organic certification and /or scale up development). However, the 1,000 liter scale is likely inadequate for industry. Scaling up the technology may involve working with industry to use commercially available equipment to develop robust support equipment for feeding, dosing and harvesting the bioreactor. Attachment 1 highlights the economic analysis (Net Present Value, Benefit-Cost Ratio) assumptions and data.

Regulatory Authorities are required to define and assess applicable field and greenhouse regulations. Since the product is neither manure nor is it compost (nor compost tea).

Section C: Project details

1. Project team (max ½ page)

Describe the contribution of each member of the R&D team to the functioning of the project. Also describe any changes to the team which occurred over the course of the project.

Emmanuel Laate, Project Economist, provided the report's economic analysis.

Marc Legault, Project Manager and Engineer, developed the processes, data analysis, documentation and coordinated field trials.

Project Team Changes:

i. Dr. Nick Savidov left Alberta Agriculture and Forestry prior to the project's commencement
ii. Yingli Wang, technician, as of the Fall of 2016, was no longer involved with the project

2. Background (max 1 page)

Describe the project background and include the related scientific and development work that has been completed to date by your team and/or others.

The project's fermentation technology is a refinement of aerobic digestion. Aerobic digestion was first used by Alberta Agriculture and Forestry in the development of aquaponics where fish manure is used to fertilize plants. Fish manure is inherently diluted by water although strategies can be developed to concentrate the manure; the percent dry matter likely remains far less than other livestock manures. The advantage to digesting manures containing higher percent dry matter is that more concentrated nutrient solutions can be produced. A technical challenge for the project was to ferment as much manure as feasible per batch in order to produce concentrated biologically active nutrient solutions.

To facilitate industry adoption of the technology, this project focused on utilizing existing greenhouse equipment and techniques to demonstrate the use of these biologically active nutrient solutions to safely grow greenhouse crops. The biological activity of these nutrient solutions differentiates them from traditional synthetic fertilizers; as a consequence greenhouse techniques had to be modified in order to use these solutions. The incorporation of water recycling also necessitated the modification of existing greenhouse practices to maintain a sufficient nutrient chemistry and to monitor for toxic buildups; in particular, sodium levels.

The fermentation of poultry manure to produce non-pathogenic biologically active nutrient solutions, involved two central principles of operation. One, the control of pH to induce thermophilic pasteurization conditions and avoid nutrient loss by: phosphate divalent cation precipitates and ammonia off-gassing. Secondly, the constant addition of relatively pure oxygen was done to ensure aerobic conditions in order to maintain an odorless microbial decomposition of organic matter.

Only organisms native to manure were used in this non-aseptic fermentation process. All solutions were analyzed for pathogens (*E. coli, Salmonella* and other fecal coliforms) and periodic aerobic plate counts (i.e. total number of aerobic organisms). An aliquot of solution was tested [a DNA scan] for

thirty root rot pathogens, which was confirmed to be free of all pathogens [see Attachment 2 for details]. The solutions can be considered "safer" than untreated manure with regard to animal and root pathogens.

Project development was achieved through two stages, first it was necessary to develop, characterize and optimize the fermentation process while simultaneously developing the greenhouse processes and building the support infrastructure.

Specific agricultural and greenhouse regulations with regard to chemistry and microbiology were difficult to determine since the product is neither manure nor compost. As is identified in this report, soil contamination / remediation and compost regulations were used as guidelines.

3. Objectives and deliverables (max 1 page)

State what the original objective(s) and expected deliverable(s) of the project were. Also describe any modifications to the objective(s) and deliverable(s) which occurred over the course of the project.

Objectives - Original Project Objectives Prior to Staff Changes

I. Characterize the fermentation process to ensure pathogen kill.
II. Optimize the fermentation process to yield stable nutrient product solutions.
III. Optimize the fermentation process to maximize the economic and nutrient value with regard to acid addition.

Deliverables - Original Project Deliverables Prior to Staff Changes

I. An economic assessment of:
    i. The financial suitability to use BANS to grow greenhouse crops
    ii. The cost to ferment poultry manure to produce BANS
II. A comprehensive "How to Manual" to describe in detail ARD's aerobic digestion technology to produce BANS, biologically active nutrient solutions
III. Standard Operating Procedure, SOP's, to:
    i. Ferment poultry manure to attain pathogen safe stable nutrient solutions
    ii. Store, maintain viability of and use BANS
    iii. Greenhouse production based on BANS as a source of plant nutrients
IV. Data on the potential accumulation of sodium and other minerals associated with recirculation of hydroponic solutions containing BANS
V. Data with regard to the impact of soil biology when using repetitive field BANS applications.
VI. Economic analysis of producing BANS from poultry manure for greenhouse production
VII. Final report

Objectives - Modifications with regard to staff changes

I. Produce safe and effective plant nutrient solutions from poultry manure
II. Develop a robust and industrialized manure fermentation technology
III. Facilitate industry adoption of the technology by:

    i. Demonstrating the use of solutions to grow greenhouse and field crops.

The project developed greenhouse and outdoor practices to use the nutrient solutions.

ii. Detailing process design including 'scale-up' considerations

Considerations for utilizing other manures were included in addition to scale up challenges.

iii. Providing detailed SOP's, batch records and data presentation

Process Control Forms for data capture were also developed.

Unless otherwise specified, the objectives of this project were successfully achieved.

Project Modification of Key Results: *the experimental use of reagents [ammonium hydroxide (to enhance microbial biomass), vinegar (an antifoam agent), methanol (a soluble carbon source) and iron sulphate (to enhance iron concentrations)] were added as the project progressed.*

Deliverables Modifications with regard to staff changes
Key Results Expected

I. A detailed characterization of the fermentation process including in-depth nutrient analysis to document the optimization path and process scale-up considerations

This characterization process will address both controlled and uncontrolled process variables.

Process variables controlled or manipulated	**Process variables NOT controlled nor manipulated to date**
i. pH control agents ii. pH setpoints iii. dO, dissolved oxygen iv. gas (oxygen) flowrate v. antifoam agents vi. % DM, amount of manure per batch vii. duration viii. agitation ix. mother liquor	i. manure feedstock variability project will try to process manures containing antibiotic residues ii. microbiology - the project will investigate: - if nutrient solutions impact the Rhizobium inoculation of legumes the continual use of mother liquor to optimize the fermentation iii. temperature

II. Industry trials and assessment of plant nutrient solutions derived from poultry manure
(The assistance from CARA, Chinook Applied Research Association was a modification)

III. Demonstrate and assess greenhouse strawberry production techniques using nutrient solutions derived from poultry manure. Echinacea raft and substrate culture will be trialed

The cost of microbiological characterization hindered the quantification of the microbial contribution to the nutrient profile ($12,000 for a single DNA scan to $40,000 to assess plant growth, bio-

stimulant properties and basic microbiology). Project did not address the microbiology impact on *Rhizobium* inoculation.

4. Research design and methodology (max 4 pages)

Describe and summarise the project design, methodology and methods of laboratory and statistical analysis that were actually used to carry out the project. Please provide sufficient detail to determine the experimental and statistical validity of the work and give reference to relevant literature where appropriate. For ease of evaluation, please structure this section according to the objectives cited above.

Subject matter experts were contracted whenever possible throughout the project. Contracts were established for chemical and microbial analysis and wastewater treatment options. This project is grateful for the help from the Oyen, AB producer research group, the Chinook Applied Research Organization (CARA) in providing soil health experiment planning and advice. Alberta Agriculture and Forestry's Food Safety and chemical analysis subject matter experts (OS Longman colleagues) greatly assisted in the interpretation of pathogen and chemical analysis data.

Producing over 3,000 liters of biologically-active nutrient solutions for greenhouse and field trials was the driving force of this project, while the objective was to chart and document the developmental work for each batch.

The project tried to focus on two economic (and technical) factors:

i. the more manure processed per batch, the better the potential economic returns
ii. the quicker the production the better the potential economic returns

The 'Trial and Error' method was used to investigate the variables given below:

Process variables controlled or manipulated	Process variables NOT controlled nor manipulated to date
i. pH control agents ii. pH setpoints iii. dO, dissolved oxygen iv. gas (oxygen) flowrate v. antifoam agents vi. % DM, amount of manure per batch vii. duration viii. agitation ix. mother liquor	i. manure feedstock variability project will try to process manures containing antibiotic residues ii. microbiology - the project will investigate: - if nutrient solutions impact the Rhizobium inoculation of legumes the continual use of mother liquor to optimize the fermentation iii. temperature

This methodology was utilized to determine the optimal solution harvest technique. The objective was to maximize the amount of solution per batch while minimizing solution dilution. The project contracted a wastewater company, ClearTech, to investigate the optimal flocculation agent for the process.

Harvesting the residual solids that originate from bedding materials became an objective as well as determining end-use applications for this solids product.

Objectives of the Project

I. Produce safe and effective plant nutrient solutions from poultry manure

Accredited 'Lab Houses' were contracted to assay the biologically active nutrient solutions. The effectiveness of the solutions to grow crops was qualitative, based upon the experience of qualified colleagues and visitors. Tree farm trials provided qualitative input. Solution safety was based upon the services of an accredited 'Microbiology Lab'; the most difficult pathogens to kill (*E. coli, Salmonella* and fecal coliforms) were chosen for investigation.

II. Develop a robust and industrialized manure fermentation technology

Poultry manure (complete with bedding, feathers, eggs, mites and miticides etc.) was added to water, followed by oxygen. Section C, subset 5, details the subsequent *"*Trial and error*"* methodology to characterize the fermentation (and harvest) trials, the greenhouse and field methods, to successfully utilize the biologically-active nutrient solutions.

III. Facilitate industry adoption of the technology by:

i. Demonstrating the use of these fertilizer solutions to grow greenhouse and field crops.

The solutions were trialed indoors and outdoors for three years. Greenhouse trials and a novel innovative outdoor market garden trial were conducted at Crop Diversification Center, CDC, North; both trials utilized the biologically active nutrient solutions while continuously recycling all water. A southern Albertan tree farmer trialed the solutions for three years. [At worst: the solutions are as effective as synthetic fertilizers.] The producer research group, CARA, is currently investigating the solutions' (in particular the microbiological) impact on soil health.

ii. Detailing process design including 'scale-up' considerations

Process challenges and scale-up considerations were recorded as they were observed on Fermentation Run sheets and Batch Records. Project presentations and webinars outlined the technology's successes, challenges and pitfalls as detailed below in Section 5.

Much effort was directed in developing a robust harvest process involving equipment and pH manipulation. Overcoming the problematic lignocellulosic bedding materials for decant (settling) based harvest strategies was a challenge.

iii. Providing detailed SOP's, batch records and data presentation

Standard Operating Procedures (SOPs) and document control forms (Fermentation Run and Batch Record templates) used to capture data, are attached to this report [see Attachment 3 for Details].

The focus of this project was to help industry adopt this technology. In addition to the above documentation, the data presentation consists of fourteen fermentation and nutrient graph sets - one set per fermentation trial, (due to the cost associated with Trial 2, the second trial without pH control, does not have a nutrient profile like Trial 1).

The graphs are near continuous depictions of data along the fermentation runtime from t = 0 to harvest and often includes Quarantine Tank data. The graphs depict Temperature, Dissolved Oxygen and pH vs Time (in Hours and Days) using ten-minute graphing intervals. The graph title provides the pH and +/- setpoints, the date and the acid control agent other than phosphoric acid.

Each graph has a Text Box in order to readily compare:

i. Volume of Mother of Liquor used, if any
ii. Broth Volumes at t = 0, at harvest and amount of decant harvested
iii. Bioreactor loading i.e. % dry matter and number of 20L manure pails added
iv. The type and amount of acid and base used to control the pH

Process upsets (power loss, foam outs, etc.) and reagent additions (antifoam agents, methanol, vinegar, ammonium aliquots, iron supplements, etc.) are all denoted with respect to time on each Fermentation Runtime graph. The nutrient graphs illustrate the direct relationship between foam loss and nutrient loss.

The ideal way to display the graphs was to place the nutrient data graphs below the fermentation runtime graphs so as to provide a visual correlation between fermentation events and the corresponding nutrient profile graphs. Each run (except Trial 2) has either four or six corresponding nutrient profile graphs; the discrepancy is for extractable metals when the results were reported as less-than-values and consequently, are of little benefit.

NOTE: For future chemical and possibly microbial analysis, all fermentation runs (including the aborted ones) have retained broth samples stored at 2 - 5°C; these samples exist all along the runtime including the final harvest. Although Trials 15 and 16 were terminated, retained samples exist as there may be cellulose degrading organisms present.

Project activities with regard to greenhouse materials, methods and observations are given in Appendix C Greenhouse Methods and Observations.

Project activities with regard to engineering challenges and considerations including original apparatus are given in Appendix D Engineering Challenges and Considerations.

5. Results, discussion and conclusions (max 8 pages)

Present the project results and discuss their implications. Discuss any variance between expected targets and those achieved. Highlight the innovative, unique nature of the new knowledge generated. Describe implications of this knowledge for the advancement of agricultural science. For ease of evaluation, please structure this section according to the objectives cited above.
NB: Tables, graphs, manuscripts, etc., may be included as appendices to this report.

The project demonstrated the feasibility of fermenting organic wastes to produce biologically active nutrient solutions. The pH was controlled to maximize the nutrient yield (from organic decomposition) and more importantly near-guarantees the thermophilic step to kill pathogens. Competition and predation from other organisms are other hurdles for pathogen survival within the solutions. The goal was to destroy pathogens and increase the non-pathogenic microorganisms that are believed to be key to the nutrient-rich solutions' success.

The project attempted to focus on two economic (and technical) factors:

i. The more manure processed per batch the better the potential economic returns.
   The more manure per batch the more concentrated the nutrient solution, and therefore, the more favourable the economics of transport becomes.

ii. The quicker the product is produced, the better the potential economic returns.

Discussion of process variables listed in Section C, subsection 4 Research design and methodology:

Process variables controlled or manipulated:

i.) pH Control Agents

The project trialed various pH control agents to determine the advantages and disadvantages of each; [See Appendix B, Table 1 pH Control Agents for details]. When using sulphuric acid it appears "foam-outs" occur at the end of the run, whereas when using phosphoric acid, foam losses occur in the beginning. A blend of 1 part sulphuric acid to 4 to 5 parts phosphoric acid appear to be the most favourable acid agent. Nitric acid produced the most manageable foam (easily collapsed on itself) but nitric acid usage was discontinued since the buildup of nitrate caused the solutions to be over diluted in order to attain safe nitrogen levels. Unlike ammonium, nitrate ions do not appear to be incorporated into microbial biomass thus leading its accumulation. A 100 fold or greater dilution was required to have safe ammonium and nitrate levels; this caused an over dilution of trace elements, especially iron. Additionally, nitric acid is a non-permitted substance for Canadian organic compliance. The preferred base control agent was 2 parts potassium hydroxide (caustic potash) to 1 part ammonium hydroxide.

In general terms, when using phosphoric acid and ammonium hydroxide as pH control agents, ammonium and phosphate ions (as expected) both tended to increase while calcium, magnesium, iron and manganese ions showed the classical asymptotic loss curve (steep decline followed by modestly stable value). When nitric acid is the control agent, ammonium ions tended to climbed in value (due to in part to ammonium hydroxide as the base agent) whereas phosphate ions (which was not added)

was relatively constant in two cases but gradually declined to less than half in the third case, suggesting loss due to precipitation.

ii.) pH Setpoints

The optimal fermentation pH setpoint is a tradeoff between:

i. near neutral pH 7 for maximum thermophilic pathogen elimination temperatures

ii. lower pH's near or below pH 6.6 to prevent nutrient loss from ammonia off gassing and phosphate cation precipitates

Two pH setpoints may be an option; the initial pH setpoint (near neutral pH) would induce thermophilic conditions. After the pathogen elimination step, the pH setpoint could be lowered to maximize nutrient stability.

iii.) Dissolved Oxygen, iv.) Gas Flow Rate v.) Antifoam Agents

The bioreactor utilized manual control of oxygen and antifoam.

The cause and effect inter-reactions between foam control, temperature, agitation, % dry matter (%DM**)** and oxygen flowrate were routinely noted. The more manure fed to the bioreactor (i.e. higher % DM), the more oxygen and agitation required to support metabolic activity. The greater the metabolic activity, the greater the temperature, and the higher the temperature, the more likely microorganisms will die. Upon death, cells rupture and release proteins into the broth, and the more protein in the broth, the more likely foaming will occur. Also, the higher the oxygen flow rate, the greater the likelihood of foam formation. Foam also insulates the open bioreactor from heat loss. As such, foam loss may lead to heat loss. However, if the process utilized air instead of relatively pure oxygen, the higher flow rates (to attain the same oxygen level) would induce more foaming and temperature stripping.

Antifoam is a barrier to oxygen transfer since oxygen bubbles, cells and instrumentation become coated in antifoam consequently it has a great negative impact on oxygen transfer to cells. The less oxygen to cells, the lower the metabolic activity, thus leading to an immediate decrease in broth temperature (loss of foam insulation can contribute to heat loss). All fermentation temperature graph lines are 'saw tooth'; this highlights the negative impact of antifoam on temperature. Earlier runs have handwritten arrows to denote antifoam addition times that correspond to an immediate temperature loss. Many temperature graphs have an overall cyclic (sinusoidal) aspect over the fermentation runtime; it has been speculated that this may correspond to the rise and fall of different microbial populations.

vi.) % Dry Matter (% DM), amount of manure per batch

The more manure per batch, the more concentrated the resulting nutrient solution, and as such, the more favourable the economics of transport becomes. To this end, each batch saw an increase in manure loading; initial batches were near 4% DM while dry matter increased up to 15% DM (Trial

11), which was far too much for the given agitation assembly (3 hp motor and 2 aggressive impellors). At 15% DM, the broth was not homogenous in temperature and possibly pH (the pH probe in question may have been failing), the oxygen distribution was also hampered. Based on temperature uniformity, 13 % DM proved to be the maximum load for the given bioreactor.

NOTE: *Temperature measurement is considerably more robust than pH; consequently there's less likelihood that conflicting temperature measurements are from failing temperature sensors. Constant exposure to thermophilic temperatures in addition to abrasion from suspended grit was extremely problematic for pH probes.*

For the given bioreactor assembly, 10 to 12% DM is ideal especially if the manure contains bedding. Bedding material is required for efficient nutrient solution harvest.

vii.) Duration

The quicker the product is produced the better the potential economic returns. For this reason, the fermentation runtimes (reaction times) were cautiously decreased. Initial reaction times for the first few runs which also had lower % DM loading were quite long at thirty days or more. Labour logistics and harvest equipment breakdowns were responsible for one run, exceeding forty days. Towards the end of the project, reaction times decreased as the quantity of manure fed to the bioreactor increased. A conservative processing time of 12 days was used to generate economic assumptions. Therefore, it may be feasible to shorten the fermentation to as little as 7 days or less.

viii.) Agitation

Aggressive agitation enhances oxygen transfer, but the entrainment of abrasive grit is a negative consequence. Scale-up contemplations should consider the use of aggressive agitation assemblies that incorporate inline aeration; (it is recommended to substitute the aeration component with oxygen). Turborator™ is one such technology [http://www.mgdprocess.com/turborator.html].

ix.) Mother Liquor (includes discussion for process variables NOT controlled)

Typically, the inclusion of Mother Liquor, [a broth aliquot from prior run(s)], serves to hasten the thermophilic step and, possibly, the rate of mineralization, (the microbial decomposition of organic matter into plant available nutrients). Trial 6 (see Attachment 4 Trial 6 for details) may highlight a potential drawback of using too much Mother Liquor. This run had 40% Mother Liquor where it appears that nitrification (the microbial conversion of NH_4 to NO_3) may have caused the broth to become acidic; an increase in NO_3 and a decline in NH_4, supports the likelihood of nitrification occurring even though the pH was low for the nitrification due to *Nitrosomonas* and *Nitrobacter* bacteria. However, other chemotrophic organisms could also be responsible for nitrification induction. Nitrification is a negative impact since the end product nitrate is an unstable nitrogen compound with regard to shelf life. The Biological Oxygen Demand (BOD) also stalled at a high BOD point, suggesting a decrease in overall microbial activity.

Hypothesis: The broth was NH_4 rich due to Mother Liquor from Trial 5. Upon startup of Trial 6 as the broth became oxygenated. The nitrification process was initiated, which led to an

acidification from the conversion of NH_4 into NO_2 then NO_3. It is likely that the nitrifying organisms were present in the manure and not the mother liquor since typical N-cycle bacteria would have been killed from Trial 5's thermophilic step.

Process variables NOT controlled nor manipulated to date

i.) Manure feedstock variability

The project utilized two types of layer manure; one contained bedding materials and the other did not. Neither contained antibiotic residues, and consequently, the project did not run trials with manures having antibiotic residues. However, miticides and other dusting powders could be sources of contamination.

iii.) Temperature

Broth temperature was a direct result of fermentation conditions. An ideal run has an inherent thermophilic step that serves to pasteurize, if not, eliminate pathogens. Temperature increases are due to metabolic activity; often a loss of temperature signifies a decrease of activity. Most runs easily met the minimum compost pasteurization requirement of 3 days at 55°C [Guidelines for Compost Quality PN 1340, Canadian Council of Ministers of the Environment, 2005].

Reagent additions such as iron sulfate, vinegar, methanol and ammonium hydroxide were experimented with as aliquots. The goal was to further enhance the solutions' nutrients and / or the microbiology biomass [see Appendix B, Table 2 Reagents for details]. Towards the end of the project, the biologically active nutrient solutions were complete; no other plant supplements were required. The nutrients from poultry manure combined with the pH control agent additions seemed to yield near-balanced solutions for plant growth [see Attachment 5 for details].

Initially, foliar applications of iron were required for many plants. After ferrous sulphate heptahydrate was added to the fermentation broth, iron supplementation was no longer required, thus suggesting this iron continued to be plant available, or microbial activity mediated iron uptake. Supplementing the fermentations with iron does appear to create a unique foam; this foam can easily be 4 to 6 inches thick along the bioreactor walls. It is suspected that the positive ("+") iron charges are bridging with the cells' negative ("-") charges. Foam must be washed back into the bioreactor – as *E. coli* could be shielded from heat by the foam and thereby contaminate the broth, which is especially detrimental once past the thermophilic phase.

Methanol additions were first tested at a 150 L scale using an oxygenated post-thermophilic (approximately 50°C high point) aliquot from Trial 6. Once the culture was fed 3.78 L of methanol, the temperature went up immediately by over one degree Celsius. Three days later, the culture was fed sugar (the amount to saturate a 3.78 L solution at ambient temperature). This had no effect, other than temporally decreasing the dissolved oxygen, suggesting an increase in metabolic activity was observed but it was not enough to have an effect on the temperature. The following day the greenhouse emitted a yeasty, doughy smell, suggesting yeast was cultured. The 50°C broth aliquot likely contained yeast, since 50°C is not lethal to yeast survival.

The solutions typically have biological activity up to and at times beyond 10^9 +/- 10^2 cells per mL. The addition of methanol yields a thousand fold increase in cell density [see Attachment 4 Trial 11 for details].

Fermentation Observations

The technology's robustness was repeatedly highlighted; loss of: power, oxygen, agitation and /or pH control continued to yield seemingly effective nutrient solutions. At times, broth pockets were so anaerobic that characteristic foul odours were noted; reprocessing the batch appeared to yield nutrient solutions of comparable quality. Without doubt, some bacteria are more plant beneficial than others, just like some batches must be better than others. However, the noted robustness likely stems from the fact that all microorganisms can become readily available nutrients for plant uptake.

The project consisted of sixteen fermentation runs including two "baseline runs" without pH control; all runs were oxygenated [see Attachment 4 for Fermentation and Nutrient Graphs]. All other runs had pH control; four runs used phosphoric acid, three runs used nitric acid and one run used sulphuric acid. The six remaining runs used a combination of phosphoric and sulphuric acid to control pH. The base pH agent was ammonium hydroxide for the first eight pH control runs. Trial 11 used potassium hydroxide as the base agent – manual aliquots of ammonium hydroxide were often added to supplement the ammonium concentration in order to spur metabolic activity and increase microbial biomass. The five remaining runs used a combination of potassium and ammonium hydroxide to raise pH levels.

Without pH control [the first two fermentation trials], the cultures quickly became 'self-limiting' from attaining a pH of near 9 and consequently, they did not attain thermophilic pasteurization temperatures; the broths barely attained 40°C [see Attachment 4 Trial 1 and 2 Fermentation Runtime graphs]. The Nutrient Profile for Trial 1 is a text book example of nutrient loss due to high pH. Significant nutrient losses were observed due to ammonia off-gassing and likely irreversible phosphate-divalent cation precipitates [see Attachment 4 Trial 1 graphs for details].

However, aside from the loss of nutrients due to high pH [see Attachment 4 Trials 1, 11], nutrient concentrations increase with decreasing pH at time of harvest [see Attachment 4 Trials 13, 14] and foam loss is a nutrient loss [see Attachment 4 Trials 9, 10 and 11], no other firm rules could be determined.

Trial 11 [see Attachment 4 Trial 11] used sulphuric acid and caustic potash for pH control; aliquots of ammonium hydroxide were added (to increase microbial biomass). In this case ammonium rose, phosphate ions were modestly stable, calcium and magnesium ions oscillated but rose overall whereas iron and manganese ions were approximately stable. In hindsight, a sulphuric acid run (and possibly a series of runs) should have been repeated.

The last three successful runs (see Attachment 4 Trials 12, 13 and 14) used the aforementioned combinations for pH control; it is considerably more difficult to identify trends since vinegar and iron were added to the broth. Ammonium ions generally rose in concentration over time whereas phosphate ions were steady for two runs. Trial 14 was very uncharacteristic, since phosphate asymptotically plummeted 30 fold from 800 to 250 ppm. Calcium ions for these runs tended to be

stable, however, Trial 12 had a rather large cyclic oscillations of calcium, magnesium and iron ions. The fact that Total Hardness (as calcium carbonate) also followed these oscillations, this suggests chemistry, rather than microbiology, is responsible.

The last two fermentations (Trial 15 and 16) stalled and did not attain the thermophilic pathogen elimination step; likely due to residual negative by-product(s) from the use of vinegar as an antifoam agent. Trial 15 and 16 had the greatest viscosity, observed by the fact that the limestone grit [1/16" to 1/8" diameter] remained suspended for days when broth aliquots were allowed to settle.

Runs 12, 13 and 14 successfully used vinegar as a means to delay the need for antifoam. The delay in using antifoam allowed the cultures to attain temperatures near and slightly above 70°C. Eventually, the cultures required antifoam, and as such, the temperatures decreased immediately. The discarded trials 15 and 16 that contained Mother Liquor from trials 12, 13 and 14 likely stalled from negative by-products from the vinegar.

It was observed that pathogens were killed due to residence time in the quarantine tank (see Trial 11 Fermentation graph).This elimination was likely due to competition with and predation by other organisms. This may be an additional means for pathogen reduction or elimination. Trial 11 obtained a thousand-fold more cells by supplementing the culture's soluble carbon with methanol. Pathogen elimination due to competition would likely still occur for solutions having lower cell counts (from not being fed methanol); however it may take longer. Prior to automatic pH control when acid was periodically added to the broth manually, *E. coli* elimination was routinely observed even though the broth did not attain the 3 days at 55°C pasteurization threshold. As previously mentioned, this elimination was likely from competition and predation.

Trial 7 used nitric acid as the control agent and the oxygen addition assembly was modified to deliver more oxygen in a controlled manner; the corresponding thermophilic step had a significant temperature rise rate and high point. When using nitric acid in comparison to phosphoric acid, the baseline temperature increased from 50°C to 60°C. It is suspected that the temperature rise was most likely due to oxygen enhancement. The run had a near 5 day plateau at 65°C, and the broth went from 10°C to over 40°C in less than 24 hours.

The project routinely tested for heavy metal contamination; Canadian Environmental Quality Guidelines Summary Table Soil Quality Guidelines for the Protection of Environmental and Human Health was used as a guideline. These guidelines report contamination values for agricultural land on a dry weight basis using mg/kg dry weight; the solution values are reported using a wet weight basis of mg/L. Bioreactor feed slurries were dried and analyzed by Contract Labs to investigate the contamination risk potential. These manure slurries typically have greater than 85% moisture content. The manure feed slurries were dried to compare the dry weight data to comparable dry weight regulations. **It is not practical (or conceivable) to dry these solutions prior to field application**. This analytical exercise was to understand the slurry's chemistry. In this instance, when drying the manure slurry, molybdenum, selenium, tin and zinc were above the limits for agricultural soils using the Canadian Environmental Quality Guidelines Summary. When reviewing this dry weight data against Alberta Tier 1 Soil and Groundwater Remediation Guidelines 2014, Soil Remediation Guidelines; for Fine Agricultural Soil, copper concentrations for the dried slurry were also elevated.

In summary, contamination risks would exist if utilizing dried product slurries. Repetitive manure slurry applications would need to be assessed, as is true with repetitive manure applications.

Using a screw press to harvest the solutions was a significant achievement. Decant and flocculation strategies for harvest worked well for broths having a low percent dry matter, % DM. For initial runs having low % DM, the biologically active nutrient solution harvest volumes were easily 50% of the total broth volume. As the % DM increased, the inefficiencies of decant strategies to harvest became much more apparent; the harvest rate declined to only 20% to 30% of total volume for high % DM broths when using the decant method. When using a screw press greater than 90% of the broth can be harvested provided that lignocellulosic bedding materials are present. The initial poultry bedding was wood chips; this material was reduced in size due to bird activity and once harvested, the residual solids resembled wet sawdust.

To harvest with a screw press, the manure should contain bedding (bovine manure containing straw could be mixed into manures not containing bedding). Lignocellulosic materials, typically associated with bedding, fill the screw press's flight screws; which serves to trap the finer materials.

Later in the project, it was determined that acidification of the broth to pH 5 to 5.5 prior to harvest was required. This step is believed to enhance nitrogen stability by discouraging nitrogen cycle bacteria to convert ammonium to nitrate and to avoid phosphate divalent cation precipitates. This lower pH also inhibits ammonia which could lead to off gassing loss. Acidifying the broth at the time of harvest raises the nutrient concentrations, especially for divalent cations including iron [see Attachment 4 Trials 13 and 14]. Due to error, Trial 11 had a quarantine tank pH greater than 7; Trial 11 Nutrient Graphs highlight the corresponding ammonium, phosphate (declined by half) and divalent cation nutrient losses (due to high pH) where magnesium ions nearly disappeared [see Attachment 4 Trial 11].

When the broth was harvested by settling and decanting an analytical comparison of the residual slurry (Mother Liquor) and the chemistry of the decant solutions showed the Mother Liquor to have considerably higher nutrient and data values except for chemical oxygen demand (COD). Although the decant and remaining residual slurry (i.e. Mother Liquor) were from the same batch, the decant was considerably lower in nutrients. [See Attachment 6 for Bar Graphs]. Prior to chemical analysis all samples are first filtered to remove solids including microorganisms; consequently the microorganism contribution to the nutrient profile is lost.

The project tested the use of flocculation agents to shorten the decant time and enhance solution clarity. A wastewater company, ClearTech, was contracted to investigate the best flocculation agent for the process. The selected agent, CP 1080 at 86 mg per L broth, enhanced the settling time and broth clarity, but this harvest strategy was abandoned due to associated nutrient losses from using flocculation agents. [See Attachment **7** for Bar Graphs].

Product stability (shelf life) was explored by comparing the chemical analysis of a sample stored at 5°C for 77 days to the same solution stored in a shed outside during the summer for the same period of time [see Attachment 8 for Bar Graphs]. The ammonium concentration did not change between the two lots. The outdoor solution was not aerated which may explain the loss of nitrate, NO_3, likely due to nitrogen gas loss from denitrification, thus confirming that NO_3 is not as stable as NH_4. Solution

aeration could encourage the conversion of ammonium to nitrate (a negative outcome); project greenhouse solutions are typically aerated to maintain viable bacteria population(s). The acidification of the nutrient solutions serves to discourage the loss of nitrogen via denitrification by the *Nitrosomonas* and *Nitrobacter* nitrogen cycle bacteria.

There was some divalent cation variability, especially the extractable metals (which is best thought of as acid soluble rather than acid extractable where weakly bound metals are solubilized by the acid preservation agent). Dissolved phosphate values were constant suggesting mechanisms other than phosphate-divalent cation precipitates were responsible for dissolved magnesium and calcium ion losses.

The above Stability Data and Bar Graphs are from a single data set. Since this data is from a single data set, firm conclusions are not realistic. However, this work may serve as a model for future stability work or studies. Involving analytical chemistry professionals for such stability work would be of immense benefit. Microbiologists would also contribute immensely, especially if microbiology profiles were determined.

The biologically active nutrient solutions were associated with robust plant biomass and harvest. In addition, it was noted that "cabbage moths" didn't really impact the outdoor trial's brassica plants (broccoli, kale and red cabbage), whereas 300 meters away, garden plots of similar vegetables were completely destroyed by these pests.

Hypotheses:

I. The robust growth observed, could be due to water recycling in particular the "Brassica-favoured" anion SO_4^- would be constantly available and is not lost as in soil applications.

Note: the SO_4 ion concentration is augmented from using sulphuric acid as a pH control agent.

II. A solution component(s) may be discouraging insects or the plants are healthier to resist the attack – hatched larvae are present but in much lower numbers.

III. Increased plant turgor pressure, due to constant water exposure prevents insect attack.

A high sulphate, approximately 5,000 ppm SO_4 stock solution (that contained approxiamately1,200 ppm NO_3 and NH_4) was trialed to see if dilutions including no dilution of this solution would discourage club root infection of canola plants. The greatest dilution had approximately 670 ppm SO_4 and approximately 370 ppm NO_3 and NH_4. This experiment was unsuccessful; a subsequent literature review suggested the nitrogen levels were too high. The stock solution only had 10^7 organisms per mL. There may be merit in trialing a low nitrogen solution with considerably more organisms such as from methanol supplementation.

6. Literature cited

Provide complete reference information for all literature cited throughout the report.

Alberta Tier 1 Soil and Groundwater Remediation Guidelines 2014, Soil Remediation Guidelines

Canadian Environmental Quality Guidelines Summary Table Soil Quality Guidelines for the Protection of Environmental and Human Health

Guidelines for Compost Quality PN 1340, Canadian Council of Ministers of the Environment, 2005. ISBN 1-896997-60-0

National Standard of Canada, Canadian General Standards Board – CAN/CGSB-32.311-2015 Organic production systems — Permitted substances lists

7. Benefits to the industry (max 1 page; respond to sections *a)* and *b)* separately)

a) Describe the impact of the project results on Alberta's agriculture and food industry (results achieved and potential short-term, medium-term and long-term outcomes).

Fermenting organic wastes to produce biologically active plant nutrient solutions benefits Alberta's agri-food industries by providing an additional option for the province's organic waste. These biologically active nutrient solutions would be ideal for field fertigation, which is the technique of adding liquid fertilizer solutions while irrigating. Existing liquid manure injection equipment could be used to inject these readily plant available nutrient solutions for field applications.

The information generated by the project confirms waste is a resource. The following challenges need to be addressed before industry will implement this concept into a business model: identifying applicable greenhouse and field regulations, and scale up considerations. Industry can make this into a profitable business, especially if organic certification can be achieved. Organic certification should be feasible since the technology uses the same reagents that are permitted in the production of organic fish-based fertilizers.

Water recycling and the use of biologically active nutrient solutions have benefited the Alberta Greenhouse Industry by demonstrating this proof of concept for over two years, and obtaining organic status would likely foster industry adoption of the technologies.

b) Quantify the potential economic impact of the project results (e.g., cost-benefit analysis, potential size of market, improvement in efficiency, etc.).

An initial economic analysis for the 1,000 liter scale [not including facility, overhead and utilities costs] indicates it may be economically feasible to pursue this technology. The Return on Investment (ROI) can be between 250% to 600% depending on the wholesale value of the biologically-active nutrient solution (three scenarios based on wholesale pricing are provided). The ROI values do not account for basic business overhead costs; although the assumptions are conservatively realistic the outcome may be overly optimistic. Attachment 1 highlights the economic assumptions, Net Present Value and Benefit-Cost Ratio analysis, data and assumptions

The 1,000 liter scale is likely too small for an industrial setting. Scaling up the technology involves integrating existing equipment to develop robust support equipment in particular feeding, dosing and harvest functions.

8. Contribution to training of highly qualified personnel (max ½ page)

Specify the number of highly qualified personnel (e.g., students, post-doctoral fellows, technicians, research associates, etc.) who were involved in the project.

The project greatly complemented the project engineer's fermentation and greenhouse applications background. The project provided exposure to current and future greenhouse applications and challenges. Developing the methods and infrastructure to utilize biologically-active nutrient solutions and water recycling was a much appreciated skill set enhancement.

9. Knowledge transfer/technology transfer/commercialisation (max 1 page)

Describe how the project results were communicated to the scientific community, to industry stakeholders, and to the general public.

The results from this project were presented to the scientific community, industry stakeholders and the general public via:

a) Scientific Presentations
   - Cultivating Connections Alberta Regional Food Systems Forum February 2017 – presented

b) Industry-oriented presentations
   - Green Industry Show & Conference, Edmonton November 2016 – presentation booth
   - The Festival of Big Ideas, Edmonton June 2017 – presentation booth
   - Northlands Farm Fair, Edmonton November 2017 –booth complete with hydroponic demonstration (live plants including active water recycling
   - Green Industry Show & Conference, Calgary November 2017 - presentation booth

c) Media activities
   - 45 minute Webinar February 2017 https://gov-ab.webex.com/govb/lsr.php?RCID=efad04702be168748dcef68e2a55633c.

d) Commercialisation activities or patents
   - The CDC North site hosted many tours most notably:
   - Chinese Delegation July 2016 – 6 individuals
   - U of A Permaculture Group – 40 individuals

The opportunity to showcase the technology's products was an excellent means to advertised Alberta Agriculture and Forestry's efforts to utilize waste as a resource.

Section D: Project resources

1. **Statement of revenues and expenditures:** *Please see attached spreadsheet*
   a) **In a separate document certified by the organisation's accountant or other senior executive officer, provide a detailed listing of all cash revenues to the project and expenditures of project cash funds.** Revenues should be identified by funder, if applicable. Expenditures should be classified into the following categories: personnel; travel; capital assets; supplies; communication, dissemination and linkage; and overhead (if applicable).

Please see attached 11 x 17 spreadsheet, Project C028R Final Report Financial Statement.

Provide a justification of project expenditures and discuss any major variance (*i.e.*, ± 10%) from the budget approved by the funder(s).

		Budget *over Project 2.25 year Duration*			
Item	**Description**	**Original**	**Actual**	**% Variance**	**Justification of Expenditures and Variances**
1	People	$326,250.00	$256,450.16	27	Reflects Project Loss of: -2.25 years fulltime scientist position - 1.25 years fulltime project technician
2	Travel	$4,500.00	$12,177.91	-63	Increase travel to S. Alberta - CARA in Oyen, AB - tree trials Strathmore, AB - weekend, afterhours greenhouse / bioreactor monitoring - mileage
3	Capital Assets	$83,076.00	$35,513.92	134	Project saved funds by borrowing equipment and building in-house greenhouse infrastructure
4	Supply	$61,425.00	$143,308.52	-57	Analytical costs were substantial in addition to material and supplies for building greenhouse infrastructure
5	CDL	$4,000.00	$2,747.30	46	Project was unable due to labour restrictions to organize a workshop

Original budget included $19,258 for Overhead which was distributed among the other categories.

2. Resources:
Provide a list of all external cash and in-kind resources which were contributed to the project.

Total resources contributed to the project		
Source	**Amount**	**Percentage of total project cost**
Funders ACIDF maximum value $200,175.00	$199,518.51	44.3 %
Other government sources: Cash	$59,492.48	13.2 %
Other government sources: In-kind	$181,186.82	40.3 %
Industry: Cash	$10,000.00	2.2 %
Industry: In-kind		
Total Project Cost	**$450,197.81**	**100%**

Please see attached 11 x 17 spreadsheet, Project C028R Final Report Financial Statement.

External resources (additional rows may be added if necessary)		
Government sources		
Name (only approved abbreviations please)	Amount cash	Amount in-kind
Alberta Agriculture and Forestry	$59,492.48	$181,186.82
Industry sources		
Name (only approved abbreviations please)	Amount cash	Amount in-kind
Sustainable Poultry Farming Group, BC	$10,000.00	

Section E: The next steps (max 2 pages)

Describe what further work if any needs to be done.

a) Is new research required to deal with issues and opportunities that the project raised or discovered but were not dealt with within the current project?

To advance the conclusions of this project, other academics and researchers should be involved to study and validate the process/ products. We would expand the collaboration to include other Albertan Producer Research groups. The Chinook Applied Research Association, CARA, in Oyen, AB will be starting up a lab specializing in soil health assessments. This is a great opportunity to work with producers in the emerging soil health field.

Another area of opportunity is developing co-fermentation strategies where trialing other organic feedstocks, in particular other manures, with and without a waste soluble carbon feedstock would be of interest to the industry.

b) Is there related work that needs to be undertaken to continue advancement of the project technology or practice?

To continue advancing this project, agronomy trials should be conducted to quantify the solutions impact on plant growth and harvest.

Hypothesis: The solutions' microbiological activity would suppress soil pathogens.

i. Hypothesis could be trialed (as an organic process) in both greenhouse and field settings.

ii. Although water recycling is currently not an organic practice – this is an opportunity to lead by demonstrating the merits of water recycling for soil based growing systems.

It appears Brassica plants respond very well from continuous exposure to sulphate; which suggests growing Chinese vegetables as a demonstration crop could be useful for organic practices and water recycling may be feasible.

c) Did the project identify any new technology or practice that needs to be developed?

d) What suggestions do you have that increase commercial use of results by farmers and/or companies. These may be:

1. Commercial Uptake.

Commercial uptake is more likely to occur once organic certification for the process is obtained, due to the high interest from the organic industry/farmers.

2. Further Research Toward Commercial Use

Scaling-up the technology to 5,000 liter scale will demonstrate the process at an industrial scale. It is suggested that utilizing an industrial computer control system to control pH, dissolved oxygen and antifoam addition would be particularly useful. A robust manure feeding mechanism is required, as

well as an auger system that may also entrain the manure into a slurry. A bioreactor tank design is recommended to allow for the removal of grit and ease of harvest.

The products are neither manure nor compost (nor compost tea), therefore manure and compost regulations may not apply. The solutions can be considered "safer" than untreated manure. An aliquot of solution was tested [a DNA scan] for thirty plant root pathogens, which was free of the tested pathogens. A poultry amended soil sample tested positive for five pathogens (it is unknown if these pathogens were present in the soil and / or manure [see Attachment 2 for details]. A thorough assessment by qualified professionals is required to evaluate associated contamination risks.

Product stability studies would be required for marketing and application purposes.

This technology could be exploited as a heat source (in addition to a CO_2 source) for greenhouses. This involves operating the bioreactor in a semi-continuous manner. Carbon dioxide emission from the bioreactor into the greenhouse is favourable during photosynthesis. Theoretically, it should be possible to operate the bioreactor in a near-continuous thermophilic phase which would provide a 60°C to 70°C heat source. Since the solutions are concentrated, it is possible to harvest 10 to 20 L after the pathogen elimination step and then feed the bioreactor more manure (and soluble carbon) to possibly maintain the heat phase.

The work sought out in the Microbial Biomass Identification and its Contribution to the Nutrient Profile, would likely be a pre-requisite for CFIA acceptance of the products.

It is recommended to test the solutions bio-stimulant effects with regards to plant and soil health.

A consideration which was out of this project's scope included the regulatory compliance to address virus risks, in particular the "Bird Flu" risk. Moving forward, the product(s) will be required to conform to CFIA fertilizer or amendment regulations.

3. Extension and information disbursement

Section F: Research Team Signatures and Employers' Approval

The team leader and an authorised representative from his/her organisation of employment MUST sign this form.

Research team members and an authorised representative from their organisation(s) of employment MUST also sign this form.

By signing as representatives of the research team leader's employing organisation and/or the research team member's(s') employing organisation(s), the undersigned hereby acknowledge submission of the information contained in this final report to the funder(s).

Team Leader's Organisation

Team Leader	
Name: Marc Legault	**Title/Organisation:** Bio-Industrial Engineer Alberta Agriculture and Forestry
Signature:	**Date:** 01 February, 2018
Team Leader's Employer's Approval	
Name: Honq Qi	**Title/Organisation:** Director, Bio-Industrial Opportunities Section Alberta Agriculture and Forestry
Signature:	**Date:** Feb 2. 2018

Research Team Members (add more lines as needed)

1. Team Member	
Name: Emmanuel Laate	**Title/Organisation:** Senior Crop Economist Alberta Agriculture and Forestry
Signature:	**Date:** Feb 1, 2018
Team Member's Employer's Approval	
Name: Philippa Rodrigues	**Title/Organisation:** Director, Economics Section Alberta Agriculture and Forestry
Signature:	**Date:** Feb 1 /18

Appendix A. Permitted Organic Substances
Excerpts from: National Standard of Canada, Canadian General Standards Board – CAN/CGSB-32.311-2015; Organic production systems — Permitted substances lists

Highlights denote substances used in poultry manure fermentation project

Table 4.2 – Soil amendments and crop nutrition

Fish meal, fish powder, fish wastes, hydrolysate, emulsions and solubles	The following fish products are permitted: fish meal; fish powder; and hydrolysate, emulsions and solubles. Fish farm wastes shall be composted. Ethoxyquin or other synthetic perservatives, fertilizers and other chemically synthesized substances not listed in this standard shall not be added to fish products. Chemical treatment is prohibited, except that liquid fish products may be pH adjusted with the following, in preferential order: 1. a) vinegar; 2. b) non-synthetic citric acid; 3. c) synthetic citric acid; 4. d) phosphoric acid; or 5. e) sulphuric acid. The amount of acid used for pH adjustment shall not exceed the minimum needed to stabilize the product.
Iron	The following sources of iron are permitted, to correct documented iron deficiencies: ferric oxide, ferric sulphate, ferrous sulphate, iron citrate, iron sulphate or iron tartrate. See Table 4.2 Micronutrients.
Surfactants	Non-synthetic substances. See Table 4.2 Formulants, Table 4.2 Wetting agents, and Table 4.3 Soaps; table 4.3 Vegetable oils.

Table 6.5 - Processing aids

Oxygen	No annotation.
Potassium hydroxide (caustic potash)	For pH adjustment. Prohibited for use in lye peeling of fruits and vegetables.

Appendix B. Table 1 pH Control Agents

pH Agent	Pro's	Con's
Phosphoric Acid	relatively safe increases phosphate concentrations foam occurs at the beginning(?) a permitted organic reagent	increases the likelihood of nutrient loss especially divalent cations: iron, calcium, magnesium, manganese foaming tends to be greater most expensive
Nitric Acid	appears to increase the baseline temperature from 50 to 60°C increases nitrate concentration less nutrient loss (due to foam cheap higher concentrations of divalent cation nutrients i.e. no phosphate-precipitate losses the associated foam is more manageable (readily collapses)	rather dangerous most nutrients become over diluted since greater solution dilutions are required due to high nitrate concentrations **not a permitted organic reagent**
Sulphuric Acid	Cheap, Increases sulphate concentration Appears to have better feather degradation Foam occurs towards the end (?) a permitted organic reagent	rather dangerous may appear to produce (almost) off-odours
Potassium hydroxide	Increases the potassium concentration a permitted organic reagent	
Ammonium hydroxide	Increases the ammonium concentration likely leading to an increase in biomass	**May not be a permitted organic reagent(?)**

Appendix B. Table 2 Reagents

Reagent Proposed Effect	Pro's	Con's
Ammonium Hydroxide	Increase available ammonium to increase biomass, metabolic activity	**May not be a permitted organic reagent(?)**
Methanol Increase soluble carbon content	1000 fold increases microbial density, cells per mL Can be used to increase the citric acid concentrations in stock solutions	Expensive **May not be a permitted organic reagent(?)**
Antifoam Agents [10, 20 and 30% emulsions of silicon were trialed]	Arrests foam / nutrient loss Can be a permitted organic reagent	Decreases oxygen transfer leading to temperature loss
Vinegar, CH_3COOH Antifoam agent	Greatly delays the need for antifoam especially if added at the start of the fermentation, allows broth to attain 70°C temperatures a permitted organic reagent	Appears to create a buildup of compound(s) that negatively impact the fermentation (stalled reactions) Especially pronounced in the carryover of Mother Liquors Additions late in the fermentation run may trigger exceptionally large "foam outs" leading to major nutrient losses
Iron sulphate Increase the iron concentration ideally sequestered in microbial biomass	Iron concentrations increased upto10 fold Unknown if sequestered in microbial biomass Appears to be plant available since chlorosis has not been observed a permitted organic reagent	Appears to create a dense and thick foam - suspect the '+' charge is bridging '-'charged cells and / or cell debris. These foam layers (nearly crusts) can easily be 6" thick.
Citric acid, $C_6H_8O_7$ A possible organic pH control agent [may be too weak]	**not assessed** – greater concentration if mixed with alcohol – may serve to enhance soluble carbon a permitted organic reagent	

Appendix C. Greenhouse Methods and Observations

Greenhouse biologically active nutrient solutions were trialed at Crop Diversification Centre (CDC) North. The trials involved raft and drip irrigation of soil-less substrate culture; the soil-less mix consisted of equal parts coconut coir, a mycorrhizal inoculated peat moss (containing perlite) and perlite. Greenhouse trials and an outdoor tree farmer trial were started one year prior to the project; both continued for two additional years.

The project had to develop or refine existing greenhouse culture techniques to utilize the biologically active nutrient solutions. These solutions were trialed for three years in both a soil-less greenhouse setting which recycled all water and outdoor tree trials. Outdoor trials, including soil-less substrates (perlite, sand, biochar, humalite and the above soil-less mix) were tested while continuously recycling all water. Masonry sand as a substrate had the poorest drainage of all tested substrates. Humalite possibly due to microbial activity appeared to raise the feed solutions' sodium and iron concentrations.

Plastic troughs served as the indoor and outdoor plant beds; a 3 cm thick rigid porous plastic sheet was placed along the entire bed bottom which was then covered with landscape fabric which also covered the bed walls. The soil-less substrate placed inside these plant beds was contained by the landscape fabric.

Sediment containment traps were required to eliminate sediments from the recycled water and thereby prevent sediment introduction into the feed tanks. Clean-out traps are recommended especially if plants are to be cultured beyond two years; this requires an easily accessible piping penetration to allow the removal of piping plugging due to root growth.

Biologically active nutrient solutions differ from other nutrient solutions as they are rich in microorganisms. The billion organisms per mL in these solutions are "nutrient storehouses" in addition to the solution's chemical nutrients.

Hypothesis: *The microbial biomass act as a slow-release source of nutrients that complement existing nutrients in the manure-derived, biologically active solutions for plant uptake.*

In regards to manure not containing bedding materials, it was difficult to clarify the fine lignocellulosic materials. During greenhouse trials, these lignocellulosic materials quickly plugged up inline feed filters. Fortunately, these materials do not plug up irrigation drip emitters, and as such, inline filter use was abandoned.

The concentrated solutions (up to 50 to 60 fold dilution required) contain ammonium ions, NH_4, as the dominant nitrogen form. For greenhouse applications where plants tend to prefer nitrate, NO_3 ions; it is best to inoculate the feed tank with nitrogen cycle bacteria to convert NH_4 into NO_3. Such inoculums are readily available in aquarium stores. Inoculation is unnecessary for field applications due to the presence of soil nitrogen cycle bacteria.

The pH of the plant feed tanks (diluted biologically active nutrient solutions) were maintained between 5 and 6.5 as the best (speculated) trade-off to promote root/plant health and microbial

processes. The electrical conductivity (EC),) was targeted to be between 2 and 4 mS/cm; the dissolved oxygen was targeted to be above 5 ppm to encourage aerobic microorganisms and discourage root zone attack by pythium especially true for raft culture. Daily monitoring (pH, temperature and dO) was recorded for both nutrient feed tanks. A 1,000 liter feed tank was primarily used for the culture of fig trees, the other tank (approxiamately3,000 liter) was for tomatoes, tomatillos, hot peppers, purple string beans and passion fruit. Both tanks were oxygenated to maintain the aerobic organisms and to supply oxygen to the root zone.

Hydroponic operations that utilize synthetic fertilizers typically need to acidify their feed tanks; in contrast, the project's hydroponic use of biologically active nutrient solutions needed to constantly raise the pH of its feed tanks using potassium bicarbonate. Microbial activity is believed to be responsible for this acidification. However, the fig feed tank (that utilized the same biologically active nutrient stock solution) for a period of time was opposite in that phosphoric acid was required to maintain the pH setpoint.

The process yields a liquid and a solids product. The bio-nutrient solutions were associated with robust plant biomass and harvest. The solids were used to grow oyster mushrooms and a grower successfully grew portabella mushrooms. Soil remediation professionals are interested in this product since it augments soil carbon without nutrient overloading as may be the case with manure. The final outdoor trial mixed bovine bone chips into the soil-less substrate to encourage robust microorganism growth since the porosity provides protected shelter.

The growth of strawberry cultures was unsuccessful, although partial success was achieve with the growth of Echinacea. To optimize the growth of strawberry cultures, an experienced greenhouse grower would be required.

Appendix D. Engineering Challenges and Considerations

The bioreactor / fermenter was an insulated 1,000 liter plastic bottom hoppered vessel. A robust agitation assembly was mounted above the vessel. Argus, an industrial greenhouse automation system, was used to control pH; pH control involved two pH probes, a guard probe and a control probe. The Argus system also provided data acquisition for pH, temperature and dissolved oxygen. The guard probe was used to check and confirm the measurements of the control probe (from which pH control was based on). Acid or base was slowly added into the fermentation broth complete with a lag time between doses to avoid inducing pH fluctuations.

The presence of grit in the manure (limestone grit for gizzard health and oyster shell fragments for egg shell development) was a challenge. Aggressive agitation of the broth enhances oxygen transfer which also entrains this grit causing abrasive damage to the instruments. Placing the pH probes into plastic sleeves with large perforations prevented damage to a certain degree. Pipe wall segments between the perforations served to reduce broth velocity, thereby causes the grit to settle out before impact with the probes. A double perforated wall may improve this considerably, but the perforations must be large enough to prevent plugging due to the presence of feathers and straw.

The addition of antifoam for the first runs was typically in increments of 5 and 10 mL; these smaller aliquots caused slight but noticeable decreases in broth temperature. As the project progressed, the aliquots of antifoam became larger in order to investigate the process's robustness. These larger additions of antifoam were often too much (especially 200 mL of canola oil) and may have irreversibly impacted the reaction times. A computer controlled dosing (or spraying) of smaller volumes of antifoam into the bioreactor would be of considerable benefit in attaining and maintaining the thermophilic pathogen elimination step.

It would be beneficial to incorporate or develop a shaker mechanism to add powdered lime as a pH base control agent. This would augment the solution's calcium content (and magnesium depending on the grade of lime) and may decrease costs (overtime) since lime is inexpensive compared to the other base agents.

A rough rule of thumb for oxygen delivery "system sizing" is 0.1% lpm relatively pure oxygen per batch volume for approxiamately10% DM. The project is in possession of oxygen and antifoam control instrumentation; however additional funds need to be secured for their installation.

Incorporating a steam kill step may be required to denature bird flu viruses especially if the solutions were to grow poultry feed. Although bird influenza viruses are considered denatured at 75°C, it is a microbiology practice to expose such fluids to 85°C for 5 minutes to denature viruses.

Regulatory and Animal Health Authorities (subject matter experts) are needed to assess the risk(s) and determine mitigation strategies.

Attachments

1) Economic Analysis, Data and Assumptions

2) DNA Pathogen

3) SOP,s and Control Forms

4) Fermentation and Nutrient Graphs
- Trials 1 to 14 inclusive

5) Nutrient Analysis for Specific pH Agents

6) Residual vs Decant Nutrient Bar Graphs

7) Loss Due to Flocculation Agent

8) Nutrient Stability Bar Graphs

Attachment 1 Page 1 of 8

Aerobic Digestion of Poultry Manure Economic Analysis

Aerobically digested poultry manure can be used indoors and outdoors to grow crops. A net present value model is used to determine whether it makes sense financially to invest in a 1,000 litre bioreactor to produce concentrated bio-nutrient solution. Table 1 shows the investment costs, operating costs and revenues based on three wholesale prices for the bio-nutrient solution.

Table 1 Bioreactor Costs and Returns

Volume of Bioreactor	1,000 Litres		
Number of batches of nutrient solution produced per year	30	30	30
A). Revenue			
Quantity of concentrated bio-nutrient solution (L/batch)	675	675	675
Wholesale price of concentrated bio-nutrient solution - ($/L)	$7.5	$10.0	$15.0
Annual Gross Revenue	**$151,875**	**$202,500**	**$303,750**
B). Operating costs per Bioreactor Batch			
Pathogen testing			
E. coli	$45.0	$45.0	$45.0
Salmonella	$32.0	$32.0	$32.0
fecal coliforms	$32.9	$32.9	$32.9
Aerobic plate count	$22.8	$22.8	$22.8
Total pathogen testing per batch	**$132.65**	**$132.65**	**$132.65**
Reagent Costs			
Phosphoric acid 75% Food Grade	$5.7	$5.7	$5.7
Sulphuric acid 66 BE	$2.1	$2.1	$2.1
Nitric acid	$1.9	$1.9	$1.9
Caustic Potash 45 % Solution	$4.7	$4.7	$4.7
Ammonium hydroxide	$3.0	$3.0	$3.0
Ferrous sulphate	$1.8	$1.8	$1.8
Antifoam Silchem	$22.0	$22.0	$22.0
Total Reagent Costs per batch	**$41.08**	**$41.08**	**$41.08**
Total Pathogen and Reagent Costs per Year	**$5,211.9**	**$5,211.9**	**$5,211.9**
Annual Consumable Instrumentation Costs			
Two (2) set of PH probes	$4,400.0	$4,400.0	$4,400.0
Dissolve oxygen	$1,000.0	$1,000.0	$1,000.0
Electrical conductivity	$1,000.0	$1,000.0	$1,000.0
Hand held meter	$3,500.0	$3,500.0	$3,500.0
Annual Consumable Instrumentation Costs	**$9,900.0**	**$9,900.0**	**$9,900.0**
Labour	**$1,825.0**	**$1,825.0**	**$1,825.0**
Total Operating Costs (not including energy and other overhead costs)	**$16,936.9**	**$16,936.9**	**$16,936.9**
D). Gross margin (not including energy and other overhead costs)	**$134,938.1**	**$185,563.1**	**$286,813.1**
E). Capital costs			
Bioreactor			
Plastic tank w\ hopper bottom	$1,000	$1,000	$1,000
Insulation	$1,000	$1,000	$1,000
Agitator	$7,000	$7,000	$7,000
Control system	$25,000	$25,000	$25,000
Oxygen system	$70,000	$70,000	$70,000
pH control skid	$15,000	$15,000	$15,000
Foam and Oxygen control (optional)	$30,000	$30,000	$30,000
Harvest Equipment			
Screw press	$45,000	$45,000	$45,000
Bag filters	$15,000	$15,000	$15,000
Quarantine Tank 1000 L	$350	$350	$350
Nutrient Storage Tank 5000 L	$1,500	$1,500	$1,500
Transfer and other equipment			
Peristaltic pump w\ VFD	$12,000	$12,000	$12,000
Sump / recirculation pump(s)	$2,000	$2,000	$2,000
Total Capital Costs	**$224,850**	**$224,850**	**$224,850**

Prepared By: Emmanuel Anum Laate, Senior Crop Economist, Economics Section.

The total capital cost per bioreactor of $224,850 was assumed to be financed. A 10-year Alberta farm loan with an interest rate of 4.45% was used for the analysis. A summary of the economic indicators from the model are presented in Table 2.

Table 2 Summary of Economic Indicators

Volume of Bioreactor	1,000 Litres		
Wholesale price of concentrated bio-nutrient solution - ($/L)	$7.50	$10.00	$15.00
Net Present Value (NPV)	$1,025,657	$1,505,131	$2,464,080
Benefit cost ratio (BCR)	3.48	4.65	6.97
Payback if financed	2	2	2
Return on Investment (ROI)	248%	365%	597%

The results show that using a bioreactor to aerobically digest poultry manure and produce concentrated bio-nutrient solution is profitable. The NPV ranged from $1 million to approximately $2.5 million and the BCR ranged from 3.48 to 6.97.

Definition of Economic Indicators

Net present value (NPV) is the difference between the present value of cash inflows and the present value of cash outflows. If the NPV >1.0, (positive) then the investment is feasible or it makes sense financially.

Benefit-cost ratio (BCR) is the ratio of discounted benefits to discounted costs expressed in monetary terms. It is an accepted procedure for making go/no-go decisions on projects as compared to alternatives. If the value of BCR > 1, the project or investment is feasible (makes sense financially).

Payback is the length of time it will take for the investment to return its original cost or the number of years required for cash inflows to just equal cash outflows. It indicates the project liquidity rather than profitability. The computation does not address the total profitability of the project, rather it simply calculates how fast a project recovers its cash investment.

Return on Investment (ROI) measures the amount of return on an investment relative to the investment's cost. Because ROI is measured as a percentage, it can be easily compared with returns from other investments, allowing one to measure a variety of types of investments against one another.

Prepared For:
Marc Legault, P. Eng.
Bio-Industrial Engineer
Program Lead, Waste to Nutrients
Alberta Agriculture and Forestry

Prepared By: Emmanuel Anum Laate, Senior Crop Economist, Economics Section.

Item	Capital Costs for Bioreactor 1,000L	Actual Cost	Optional Costs	Notes
1	Plastic tank w\ hopper bottom	$1,000		
2	Insulation	$1,000		
3	Agitator	$7,000		
4	Control system	$25,000	[$15,000]	the current control system is Argus, greenhouse based control system - recommend a dedicated process control system
5	Oxygen system	$70,000		
6	pH control skid	$15,000		2 of each: peristaltic pump, pH transmitters. plastic tank
7	Foam control		$30,000	foam control can be a manual operation but best if automated - oxygen control can be manual
8	Oxygen control			
	1,000L Bioreactor Capital Costs	**$119,000**	**$30,000**	
	Total 1,000L Bioreactor Capital Costs	**$149,000**		
	Total Harvest and Support Equipment Capital Costs	**$75,850**	from Worksheet Harvest and Support Equipment	
	Total Capital Costs	**$224,850**		

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Item	Operating Costs per Bioreactor Batch	Actual Cost	Notes
1	Acid	$85.00	based on 30 kg consumption 1 part H_3PO_4 to 4 H_2SO_4 vol/vol
2	Base	$83.00	based on 20 kg consumption 1 part H_3PO_4 to 4 H_2SO_4 vol/vol
3	Antifoam	$22.00	
4	Ferrous sulphate	$2.50	
5	Oxygen	$65.00	based on $1,500 yearly maintenance cost shared over 2 batches per month
6	Instrumentation replacement	$481.25	based on 2 batches per month sharing the yearly cost given below
7	Pathogen testing	$132.65	
	Total cost per batch	**$738.75**	

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Instrumentation (yearly cost) may be considered a consumable cost

Item	Instrument	Cost	Notes
1	pH probes - two	$2,200	includes temperature
2	Dissolve oxygen	$1,000	estimates
3	Electrical conductivity	$1,000	
4	Hand held meter	$3,500	pH, temp, dO, conductivity

Consumable Instrumentation Costs **$7,700**

Assume

1.5 **replacements per year total instrument consumable cost is** **$11,550**

Pathogen testing per batch

Item	Pathogen	Cost	Notes
1	*E. coli*	$45.00	
2	*Salmonella*	$32.00	yet to be detected
3	fecal coliforms	$32.90	Total coiliform add'l $26.75
4	Aerobic plate count	$22.75	

Total pathogen testing per batch **$132.65**

Attachment 1 page 5 of 8

Reagent Costs	$/kg	Notes
Phosphoric acid 75% Food Grade	$5.70	
Sulphuric acid 66 BE	$2.08	
Nitric acid	$1.85	no longer used or recommended
Caustic Potash 45 % Solution	$4.69	plus $42 surcharge
Ammonium hydroxide	$3.01	
Ferrous sulphate	$1.75	
Antifoam Silchem	$22.00	

Attachment 1 page 6 of 8

Capital Costs for Harvest and Support Equipment

Item	Harvest Equipment	Actual Cost	Optional Costs	Notes
1	Screw press	$45,000		
2	Bag filters	$15,000		requires feed pump, bags would be filled with lignocellulosic solids
3	Quarantine Tank 1000 L	$350		
4	Nutrient Storage Tank 5000 L	$1,500		
	Total Harvest Equipment	**$61,850**		

	Transfer and other equipment			
4	Peristaltic pump w\ VFD	$12,000		ideal to feed bag filters
5	Sump / recirculation pump(s)	$2,000		
	Transfer and other equipment	**$14,000**		

Total Support Equipment for 1,000L Bioreactor **$75,850**

Attachment 1 page 7 of 18

 Attachment 1 Page 8 of 8 **Aerobic Digestion Technology Costs**

								Analysis % (assume wt/wt) [Bold blue font denotes advertised Label Data]									
Tag	Primary Ingredients	Odour	Dilution Notes	Dilution Factor	Volume L	Cost	Cost per Liter	N	P_2O_5	PO_4	P	K_20	K	Sulfur S	Calcium	Sodium % wt/wt	Comments
A	Seaweed	Mild	15 to 30 mL per Liter	33.3 to 66.7	0.91	$16.99	$18.67	**0.1**			**0.1**	**0.5**	0.4				See Note 2
B	Vegetable, bat guano, etc.	yes moderate	10 mL per Liter	100	1	$11.99	$11.99	**3**	**1**		0.4	**2**	1.7		**0.5**		See Note 2
C	Fish emulsion, fulvic acid, kelp, etc...	yes **Foul**	10 to 20 mL per Liter	50 to 100	1 kg	$14.99	~$15.00	**3**	**2**		0.9	**5**	4.1	**2**			See Notes 1 and 2
D	Fish emulsion	yes **Foul**	1 to 4 tablespoons per gal. 1 tbsp = 14.787 mL; 1 US Gal.= 3.79 L	64 to 256	0.946	$13.99	$14.79	**5**	**1**		0.4	**1**	0.8				See Notes 1 and 2
AF	Poultry Manure digestate	**No**		50 to 60				**0.6 to 0.3**		**0.55 to 0.1**	0.2 to 0.03		**0.5 to 0.2**	**1.2 to 0.1**	**0.23 to 0.1**	0.06 to 0.03	biologically active 10^9 organisms/mL readily-available nutrients

Notes

1 Fish, unprocessed manure based fertilizers typically need to be microbially mineralized to yield plant available nutrients

2 Fish, unprocessed manure (possibly marine plants) based fertilizers typically contain sodium

Attachment 2 – DNA Pathogen Scan

To	Date
	Thursday October 29, 2015

LABORATORY TESTING OF MATERIALS

Background

Twice this past season, materials in use at the nursery were sampled to examine at a commercial laboratory for presence of root rot pathogens.

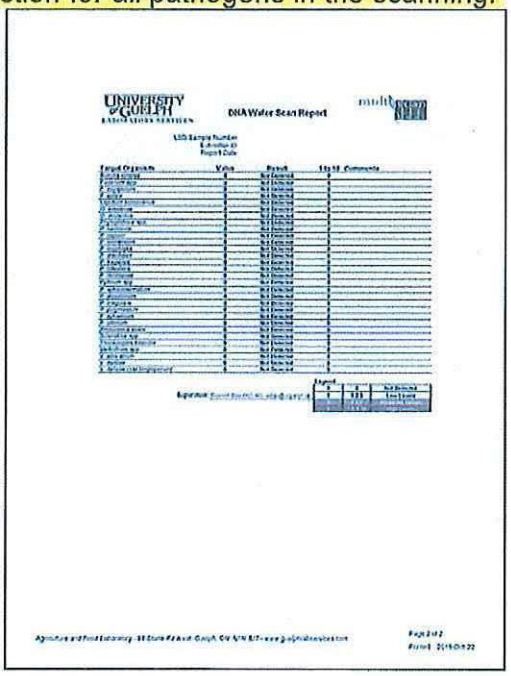
The samples were examined using DNA scanning, placing the retrieved DNA against a bank of known "markers" for multiple plant pathogens. It is currently the most advanced technique for this work and only available at Guelph University in Ontario.

The laboratory report returns a rating of 0 (none detected) up to 10 (high level) for 30 different plant pathogens. More information at http://www.guelphlabservices.com/AFL/Service_Growers.aspx (go to "Fungi").

Results for "Chicken Digest"

Sample collected by early October.

Results: No detection for all pathogens in the scanning.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DNA Water Scan Report

Results for "Manure"

Sample collected in late May . This was either leftover materials after potting trees into Air Pots, or the manure part of the mix (the information given us was unclear).

Results:

Moderate levels of *Fusarium* and *Pythium*, High levels of *Pythium ultimum*.

Comments:

The material used for potting of trees arrived with *Fusarium* and *Pythium*. This indicates material of poor quality, or poor composting, or infection from raw ingredients.

Pythium is a soil pathogen that infects plants under stress and causes root rot and stem rot. *Pythium* is found commonly in nature. A potting mix with this pathogen is a major concern for greenhouse production and during propagation. Larger plants can sustain a small amount of root rot infection.

It would be prudent to use "clean" materials for the potting mix in the future.

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Basic Soil Scan Report

multiSCAN

LSD Sample Number
Submitter ID
Report Date

Target Organism	Value	Result	1 to 10	Comments
Botrytis cinerea	0	Not Detected	0	
Fusarium spp	2	Moderate Levels	[illegible]	
F. oxysporum	0	Not Detected	0	
F. solani	0	Not Detected	0	
Olpidium bornovanus	0	Not Detected	0	
O. brassicae	0	Not Detected	0	
O. virulentus	0	Not Detected	0	
Phytophthora spp	0	Not Detected	0	
P. cactorum	0	Not Detected	0	
P. capsici	0	Not Detected	0	
P. cinnamomi	0	Not Detected	0	
P. cryptogea	0	Not Detected	0	
P. drechsleri	0	Not Detected	0	
P. fragariae	0	Not Detected	0	
P. infestans	0	Not Detected	0	
P. nicotianae	0	Not Detected	0	
Pythium spp	2	Moderate Levels	[illegible]	
P. aphanidermatum	0	Not Detected	0	
P. dissotocum	0	Not Detected	0	
P. irregulare	0	Not Detected	0	
P. polymastum	0	Not Detected	0	
P. sylvaticum	2	Moderate Levels	5	
P. ultimum	3	High Levels	10	
Rhizoctonia solani	0	Not Detected	0	
Sclerotinia spp	0	Not Detected	0	
Thielaviopsis basicola	0	Not Detected	0	
Verticillium spp	0	Not Detected	0	
V. albo-atrum	0	Not Detected	0	
V. dahliae	0	Not Detected	0	
V. dahliae (resting spores)	0	Not Detected	0	

Supervisor: ________________

Legend

0	0	Not Detected
1	1,2,3	Low Levels
2	4,5,6	Moderate Levels
3	7,8,9,10	High Levels

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Page 2 of 2
Printed 2015-Jun-02

UNIVERSITY of GUELPH
LABORATORY SERVICES
Agriculture and Food Laboratory

FINAL Report
Submission#
Reported: 2015-Jun-02

Submitted By:

DNA Multiscan

Date Authorized: 2015-Jun-02 15:29

Sample ID	
Client Sample ID	
Specimen type	**Soil**
Sampling date/time	
DNA Multiscan	Results Attached

Test method(s): PDC-114

Supervisor: Shannon Shan PhD, Agriculture and Food Laboratory 519 823 1268 ext. 57227 xshan@uoguelph.ca

FINAL Report

Submission#
Reported:

UNIVERSITY of GUELPH
LABORATORY SERVICES

Basic Soil Scan Report

multiscan DNA

LSD Sample Number:
Submitter ID:
Report Date: June 02 2015

Target Organism	Value	Result	1 to 10	Comments
Botrytis cinerea	0	Not Detected	0	
Fusarium spp.	2	Moderate Levels	6	
F. oxysporum	0	Not Detected	0	
F. solani	0	Not Detected	0	
Olpidium bornovanus	0	Not Detected	0	
O. brassicae	0	Not Detected	0	
O. virulentus	0	Not Detected	0	
Phytophthora spp.	0	Not Detected	0	
P. cactorum	0	Not Detected	0	
P. capsici	0	Not Detected	0	
P. cinnamomi	0	Not Detected	0	
P. cryptogea	0	Not Detected	0	
P. drechsleri	0	Not Detected	0	
P. fragariae	0	Not Detected	0	
P. infestans	0	Not Detected	0	
P. nicotianae	0	Not Detected	0	
Pythium spp.	2	Moderate Levels	6	
P. aphanidermatum	0	Not Detected	0	
P. dissotocum	0	Not Detected	0	
P. irregulare	0	Not Detected	0	
P. polymastum	0	Not Detected	0	
P. sylvaticum	2	Moderate Levels	5	
P. ultimum	3	High Levels	10	
Rhizoctonia solani	0	Not Detected	0	
Sclerotinia spp.	0	Not Detected	0	
Thielaviopsis basicola	0	Not Detected	0	
Verticillium spp.	0	Not Detected	0	
V. albo-atrum	0	Not Detected	0	
V. dahliae	0	Not Detected	0	
V. dahliae (var longisporum)	0	Not Detected	0	

Supervisor: _____

Legend

0	0	Not Detected
1	1,2,3	Low Levels
2	4,5,6	Moderate Levels
3	7,8,9,10	High Levels

UNIVERSITY of GUELPH

LABORATORY SERVICES
Agriculture and Food Laboratory

FINAL Report
Submission#
Reported:

Submitted By:

Owner:

DNA Multiscan

Date Authorized: 2015-Oct-22 14:57

Sample ID Client Sample ID Specimen type Sampling date/time	**CHICKEN DIGESTATE** **Liquid Sample**
DNA Multiscan	Results Attached

Test method(s): PDC-114

Supervisor: Shannon Shan PhD, Agriculture and Food Laboratory 519 823 1268 ext. 57227 xshan@uoguelph.ca

FINAL Report

Submission#
Reported:

UNIVERSITY of GUELPH
LABORATORY SERVICES

DNA Water Scan Report

multiscan DNA

LSD Sample Number:
Submitter ID: Chicken digestate
Report Date: Oct 22 2015

Target Organism	Value	Result	1 to 10	Comments
Botrytis cinerea	0	Not Detected	0	
Fusarium spp.	0	Not Detected	0	
F. oxysporum	0	Not Detected	0	
F. solani	0	Not Detected	0	
Olpidium bornovanus	0	Not Detected	0	
O. brassicae	0	Not Detected	0	
O. virulentus	0	Not Detected	0	
Phytophthora spp.	0	Not Detected	0	
P. cactorum	0	Not Detected	0	
P. capsici	0	Not Detected	0	
P. cinnamomi	0	Not Detected	0	
P. cryptogea	0	Not Detected	0	
P. drechsleri	0	Not Detected	0	
P. fragariae	0	Not Detected	0	
P. infestans	0	Not Detected	0	
P. nicotianae	0	Not Detected	0	
Pythium spp.	0	Not Detected	0	
P. aphanidermatum	0	Not Detected	0	
P. dissotocum	0	Not Detected	0	
P. irregulare	0	Not Detected	0	
P. polymastum	0	Not Detected	0	
P. sylvaticum	0	Not Detected	0	
P. ultimum	0	Not Detected	0	
Rhizoctonia solani	0	Not Detected	0	
Sclerotinia spp.	0	Not Detected	0	
Thielaviopsis basicola	0	Not Detected	0	
Verticillium spp.	0	Not Detected	0	
V. albo-atrum	0	Not Detected	0	
V. dahliae	0	Not Detected	0	
V. dahliae (var longisporum)	0	Not Detected	0	

Legend

0	0	Not Detected
1	1,2,3	Low Levels
2	4,5,6	Moderate Levels
3	7,8,9,10	High Levels

Supervisor: Shannon Shan PhD, AFL, xshan@uoguelph.ca

Attachment 3 page 1 of 13

Nutrient Recycling Group	Standard Operating Procedure Bioreactor		Food and Bio-Processing Division
Written By: Erik Bergen Marc Legault	**Approved By:**	**Page 1 of 9**	**Rev 17 Nov. 2015**

1.0 APPLICATION

1.1 To provide a procedure for the operation of greenhouse #11 Bioreactor at CDC North.

2.0 INTRODUCTION

2.1 The CDC North bioreactor breaks down poultry manure (i.e. microbial mineralization) into plant-available nutrients by culturing uncharacterized bacterial species.

3.0 SAFETY WARNINGS and PRECAUTIONS

3.1 The bioreactor aerobically digests animal manures; ***E. coli, Salmonella*, influenza, and other potential pathogens are associated with manure and are a health hazard**.

3.1.1 It is important to minimize skin exposure and other pathways into the body (inhalation, eye contact, open wounds).

3.2 The bioreactor is dosed with strong acids and strong bases therefore, it is important to **always wear** the proper PPE to minimize exposure.

3.2.1 Always have MSDS on hand or readily be available.
3.2.2 Know where the eyewash stations are
3.2.3 Ensure and test a water-pressurized hose is nearby

3.3 If the dissolved oxygen level in the bioreactor falls - the process may become anaerobic:

3.3.1 If the bioreactor turns anaerobic, strong smells (sulphur based smells like rotten eggs) will be produced as well as methane gas which is a fire and health hazard.
3.3.2 A chemical respirator is recommended when cleaning the bioreactor.

3.4 The bioreactor is situated near many control boxes, electrical outlets, and breaker panels; avoid spraying water directly on electrical devices when cleaning the bioreactor.

3.5 The bioreactor is fitted with an agitator completed with submerged blades; this agitator is aggressively rotating within the tank, **avoid placing any body parts or equipment in the bioreactor when the agitator is in motion**.

CAUTION: *To avoid entanglement with the agitator do not wear clothing with draw strings or any other accessory that could be caught in the agitator; for this reason long hair must covered.*

4.0 REQUIRED PPE

4.1 Nitrile, latex gloves,

4.2 Steel toes boots (preferred), **close toed shoes are mandatory**
4.3 Long pants,
4.4 Safety glasses, a **face shield is mandatory when working with pH** control agents
4.5 Greenhouse coat (mandatory when working with tools or chemicals),
4.6 Dust mask [particulate and aerosol protection are available]
4.7 Chemical respirator (when cleaning the bioreactor),
4.8 Full Tyvek suit with hood (recommended while filling the bioreactor).

5.0 REQUIRED DOCUMENTATION

5.1 Bioreactor Control Form
5.2 Bioreactor Loading Control Form
5.3 Rosemount Analytical Transmitter and pH Calibration SOP and Control Form
5.4 Fermentation Run Log
5.5 Batch Record

6.0 RESPONSIBILITIES

6.1 It is the Responsibility of the Technologist:

6.1.1 To perform the daily checklist and routine analysis.
6.1.1.1 Includes the manual addition of any necessary reagents
6.1.2 To wear the proper PPE at all times.
6.1.3 To keep all PPE, lab supplies, equipment, chemicals, and reagents in supply.
6.1.4 To record all data and observations legibly in the appropriate note book(s).
6.1.5 To report any malfunctions or abnormal observations

6.2 It is the Responsibility of the Supervisor / Employer:

6.2.1 To determine when the bioreactor should be decanted or cleaned.
6.2.2 To address and fix any malfunctions or mechanical breakdowns.
6.2.3 To remotely monitor the bioreactor using mobile TeamViewer

7.0 DEFINITIONS

7.1 App: Denotes software application typically for 'smart phones'

7.2 Argus System: A greenhouse control and monitoring system - See Appendix for details

7.3 TeamViewer: The smartphone App / computer program used to remotely access and monitor Argus (See TeamViewer SOP).

7.4 Decant: The removal of the top fluid layer of the bioreactor without disturbing the settled sludge or sediment.

Attachment 3 page 3 of 13

7.5 PPE: Personal Protective Equipment needed to work safely work

7.6 **KH:** Carbonate Hardness; this indicates the alkalinity of a solution. Higher alkalinity means higher carbonates which results in higher buffering capacity. This ultimately means that there is less likelihood of dramatic pH swings.

7.7 **GH:** Stands General Hardness which is the measurement of a solutions magnesium and calcium concentrations. Since these are cations, they bind to counter ions thus favoring lower pH because there are less bound hydrogen ions in solution.

7.8 **Mother Liquor** The residual fermentation broth that is left in the bioreactor to be incorporated into the next batch.

7.9 **NH_3/NH_4^+:** This property is important because it assesses the primary rate of ammonification. The higher the ammonium concentration, the faster the metabolic breakdown of manure. Ammonium is a much more stable form of nitrogen storage and is less likely to go through a denitrification process. Values greater than ***5000 ppm in the bioreactor are preferred, however, 10, 000 ppm is typically the target value.***

7.10 **NO_3^-/ NO_2^-:** Must be at a much lower concentration than ammonium; this is a much less stable form for nitrogen storage as it can go through denitrification. The ***target value is 0 ppm.***

7.11 pH: The pKa of ammonia is about 9, this means that the target pH must be lower than 7 to strongly favour ammonium and avoid denitrification resulting in irreversible nutrient loss.

7.12 pKa Acid dissociation constant quantitative measure of an acid's strength solution

7.13 Conductivity: The measurement of electrolytic or specific conductance which is a solutions ability to carry an electric charge. This gives an general estimate of how concentrated a solution is;

7.13.1 drinking water is between 0.05 and 0.5mS /cm.

7.14 Dissolved Oxygen: Dissolved oxygen, dO, is the amount of oxygen present in solution. The bioreactor and stock tanks must be ***5 ppm dO_2 at minimum*** to avoid anaerobic conditions; however, the higher the concentration of dissolved oxygen the better to ensure complete aerobic digestion of the manure

8.0 PROCEDURE

Nutrient Recycling Group	Standard Operating Procedure Bioreactor		Food and Bio-Processing Division
Written By: Erik Bergen Marc Legault	**Approved By:**	**Page 4 of 9**	**Rev 17 Nov. 2015**

8.1 Bioreactor Initialization/Start-up

8.1.1 When required the bioreactor must be thoroughly cleaned with no sediment on tank walls or the floor.

8.1.1.1 The residual amount of Mother Liquor must be calculated.

8.1.2 Fill the bioreactor with approximately **500 L of water** (50% of the tank volume).

8.1.2.1 A measuring tape is used to estimate tank volume
See Bioreactor Volume Chart for details

8.1.3 Allow the bioreactor to 'acclimatize' overnight with the agitator operating in order to off gas chlorine and attain ambient temperature.

8.1.4 pH Probe Calibration

8.1.4.1 Calibrate the pH probes as per the calibration SOP

8.1.4.2 Place all spargers and probes into the bioreactor; [oxygen, both pH]

8.1.5 "Turn on" the agitator making sure it is spinning clockwise.

8.1.6 Prime pH agent pumps

NOTE: *Prime* base pump first then the acid pump to ensure the bioreactor is acidic prior to loading.

8.1.6.1 At the Local Argus control panel place acid and base pumps to 'OFF'

8.1.6.2 Weigh the pH agent containers – place into their respective tank

8.1.6.3 At the Local Argus control panel place base pump to 'ON'

8.1.6.3.1 Continue till base (with no air bubbles) enters the bioreactor

8.1.6.3.2 At Local Panel place base pump to 'OFF'

8.1.6.4 Repeat steps 8.1.3.1 to 8.1.3.3 inclusive for acid pump assembly.

8.1.7 Confirm bioreactor is acidic if not use the acid pump to dose acid

NOTE: *Pumps are left in 'OFF' setting till the manure is loaded into the bioreactor*

8.1.8 Set the oxygen pressure regulator to between **30-40 psi**.

8.1.8.1 The bioreactor dO should be at least 20 mg/L (20 ppm).

8.1.9 Place pH dosing pumps to AUTO for automatic pH control

8.1.10 Using the bioreactor loading record log to document all activities
- **8.1.10.1** Weigh and record the weight of each pail of manure **without the lid**
- **8.1.10.2** Remove a dry weight sample as per the bioreactor loading record log
- **8.1.10.3** Empty the pail into the bioreactor
- **8.1.10.4** Weigh and record the weight of each empty pail
- **8.1.10.5** Repeat steps 8.1.10.1 to 8.1.10.4 for all pails.

8.1.11 Closely monitor the bioreactor for the first 24 hours; there may be pH swings, over foaming, and oxygen level variations.

8.1.11.1 If the bioreactor is over foaming, add a surfactant as per batch record.

8.1.11.2 If the pH is not responding fast enough to the dosing pumps, you may have to manually add a pH agent.

8.1.11.3 The Batch Record will state the target

8.1.11.4 If the oxygen level gets below 5 mg/L (5ppm), increase the pressure gradually to 50 psi.

8.1.12 SAMPLE the Bioreactor as per the Batch Record

Note: *The bioreactor "typically runs" for 2 to 3 weeks before it is decanted into a stock tank*

8.2 Bioreactor Daily Checklist

8.2.1 Make sure the bioreactor is not over foaming or has a high foam level.
- **8.2.1.1** If it does, add a surfactant as the Batch Record.
- **8.2.1.2** The foam level should dissipate within minutes, if it does not; add an additional surfactant aliquot until it dissipates.

Note: ***The oxygen level will temporarily drop when surfactant is added.***

8.2.2 When required Check water level using a measuring tape to measure volume

8.2.2.1 If the water level is too low (below 50% tank volume), add water until it is at the approximate correct volume.

Nutrient Recycling Group	Standard Operating Procedure Bioreactor		Food and Bio-Processing Division
Written By: Erik Bergen Marc Legault	**Approved By:**	**Page 6 of 9**	**Rev 17 Nov. 2015**

8.2.3 Check the acid and base pails are at least 1/3 full.

8.2.3.1 If they are not add reagent so as Not to Lose pump prime

8.2.4 pH Control Boxes are beside the bioreactor. The values displayed on the control boxes should be within range of the hand held meters.

8.2.4.1 If the boxes read "- -. - -" or "9999" values, it means there is a problem with the probes. Inform supervisor immediately.

8.2.5 Oxygen Lines: Makes sure that there are no leaks from the oxygen tank to the bioreactor.

8.2.5.1 Make sure that the oxygen tank is set on an appropriate level at least 20 psi, but depends on the bioreactor oxygen level.

8.2.6 If required: Take samples for the biomass assay and start the assay (see Biomass and percent solids SOP).

8.2.7 Fill a 500 mL beaker with sample and perform the Routine analysis tests **Record ALL data on to the Fermentation Run Log**

8.2.7.1 Dissolved oxygen, dO, level using the appropriate meter
8.2.7.1.1 Should be at least 5 mg/L (5 ppm).

8.2.7.2 The electrical conductivity and pH using the appropriate meter (see EC and pH probe SOP).
8.2.7.2.1 The pH should be '+' or '-'0.3 pH units from the setpoint
8.2.7.2.2 If pH is lower than allowed, add 100mL of ammonium hydroxide every 30 min until the pH reaches desired range.
8.2.7.2.3 If the pH is greater than target range, add 100 mL of phosphoric acid every 30 min until the pH reaches desired range.

NOTE: *If pH was not within target range inspect the dosing pumps and the Argus system to determine cause of pH control failure.*

8.2.7.3 The temperature can be checked using the hand held EC/pH meter.

NOTE: *If the bioreactor is not above room temperature, contact supervisor immediately*

8.2.7.4 Test nitrate/ nitrite levels (NO_2^- / NO_3^-, general hardness (GH), carbonate hardness (KH), and ammonium / ammonia levels (NH_3 / NH_4^+). (See Colorimetric Indicator SOP).

Attachment 3 page 7 of 13

8.2.7.4.1 The ideal nitrate and nitrite levels should be 0 ppm, but they should be at least 2 orders of magnitude lower than the ammonium / ammonia levels.

8.2.7.4.2 The ideal ammonia / ammonium levels are at least 5000 ppm but they should be at least 2 orders of magnitude about the nitrate / nitrite levels.

8.2.7.5 All of these tests can be done while waiting for the biomass / percent solids filtration step to finish.

8.2.8 Record all information and all observations into the designated bioreactor data sheet including the date and time.

8.2.9 Cross reference current temperature readings, pH readings, and oxygen level readings with Argus to validate the current stats of the bioreactor.

8.3 Decanting the Bioreactor

8.3.1 Turn off the agitator, oxygen flow, and dosing pumps (placed in 'OFF').

8.3.2 Wait at least overnight

8.3.3 Place a sump pump just under the surface of the water level and the connecting hose into the tank you wish to decant into.

8.3.4 Plug in the pump and slowly lower the pump into the bioreactor so the suction is always just beneath the water level.

8.3.4.1 The fluid should be a light to medium brown color with a watery consistency.

8.3.5 *STOP* decanting when the fluid starts to become a dark brown color and the consistency changes to a thicker sandy or sludgy consistency.

NOTE: ***Decanting is always done with 2 people present; one person focusing on lowering the pump, and the other focusing on the fluid coming out of the hose.***

8.4 Cleaning the Bioreactor

8.4.1 Turn off the bioreactor agitator and dosing pumps

8.4.2 Take the pH probes out of the bioreactor, gently clean them with a squirt bottle and swipes.

Attachment 3 page 8 of 13

8.4.2.1 Never touch the membrane or spray directly on it.

8.4.2.2 After they are cleaned, place the probes in a 5M potassium chloride (KCl) buffer or pH Buffer 4 Standard solution.

8.4.3 Remove out all other probes and oxygen spargers.

8.4.3.1 Turn off the oxygen 12- 24 hours before you clean / decant the tank.

8.4.4 Decant the bioreactor as described in the previous section.

8.4.5 Then the solution in the bioreactor becomes thick, constantly agitate and break up the sediment using a water jet while decanting.

8.4.5.1 This step is best done with 3 people.

8.4.5.2 As the solution gets thicker, the pump with start to clog.

8.4.5.2.1 Keep adding water to dilute the sediment

8.4.6 Once the volume level is less than 40 L, vacuum the remaining sediment and liquid out.

9.0 TRAINING REQUIREMENTS

9.1 Basic understanding of smartphones and windows operating system.
9.2 Proper bioreactor monitoring and routine analysis.
9.3 WHMIS.
9.4 Standard First Aid.

10.0 REFERENCES/RELATED SOPS

10.1 Bioreactor Design for Chemical Engineers, Gregory T. Benz, Benz Technology International Inc. (PDF Digital Copy Available).
10.2 Bioreactor Operations (PDF Digital Copy Available).
10.3 TeamViewer SOP
10.4 Argus SOP
10.5 Dilutions SOP
10.6 Mixing Solutions SOP
10.7 Colorimetric Indicator SOP
10.8 pH and Conductivity Meter SOP
10.9 Biomass and percent solids SOP

11.0 APPENDIX

Attachment 3 page 9 of 13

11.1 Argus System

11.1.1 The Argus System is a greenhouse control and data acquisition.

11.1.2 For the purposes of the bioreactor, Argus monitors the dissolved oxygen level, monitors and controls the pH level, and monitors the temperature.

11.1.2.1 The pH control is regulated by 2 probes, once guard probe that acts as a reference, and once control probe that tells the system to increase to decrease the pH of the system.

11.1.2.1.1 The information from these probes is stored in the Argus system software and should be monitored daily.

11.1.2.1.2 The probes should also be inspected on a weekly basis for biofilms and sediments blocking the membrane.

11.1.3 The Argus system is important for two main purposes:

11.1.3.1 The storing of important analytical data of the greenhouse and bioreactor systems and graphing this data to see trends used for process optimization.

11.1.3.2 The controlling of various functions such as pH, foam levels, and possibly automatic feeding of the bioreactor with manure and other nutrients, and possibly the controlling of the bioreactor temperature.

11.1.4 See the Argus SOP for specific instructions on how to operate the system.

11.1.4.1 For any assistance with the Argus system, call the Argus contact Jeff @ 1-888-667-2091 Ext. 115.

12.0 REVISION HISTORY

Version	Effective Date	Summary of Change

Attachment 3 page 10 of 13

Alberta Agriculture and Forestry **Bioreactor Control Form** **Nutrient Recycling Group**

1.0 APPLICATION To Provide a Documentation Form to Capture all Setup Data for the Operation of Greenhouse #11 Bioreactor at CDC North.

2.0 FERMENTATION DOCUMENTATION

2.1 Trial _____ **Date** _____ **Time** _____

2.2 Objective(s)

Trial pH setpoint
O_{xygen} lpm

3.0 PRE-FERMENTATION CHECKLIST

3.1 Adequate reagent supplies:

3.1.1 Acid for pH control Acid Type _____

3.1.2 Base for pH control Base Type _____

3.1.3 pH standards for pH calibration **Yes / No** **Type** _____

3.1.4 Surfactant (anti-foam agent) **Yes / No** **Type** _____

3.1.5 Sufficient feedstock (manure or other) **Yes / No**

3.1.6 Oxygen Purity _____

3.2 Bioreactor Preparation

3.2.1 Estimate of Residual Mother Liquor: _____

3.2.2 Initial Bioreactor Volume (typically allowed to warm up overnight): _____

3.2.3 All Probes Removed and Cleaned: Yes / No

3.2.4 Oxygen Sparger(s) Removed and Cleaned: Yes / No

3.3 Reagent **Note: If Lid is Present for the Weight**

3.3.1	Weight of Acid Before the Run:		kg
3.3.2	Weight of Acid after the Run:		kg
3.3.3	**Acid used**		**kg**

ACIDF Project \\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\Templates\
Bioreactor Control Form rev 31 Mar. 2016 .xlsxBioreactor Control Form rev 31 Mar. 2016 .xlsxControl Form Loading

Alberta Agriculture and Forestry | Bioreactor Control Form | Nutrient Recycling Group

3.3.4	Weight of Base Before the Run:		kg
3.3.5	Weight of Base after the Run:		kg
3.3.6	Base used		kg

3.4 Calibration of pH Control and Guard Probes

3.4.1 Control pH probe slope mV/pH_____ Offset _____ _____ mV/pH

3.4.2 Guard pH probe slope mV/pH_ _____ Offset _____ _____ mV/pH

Slope must be within 59.1 and 47 mV per pH unit or probe has failed.

3.4.3 Standardize Probes to Hand Held

Immerse hand-held probe in bioreactor allow to reach steady state i.e. equal temperature values with guard and control probes

3.4.4 Guard, Control Probes Standardized to _____ Hand-held pH value while immersed in bioreactor

Oxygen Sparger is nearest to the Acid / Base Outlets

dO probe is above tank floor to avoid fouling due to sediment.

3.5 All Probes Are Installed: Yes / No

3.6 Argus is Recording Data Yes / No

3.6.1 Argus pH Setpoint _____ Bandwidth _____

Prime Base Pump First Then Acid Pump

3.7.1 Dosing Pumps Primed Yes / No _____

3.7.2 If required initial acid aliquot prior to loading _____

3.7.3 Bioreactor Loading Control Form Completed Yes / No [Field Data Entries]

3.8 **Dosing Pumps Returned to 'Auto'** **Yes / No**

3.9 Bioreactor Loading

3.9.1 All Pails of Feedstock Emptied and Rinsed Yes / No

3.9.2 Agitator is Rotating Correctly Yes / No _____

3.10 Bioreactor Volume Agitation Must be 'Off' Prior to Measurement or Indicate rpm speed

3.10.1 Bioreactor Volume After Loading Liters

3.11 **Sample Bioreactor for t=0** Date Time

Nutrient Recyc Group

ACIDF ProjectAerobic D stion of Poultry Manure

Alberta Agric re and Forestry

% Dry Weight Control Form

Trial Date Time

	Total Weight of Wet Manure			% Dry Weight Determination					Record Observations on Back of Page	
	A	B	C = A - B	D	E	F	G = (E - F)/(E-D) *100	H =(F-D)/(E-D) *100	= H*C/100	
Pail # / **Sample**	**No Lid** Full Manure Pail kg	**No Lid** Empty Pail kg	**Σ Wet** [H_2O + Manure] **Weight** kg	Empty Foil Tray gm	Foil Tray + Wet Manure gm	Foil Tray + Dry Manure gm **Dried 60°C**	% Moisture	**% DM Dry Matter**	**Dry Manure Fed to Bioreactor**	**Manure Description, Source etc.**
1							#DIV/0!	#DIV/0!	#DIV/0!	
2							#DIV/0!	#DIV/0!	#DIV/0!	
3							#DIV/0!	#DIV/0!	#DIV/0!	
4							#DIV/0!	#DIV/0!	#DIV/0!	
5							#DIV/0!	#DIV/0!	#DIV/0!	
6							#DIV/0!	#DIV/0!	#DIV/0!	
7							#DIV/0!	#DIV/0!	#DIV/0!	
8							#DIV/0!	#DIV/0!	#DIV/0!	

Attachment 3 page 12 of 13

Alberta Agriculture and Forestry | ACIDF Project 2016 Bioreactor Routine Check | Nutrient Recycling Group

Trial # START:

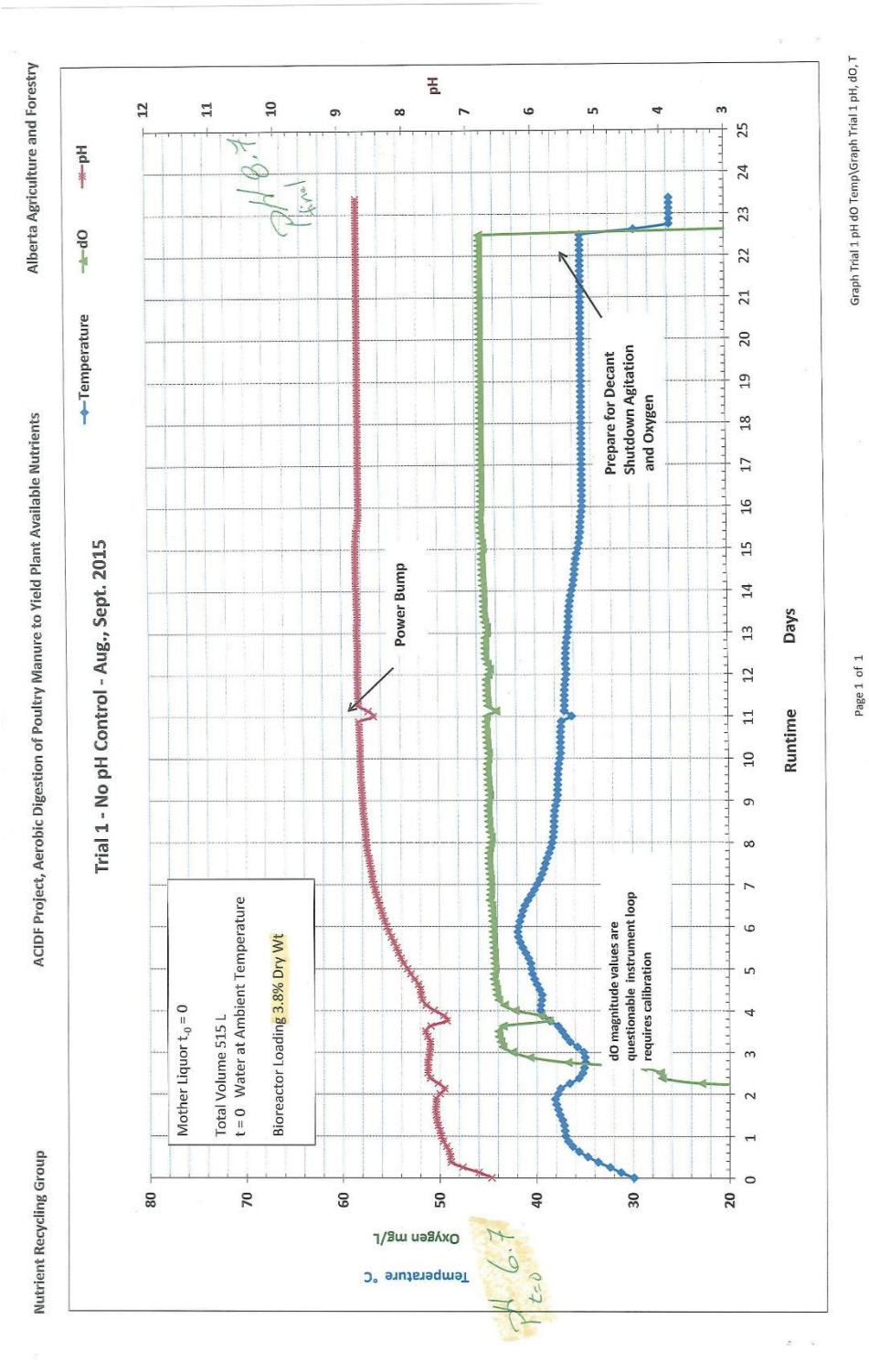
END:

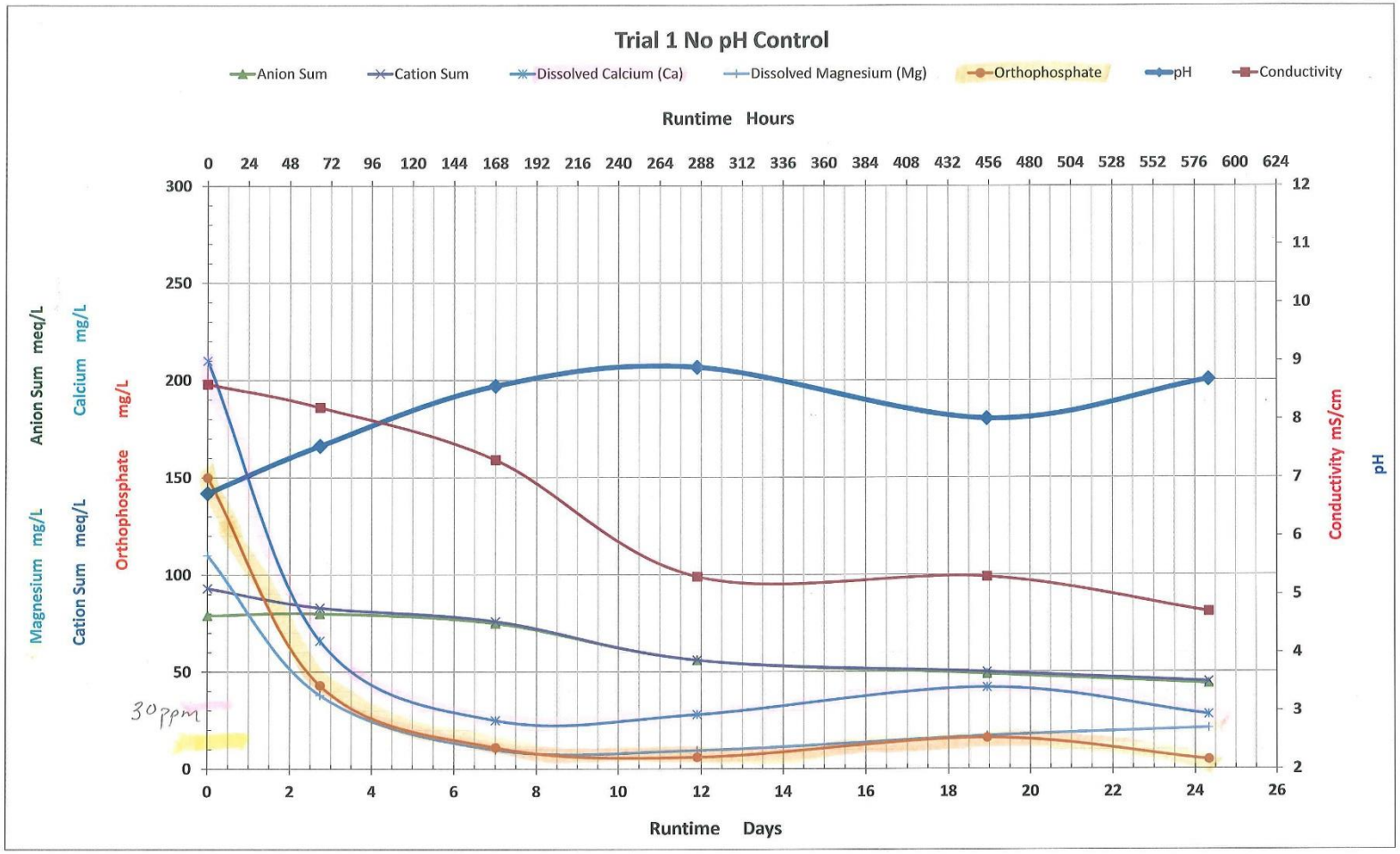
		pH Control Loop												
		pH Guard Loop		pH Control Loop							Control Oxygen lpm	Fixed Oxygen lpm		
Date yyyy mm dd	Time 24:00	Temp °C	pH	Temp °C	pH	Temp °C	pH	EC ms/cm	dO mg/L	rpm			Antifoam	Notes indicate Sample Points, antifoam additions, etc.

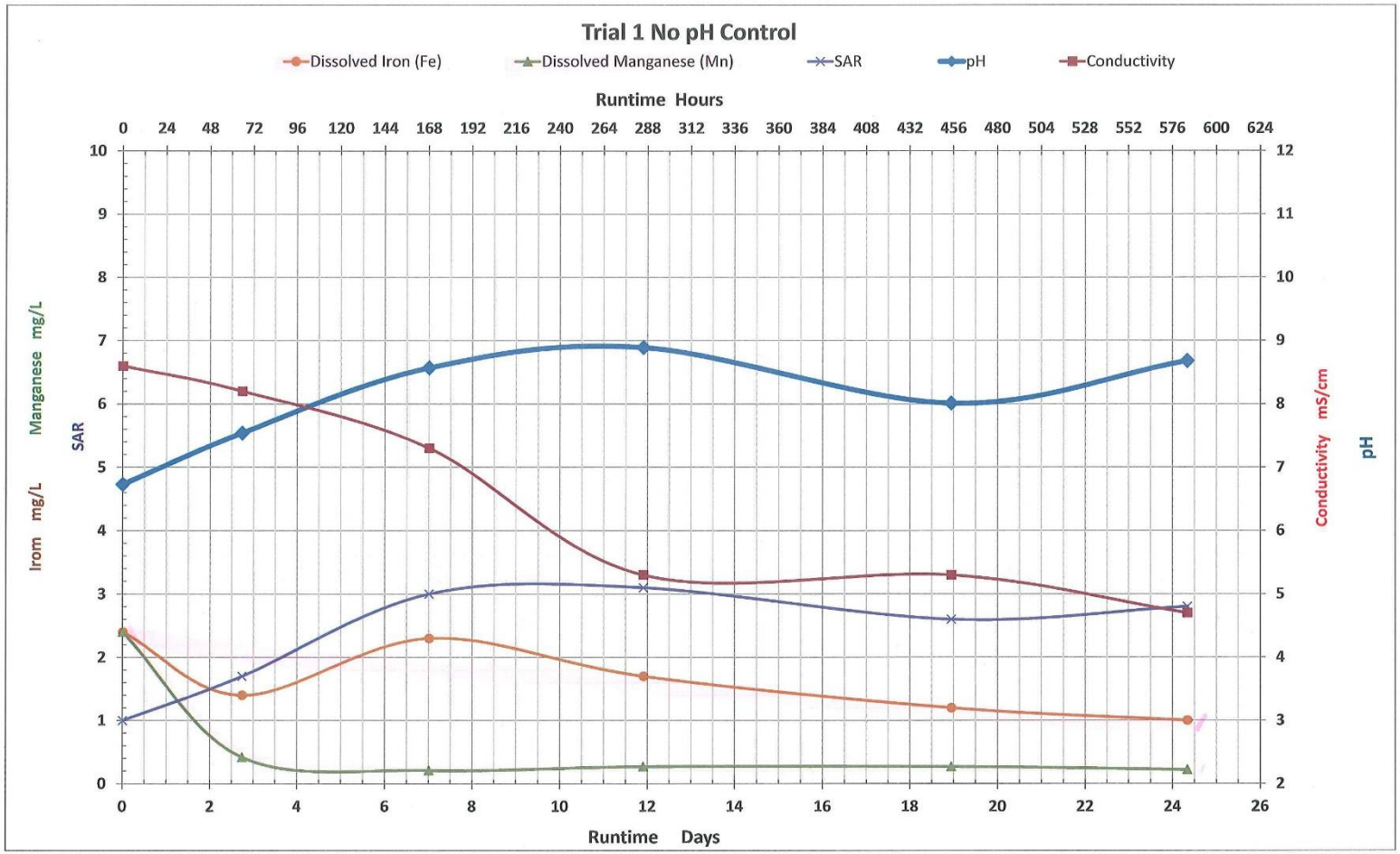
Attachment 3 page 13 of 13

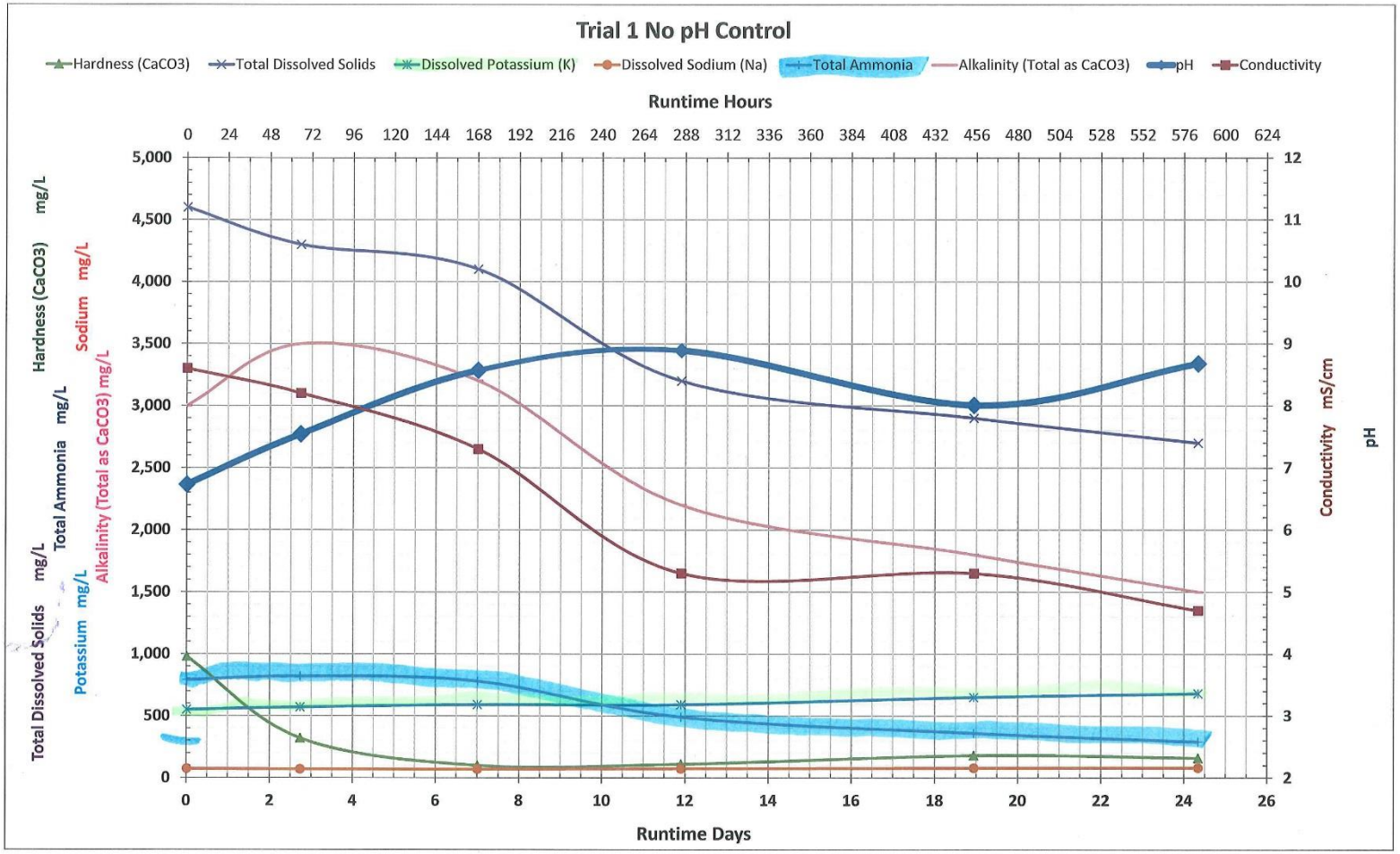
Attachment 4 – Fermentation and Nutrient Graphs

Trial 1




Trial 1 No pH Control
Anion Sum
Cation Sum
Dissolved Calcium (Ca)
Dissolved Magnesium (Mg)
Orthophosphate
pH
Conductivity
Runtime Hours
0 24 48 72 96 120 144 168 192 216 240 264 288 312 336 360 384 408 432 456 480 504 528 552 576 600 624
Anion Sum meq/L
Calcium mg/L
Orthophosphate mg/L
Magnesium mg/L
Cation Sum meq/L
Conductivity mS/cm
pH
300 250 200 150 100 50 0
12 11 10 9 8 7 6 5 4 3 2
30ppm
0 2 4 6 8 10 12 14 16 18 20 22 24 26
Runtime Days


Trial 1 No pH Control
Dissolved Iron (Fe)
Dissolved Manganese (Mn)
SAR
pH
Conductivity
Runtime Hours
0 24 48 72 96 120 144 168 192 216 240 264 288 312 336 360 384 408 432 456 480 504 528 552 576 600 624
Irom mg/L
Manganese mg/L
SAR
Conductivity mS/cm
pH
0 1 2 3 4 5 6 7 8 9 10
0 2 3 4 5 6 7 8 9 10 11 12
0 2 4 6 8 10 12 14 16 18 20 22 24 26
Runtime Days



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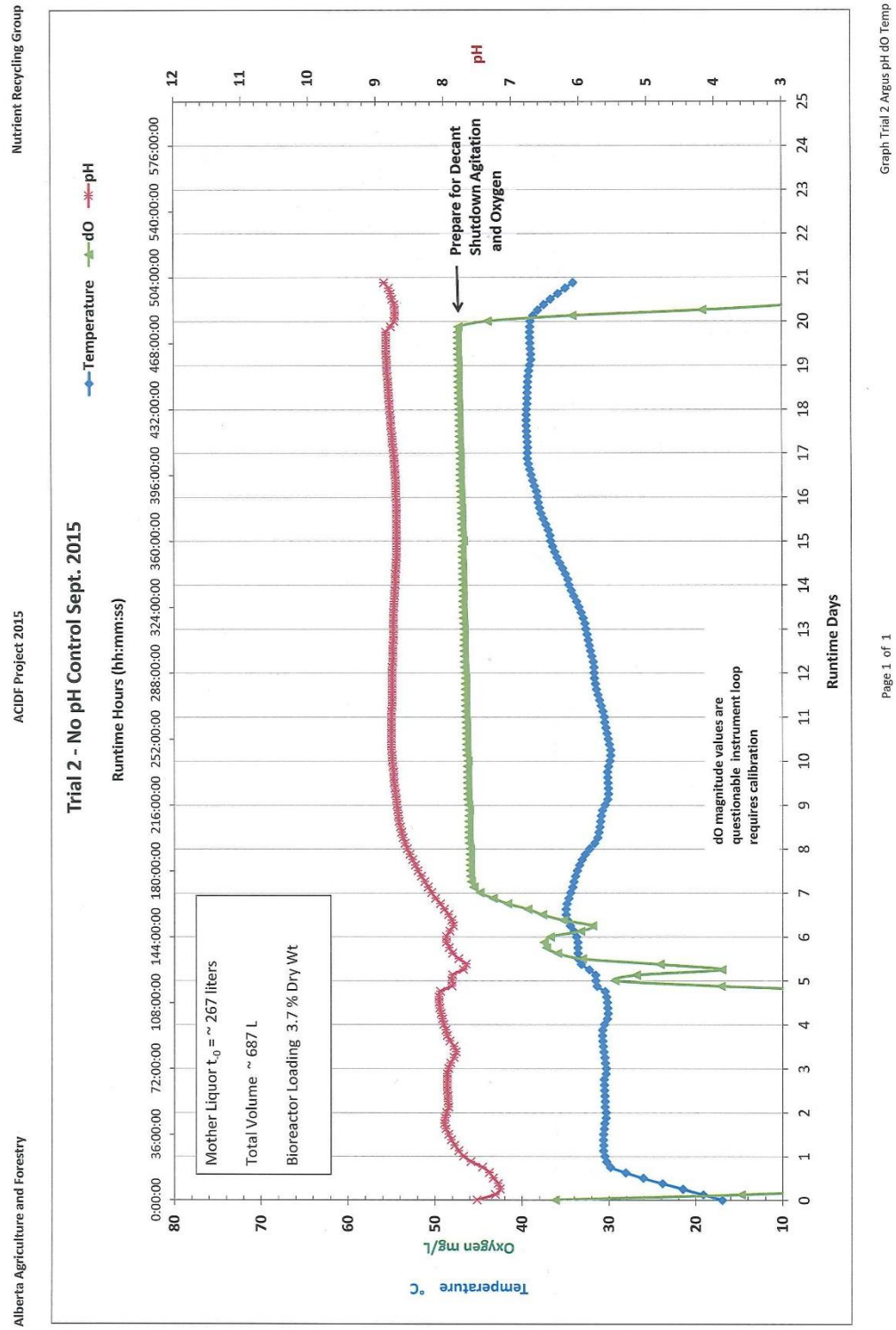
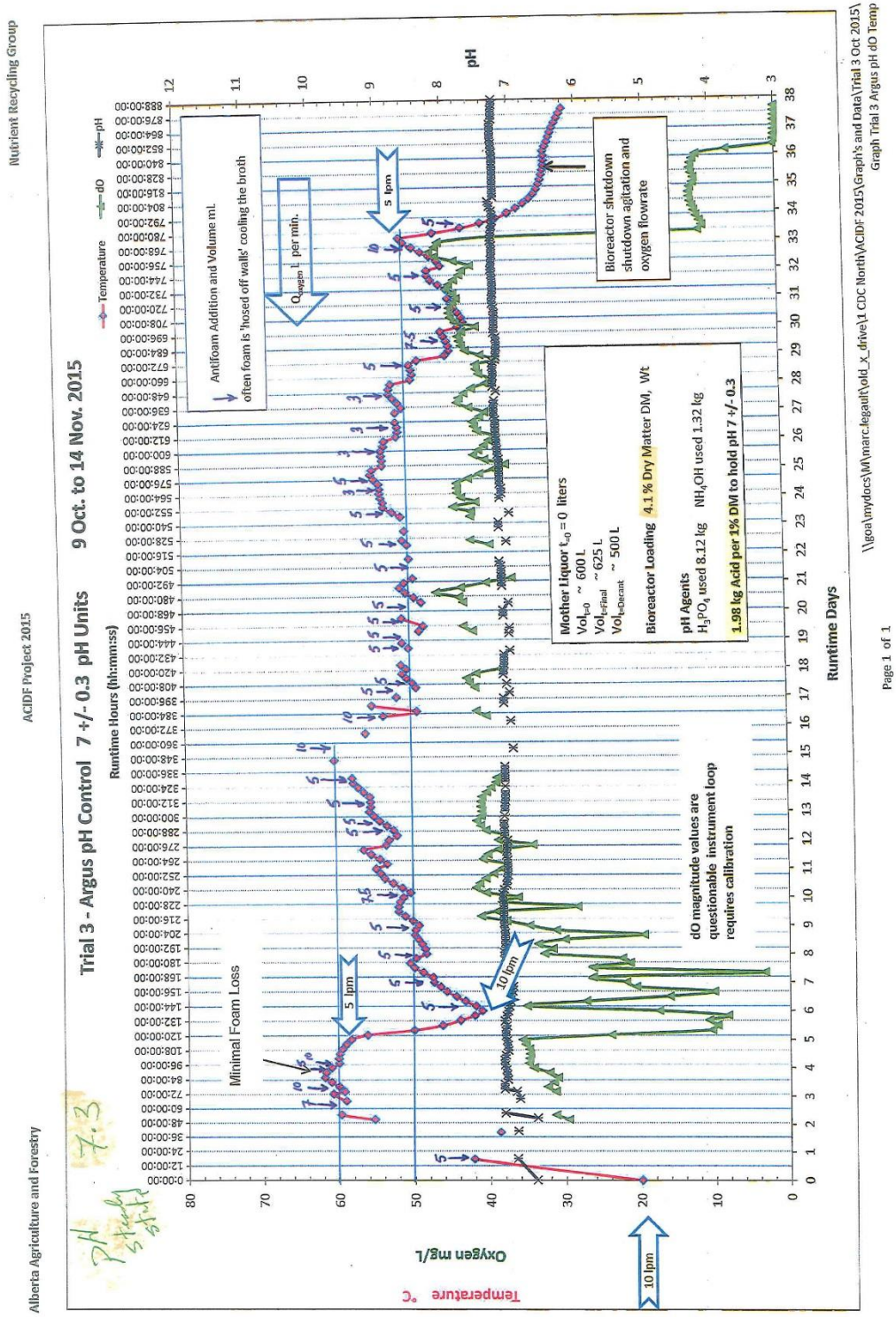
Trial 1 - Maxxam Data 20 Oct. 2015.xls\Chart 1


Alberta Agriculture and Forestry
ACIDF Project 2015
Nutrient Recycling Group
Trial 2 - No pH Control Sept. 2015
Temperature
dO
pH
Runtime Hours (hh:mm:ss)
Runtime Days
Temperature °C
Oxygen mg/L
pH
Prepare for Decant Shutdown Agitation and Oxygen
Mother Liquor t_0 = ~ 267 liters
Total Volume ~ 687 L
Bioreactor Loading 3.7 % Dry Wt
dO magnitude values are questionable instrument loop requires calibration
Graph Trial 2 Argus pH dO Temp
Page 1 of 1


Alberta Agriculture and Forestry
ACIDF Project 2015
Nutrient Recycling Group
Trial 3 - Argus pH Control 7 +/- 0.3 pH Units 9 Oct. to 14 Nov. 2015
Temperature
dO
pH
Runtime Hours (hh:mm:ss)
Runtime Days
Minimal Foam Loss
Antifoam Addition and Volume mL
often foam is 'hosed off walls' cooling the broth
Bioreactor shutdown
shutdown agitation and
oxygen flowrate
Mother Liquor $t_{=0}$ = 0 liters
$Vol_{t=0}$ ~ 600 L
$Vol_{t=Final}$ ~ 625 L
$Vol_{t=Decant}$ ~ 500 L
Bioreactor Loading 4.1 % Dry Matter DM, Wt
pH Agents
H_3PO_4 used 8.12 kg NH_4OH used 1.32 kg
1.98 kg Acid per 1% DM to hold pH 7 +/- 0.3
dO magnitude values are
questionable instrument loop
requires calibration
5 lpm
10 lpm
Temperature °C
Oxygen mg/L
pH
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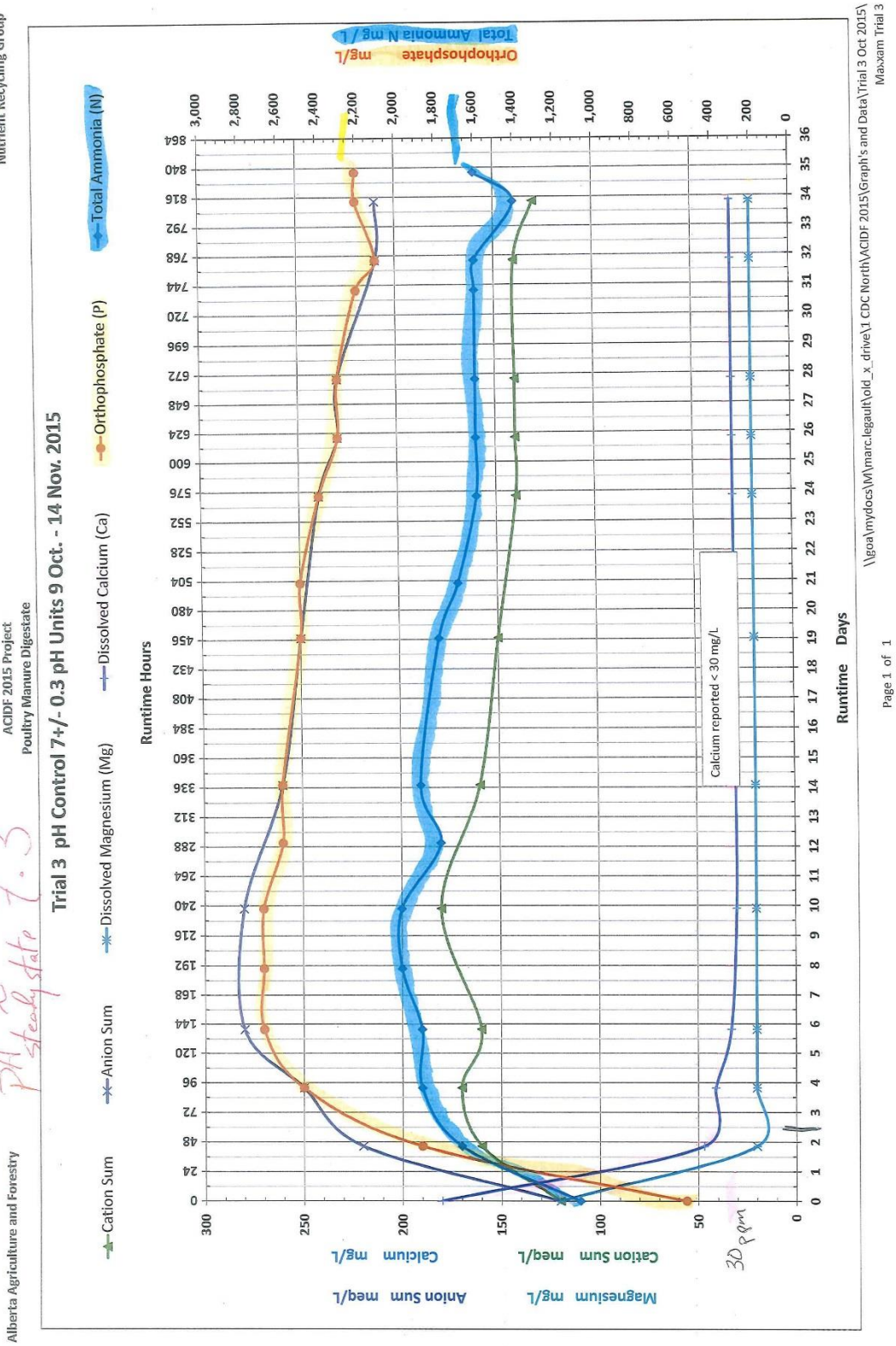
Graph Trial 3 Argus pH dO Temp
Page 1 of 1


Nutrient Recycling Group
ACIDF 2015 Project
Poultry Manure Digestate
Alberta Agriculture and Forestry
Trial 3 pH Control 7+/- 0.3 pH Units 9 Oct. - 14 Nov. 2015
pH ~ 7.3 steady state
Cation Sum
Anion Sum
Dissolved Magnesium (Mg)
Dissolved Calcium (Ca)
Orthophosphate (P)
Total Ammonia (N)
Runtime Hours
Runtime Days
Anion Sum meq/L
Calcium mg/L
Cation Sum meq/L
Magnesium mg/L
Orthophosphate mg/L
Total Ammonia N mg/L
Calcium reported < 30 mg/L
30 ppm
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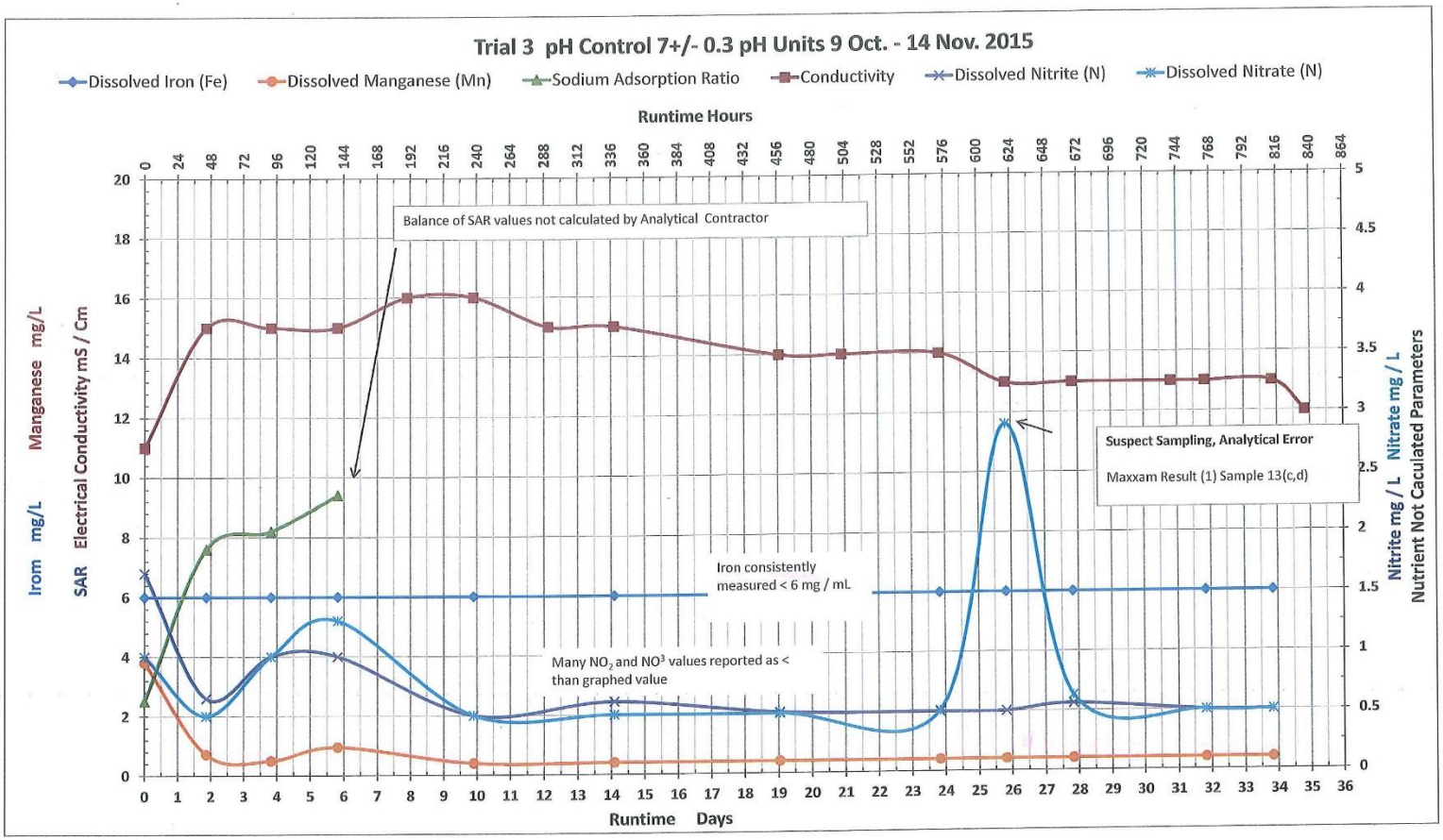
Max.xam Trial 3
Page 1 of 1



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Maxxam Trial 3.xls
Chart 2

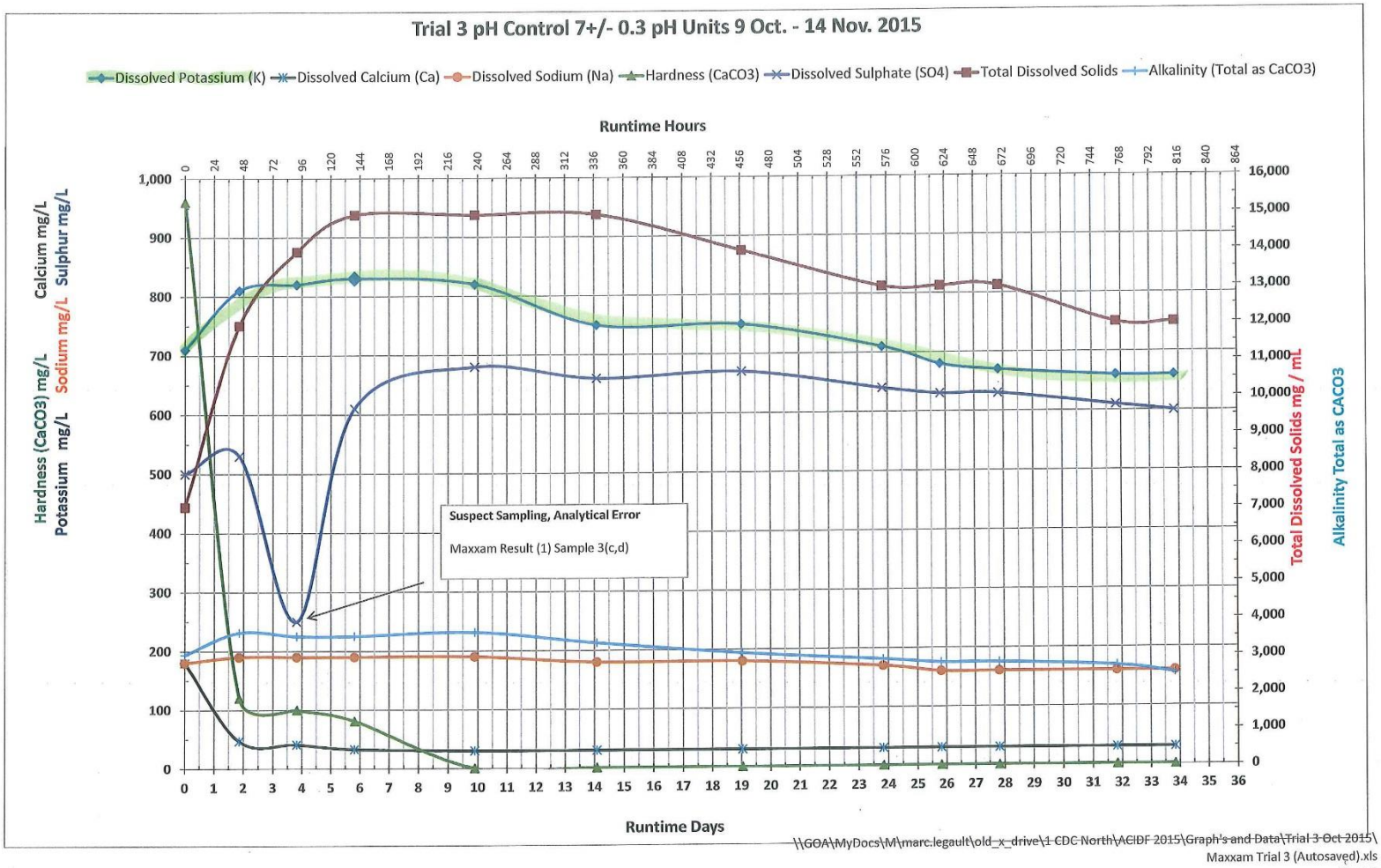

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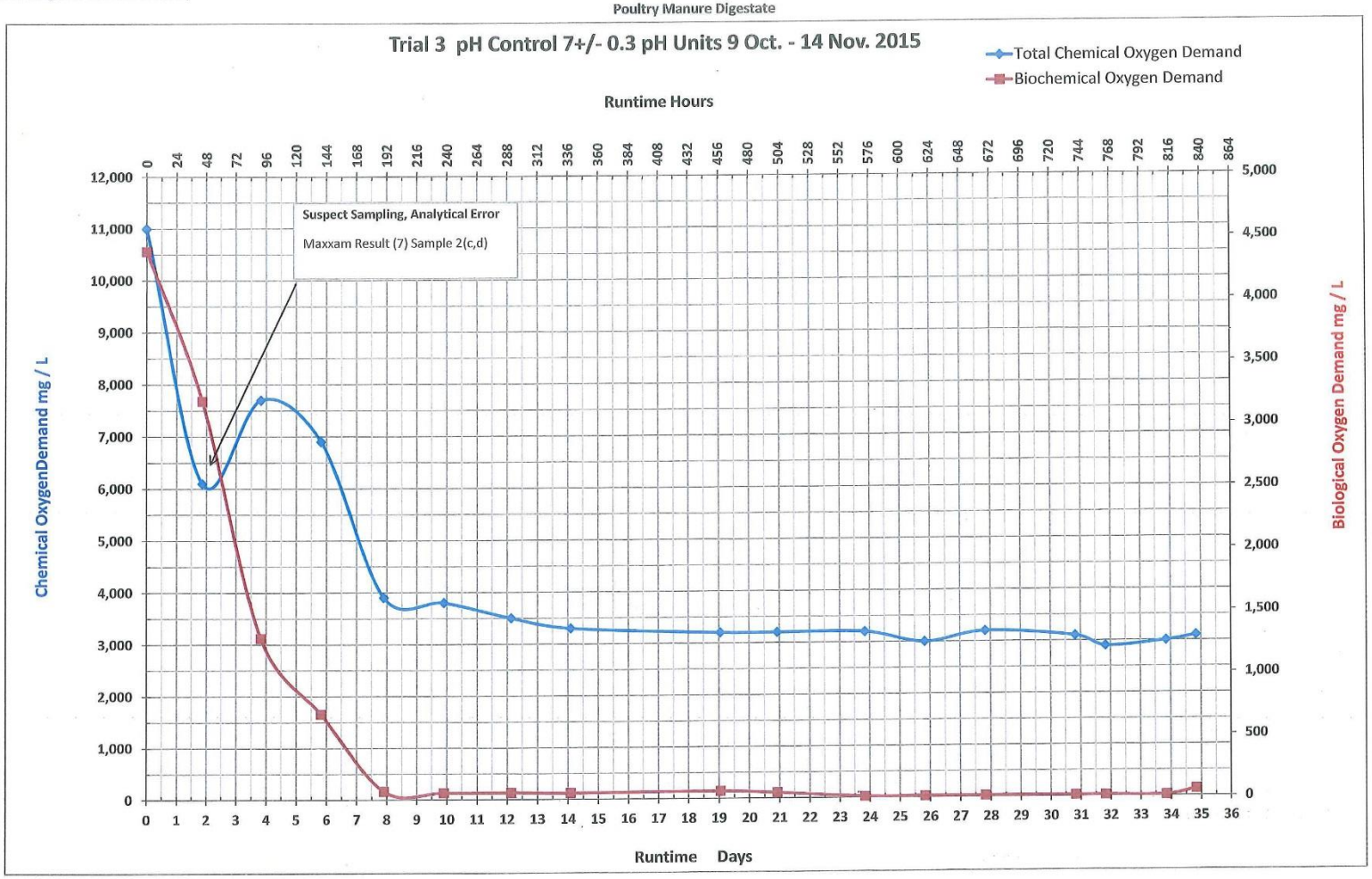
Chart 1

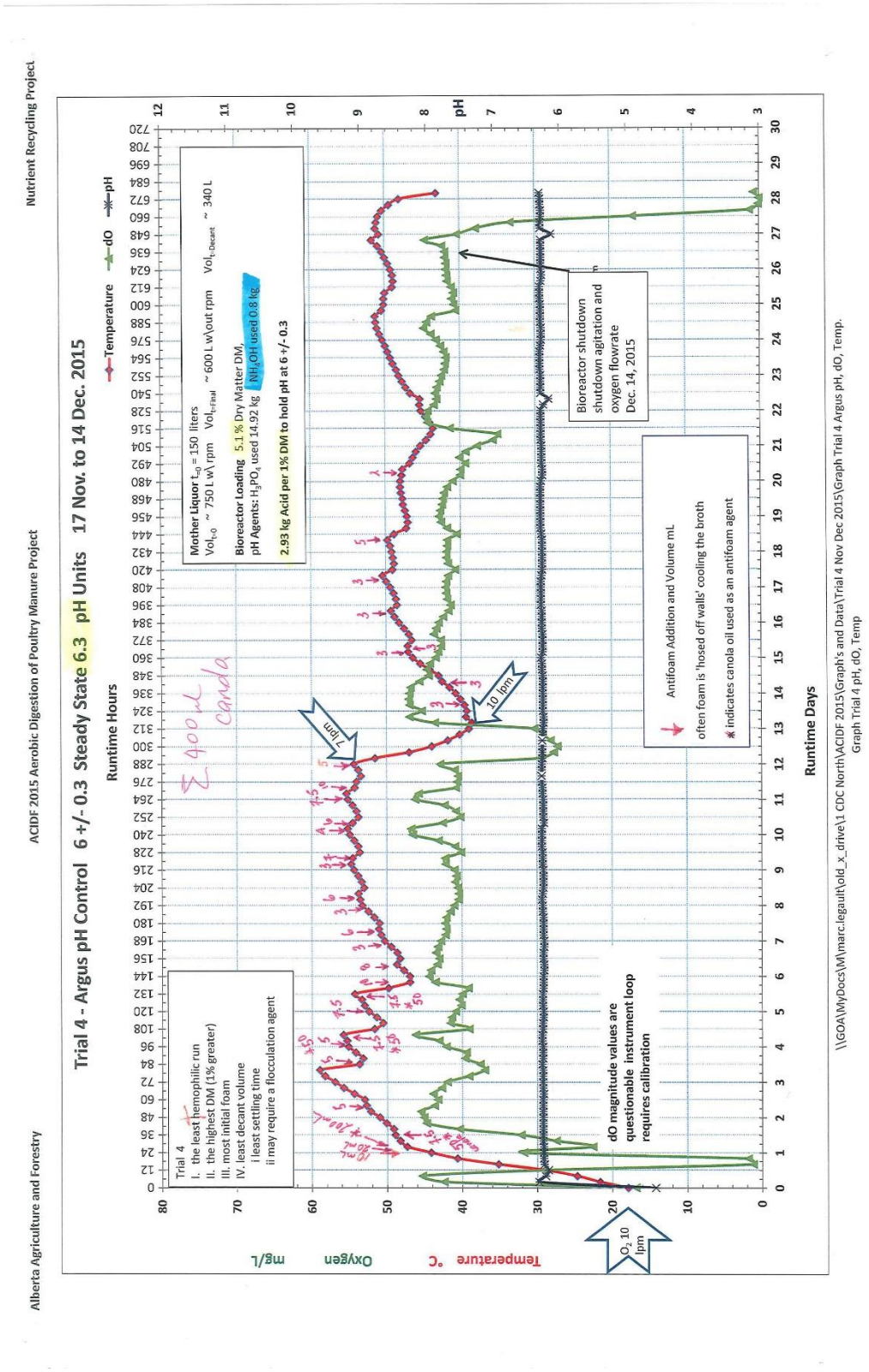
Alberta Agriculture and Forestry

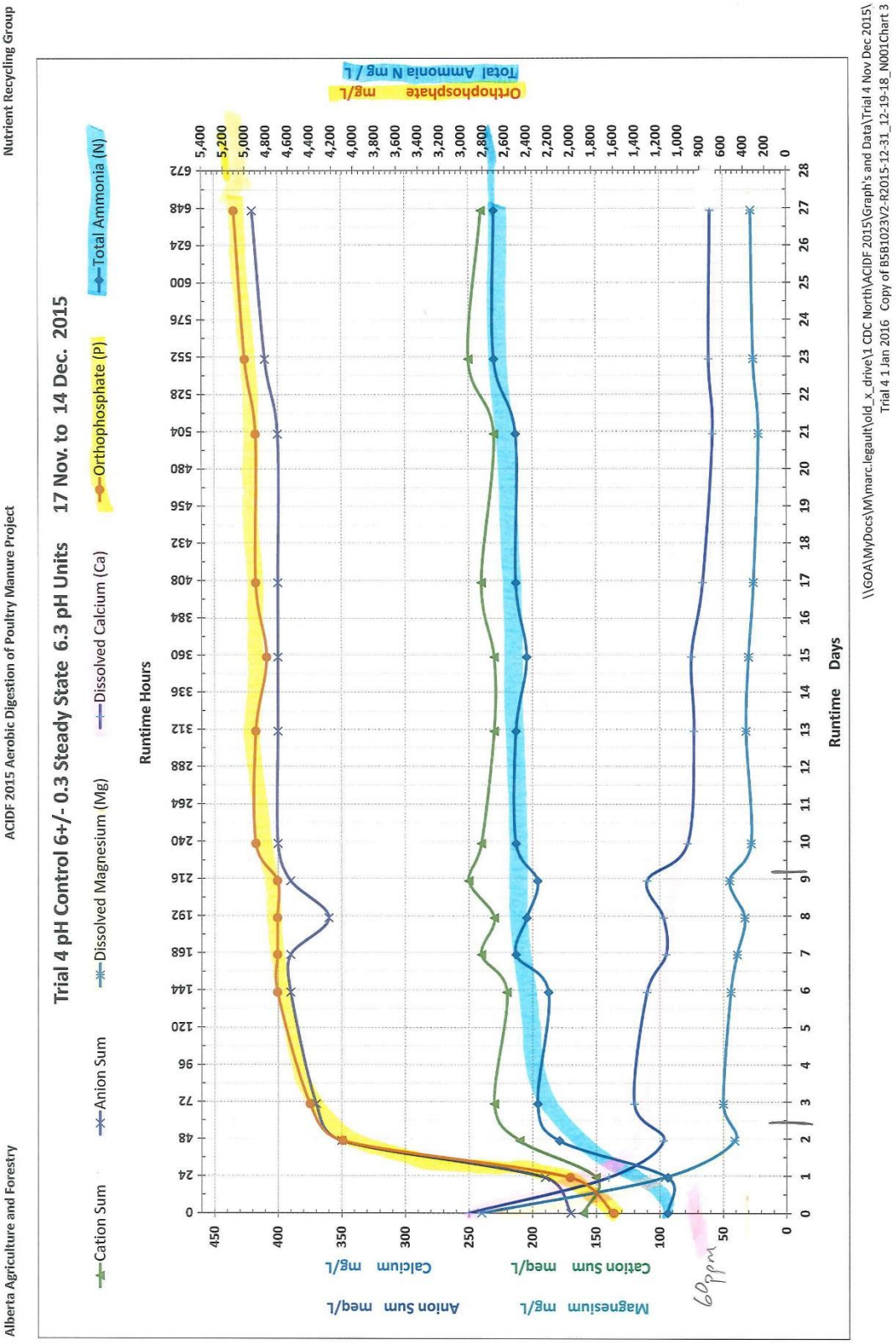
ACIDT 2015 Project
Poultry Manure Digestate

Nutrient Recycling Group



Trial 4


Alberta Agriculture and Forestry
ACIDF 2015 Aerobic Digestion of Poultry Manure Project
Nutrient Recycling Project
Trial 4 - Argus pH Control 6 +/- 0.3 Steady State 6.3 pH Units 17 Nov. to 14 Dec. 2015
Temperature
dO
pH
Runtime Hours
Runtime Days
Mother Liquor $t_{=0}$ = 150 liters
$Vol_{t=0}$ ~ 750 L w\ rpm
$Vol_{t=Final}$ ~ 600 L w\out rpm
$Vol_{t=Decant}$ ~ 340 L
Bioreactor Loading 5.1 % Dry Matter DM,
pH Agents: H_3PO_4 used 14.92 kg
NH_4OH used 0.8 kg
2.93 kg Acid per 1% DM to hold pH at 6 +/- 0.3
Trial 4
I. the least hemophilic run
II. the highest DM (1% greater)
III. most initial foam
IV. least decant volume
i least settling time
ii may require a flocculation agent
7 lpm
10 lpm
Bioreactor shutdown agitation and oxygen flowrate Dec. 14, 2015
Antifoam Addition and Volume mL
often foam is 'hosed off walls' cooling the broth
indicates canola oil used as an antifoam agent
dO magnitude values are questionable instrument loop requires calibration
O_2 10 lpm
Temperature °C
Oxygen mg/L
pH
\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015\Graph's and Data\Trial 4 Nov Dec 2015\Graph Trial 4 Argus pH, dO, Temp.
Graph Trial 4 pH, dO, Temp


Alberta Agriculture and Forestry
ACIDF 2015 Aerobic Digestion of Poultry Manure Project
Nutrient Recycling Group
Trial 4 pH Control 6+/- 0.3 Steady State 6.3 pH Units 17 Nov. to 14 Dec. 2015
Cation Sum
Anion Sum
Dissolved Magnesium (Mg)
Dissolved Calcium (Ca)
Orthophosphate (P)
Total Ammonia (N)
Runtime Hours
Runtime Days
Anion Sum meq/L
Calcium mg/L
Cation Sum meq/L
Magnesium mg/L
Orthophosphate mg/L
Total Ammonia N mg/L
60ppm
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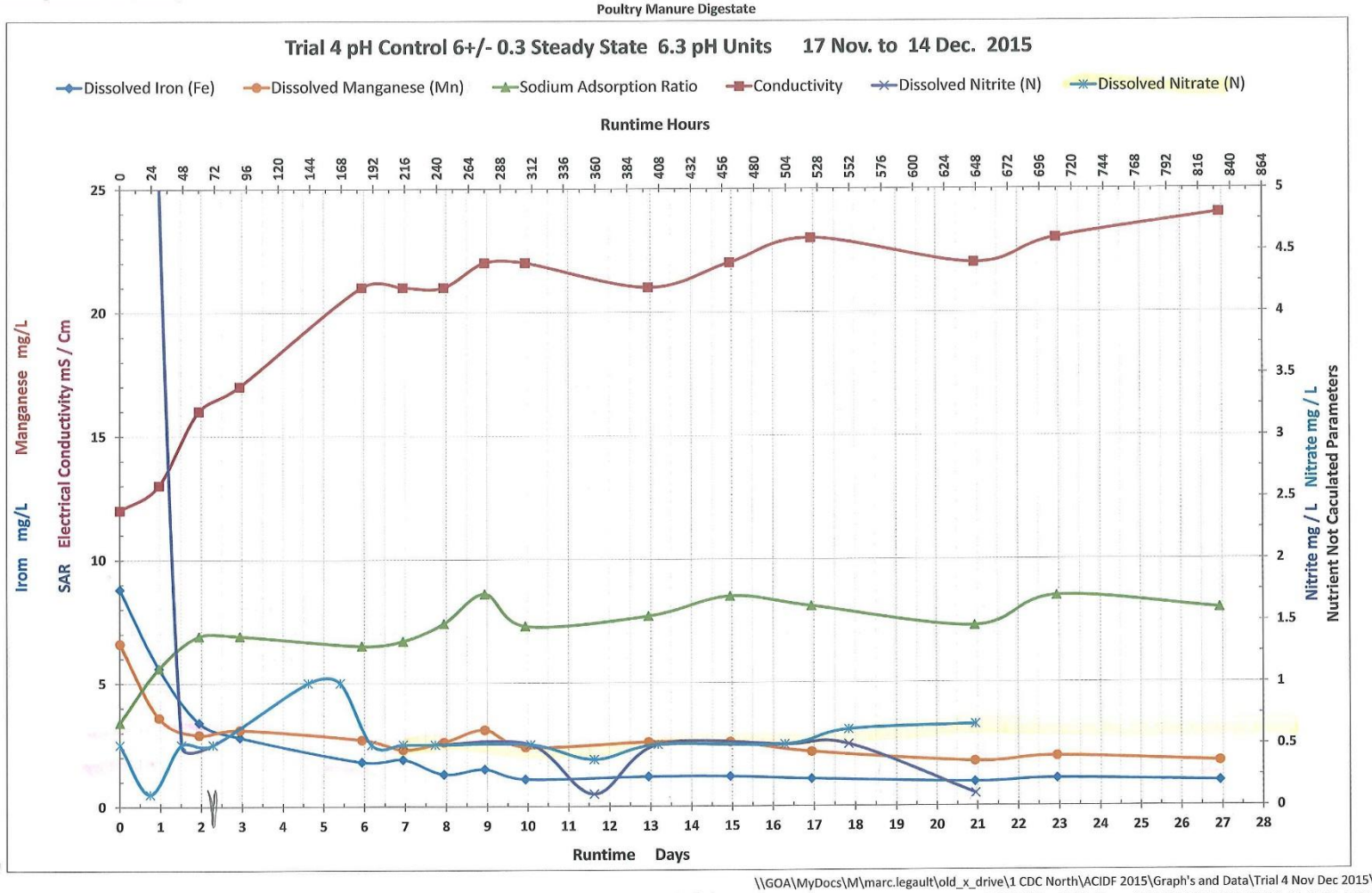
Trial 4 1 Jan 2016 Copy of B5B1023V2-R2015-12-31_12-19-18_N001Chart 3

Alberta Agriculture and Forestry

ACIDF 2015 Project
Poultry Manure Digestate

Nutrient Recycling Group



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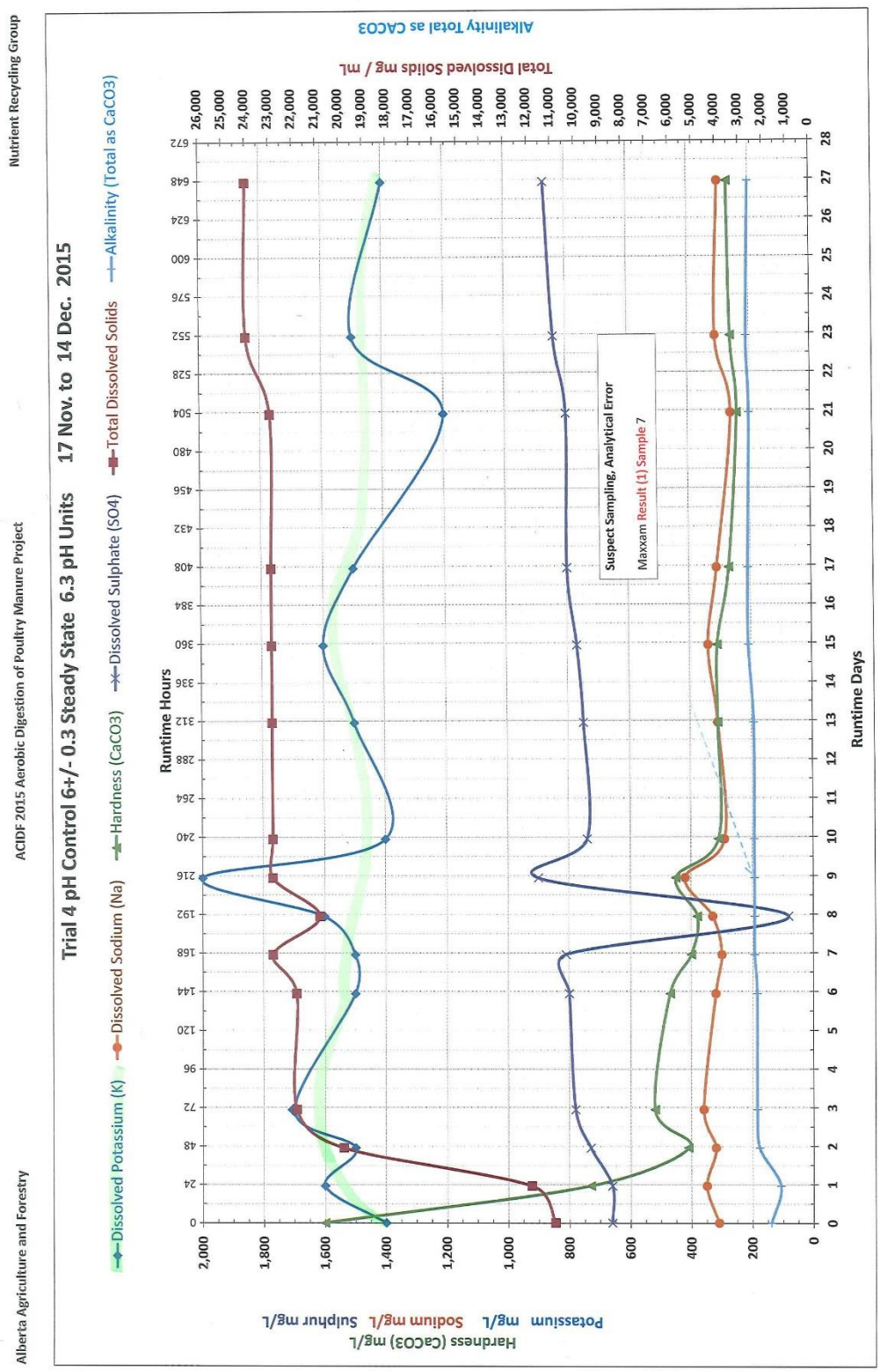
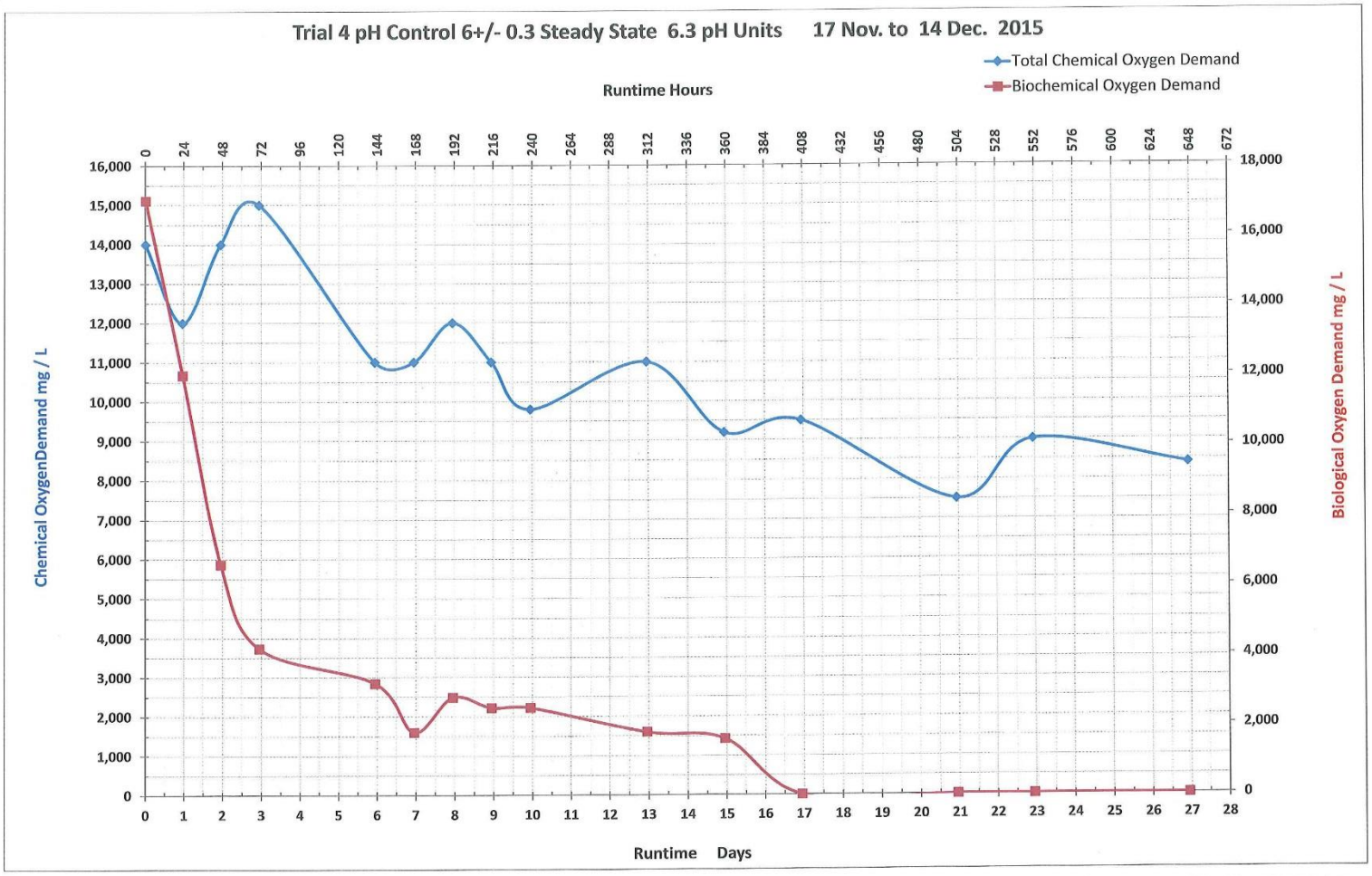
Trial 4 1 Jan 2016 Copy of B5B1023V2-R2015-12-31_12-19-18_N001


Nutrient Recycling Group
ACIDF 2015 Aerobic Digestion of Poultry Manure Project
Alberta Agriculture and Forestry
Trial 4 pH Control 6+/- 0.3 Steady State 6.3 pH Units 17 Nov. to 14 Dec. 2015
Dissolved Potassium (K)
Dissolved Sodium (Na)
Hardness (CaCO3)
Dissolved Sulphate (SO4)
Total Dissolved Solids
Alkalinity (Total as CaCO3)
Runtime Hours
Runtime Days
Hardness (CaCO3) mg/L
Potassium mg/L Sodium mg/L Sulphur mg/L
Total Dissolved Solids mg / mL
Alkalinity Total as CACO3
Suspect Sampling, Analytical Error
Maxxam Result (1) Sample 7
\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015\Graph's and Data\Trial 4 Nov Dec 2015\
Trial 4 - 4 Jan 2016 Maxxam B5B1023V2-R2015-12-31_12-19-18_N001.xlsChart 1



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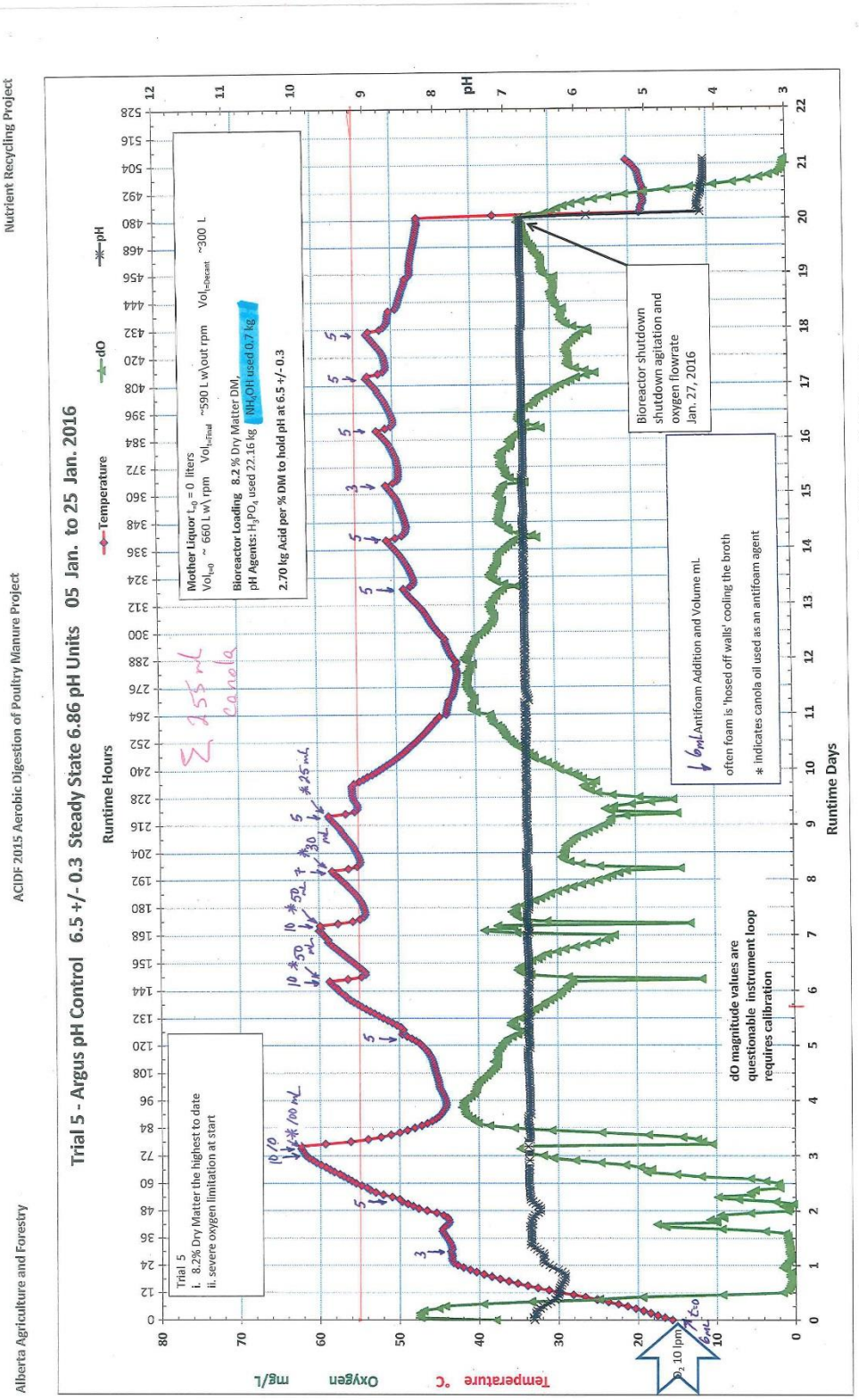
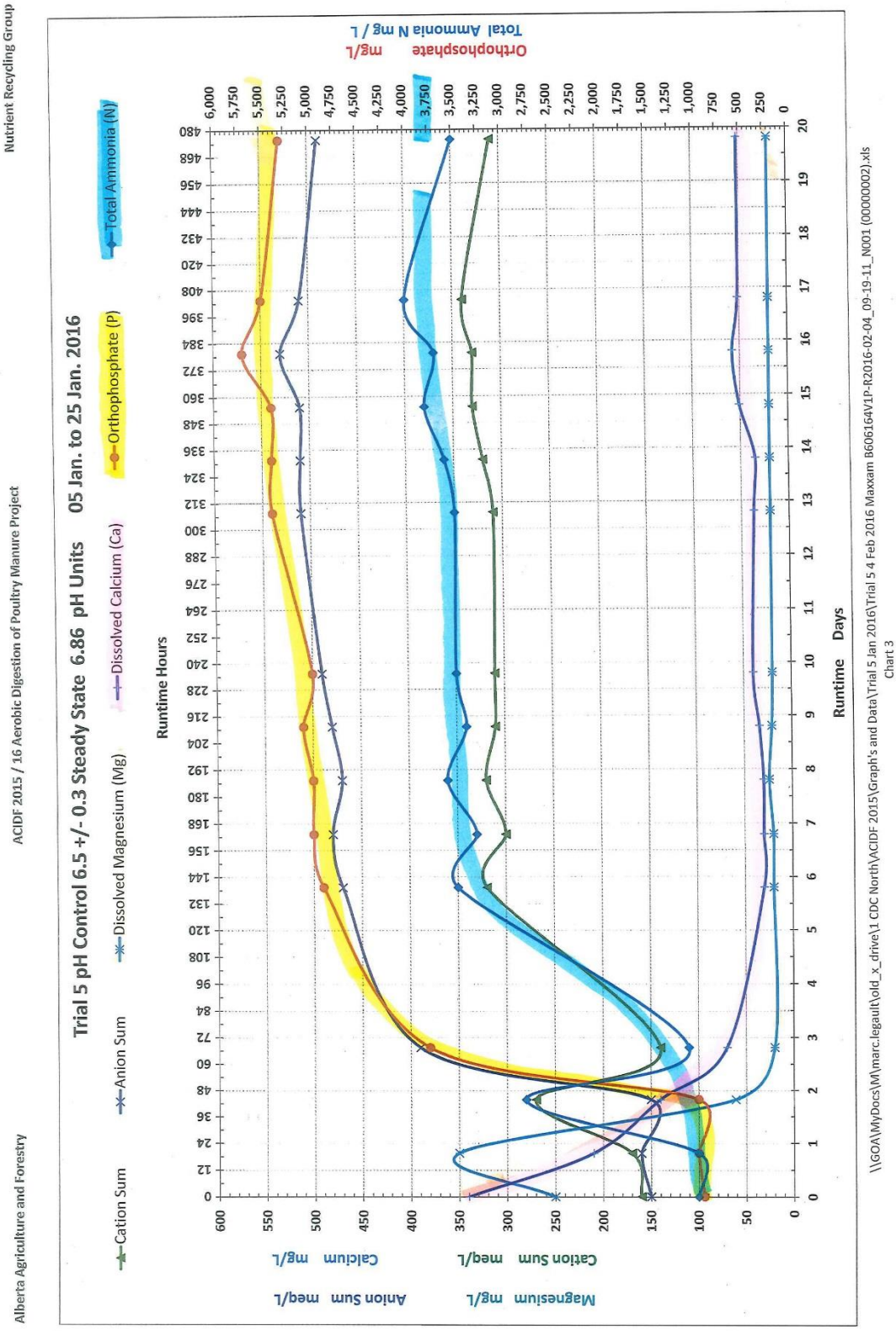
Trial 4 - 4 Jan 2016 Maxxam B5B1023V2-R2015-12-31_12-19-18_N001.xlsChart 4

Trial 5


Nutrient Recycling Project
ACIDF 2015 Aerobic Digestion of Poultry Manure Project
Alberta Agriculture and Forestry
Trial 5 - Argus pH Control 6.5 +/- 0.3 Steady State 6.86 pH Units 05 Jan. to 25 Jan. 2016
Runtime Hours
Runtime Days
Temperature
dO
pH
Trial 5
i. 8.2% Dry Matter the highest to date
ii. severe oxygen limitation at start
Mother Liquor $t_{=0}$ = 0 liters
$Vol_{t=0}$ ~ 660 L w\ rpm $Vol_{t=final}$ ~590 L w\out rpm $Vol_{t=decant}$ ~300 L
Bioreactor Loading 8.2 % Dry Matter DM,
pH Agents: H_3PO_4 used 22.16 kg NH_4OH used 0.7 kg
2.70 kg Acid per % DM to hold pH at 6.5 +/- 0.3
Bioreactor shutdown
shutdown agitation and
oxygen flowrate
Jan. 27, 2016
Antifoam Addition and Volume mL
often foam is 'hosed off walls' cooling the broth
* indicates canola oil used as an antifoam agent
dO magnitude values are
questionable instrument loop
requires calibration
Oxygen mg/L
Temperature °C
pH
\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015\Graph's and Data\Trial 5 Jan 2016\Graph Trial 5 Argus pH, dO, Temp..xlsx
Graph Trial 5 pH, dO, Temp


Nutrient Recycling Group
ACIDF 2015 / 16 Aerobic Digestion of Poultry Manure Project
Alberta Agriculture and Forestry
Trial 5 pH Control 6.5 +/- 0.3 Steady State 6.86 pH Units 05 Jan. to 25 Jan. 2016
Cation Sum
Anion Sum
Dissolved Magnesium (Mg)
Dissolved Calcium (Ca)
Orthophosphate (P)
Total Ammonia (N)
Runtime Hours
Runtime Days
Anion Sum meq/L
Calcium mg/L
Cation Sum meq/L
Magnesium mg/L
Orthophosphate mg/L
Total Ammonia N mg / L
\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015\Graph's and Data\Trial 5 Jan 2016\Trial 5 4 Feb 2016 Maxxam B606164V1P-R2016-02-04_09-19-11_N001 (00000002).xls
Chart 3

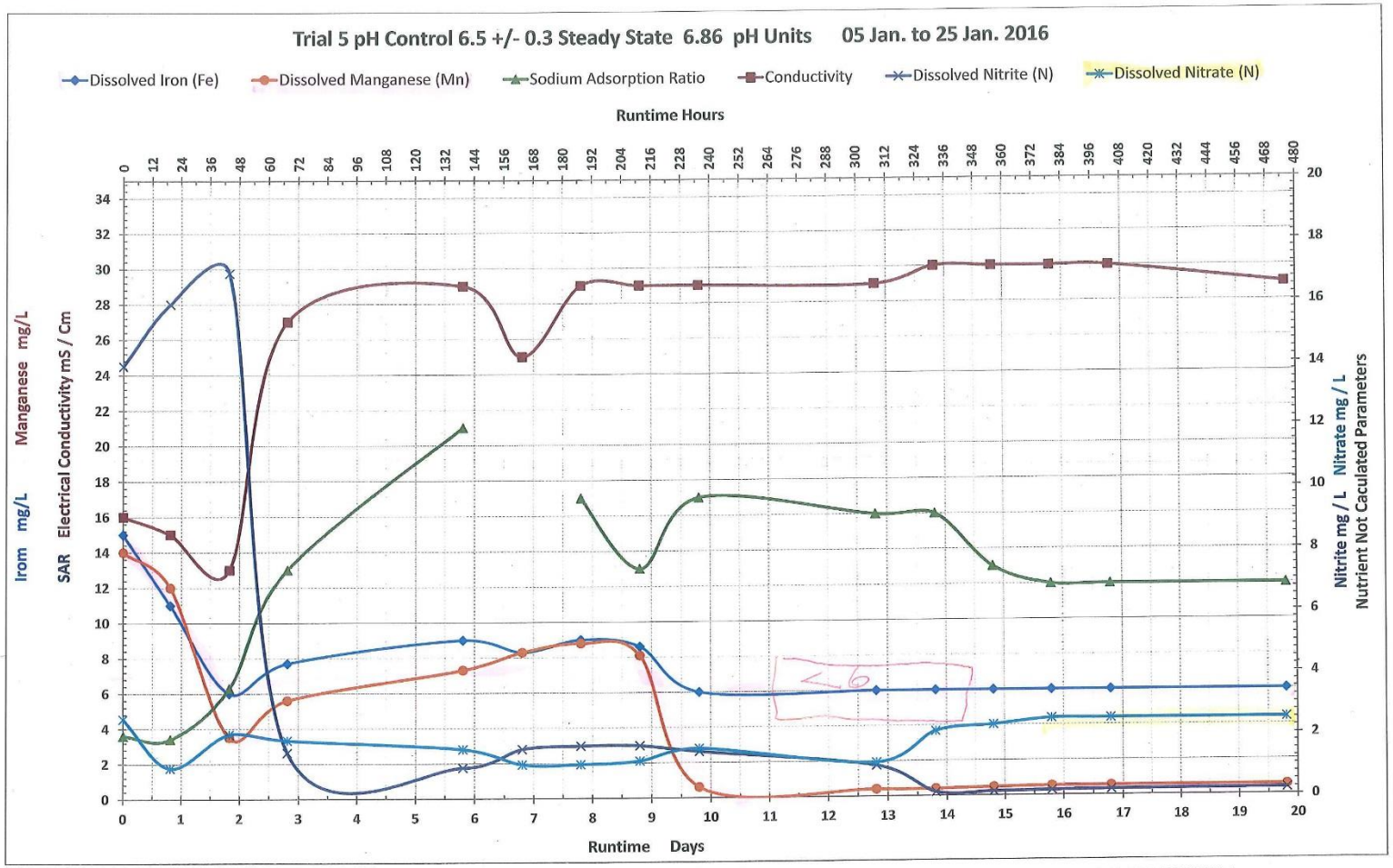


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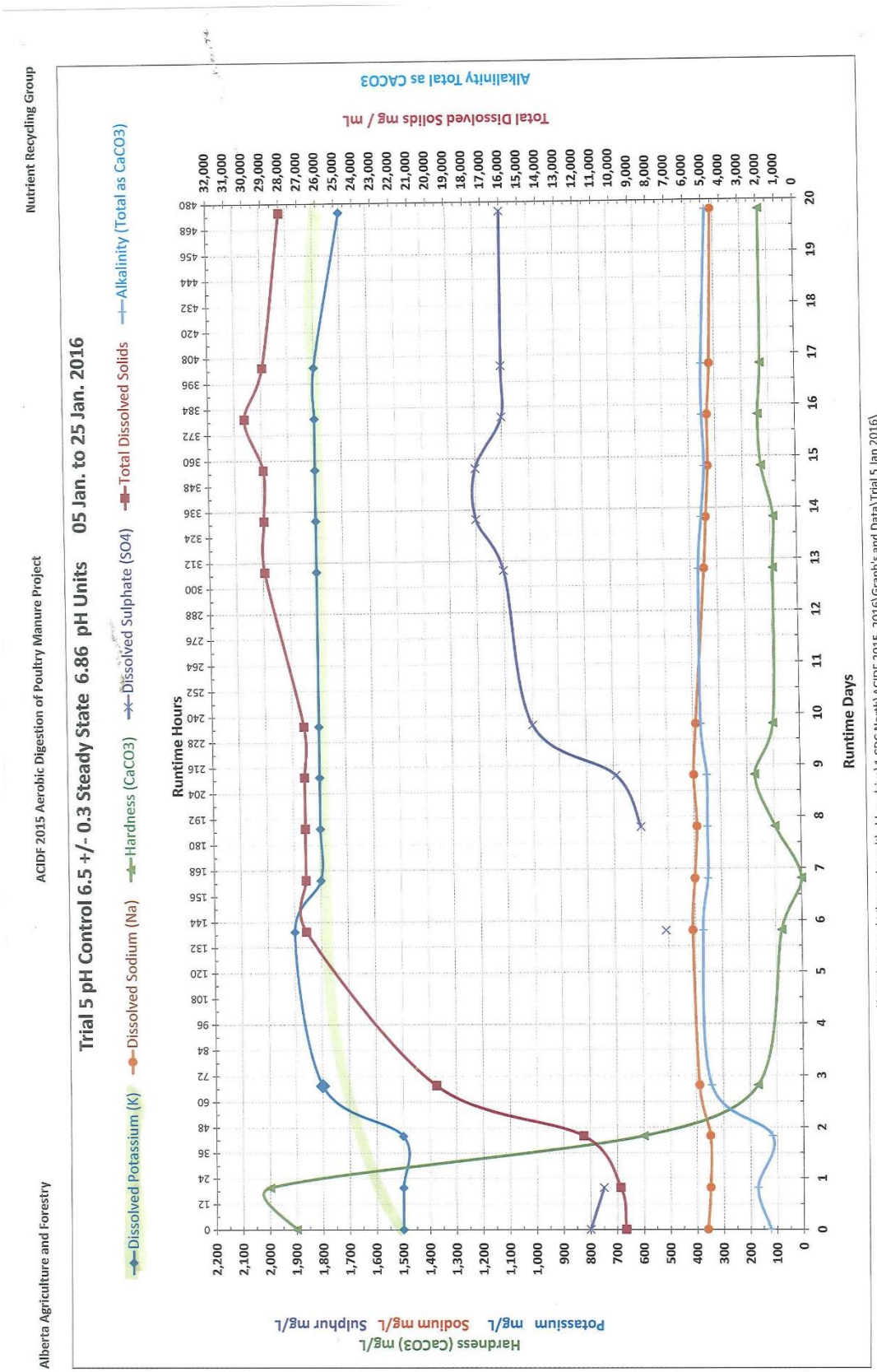
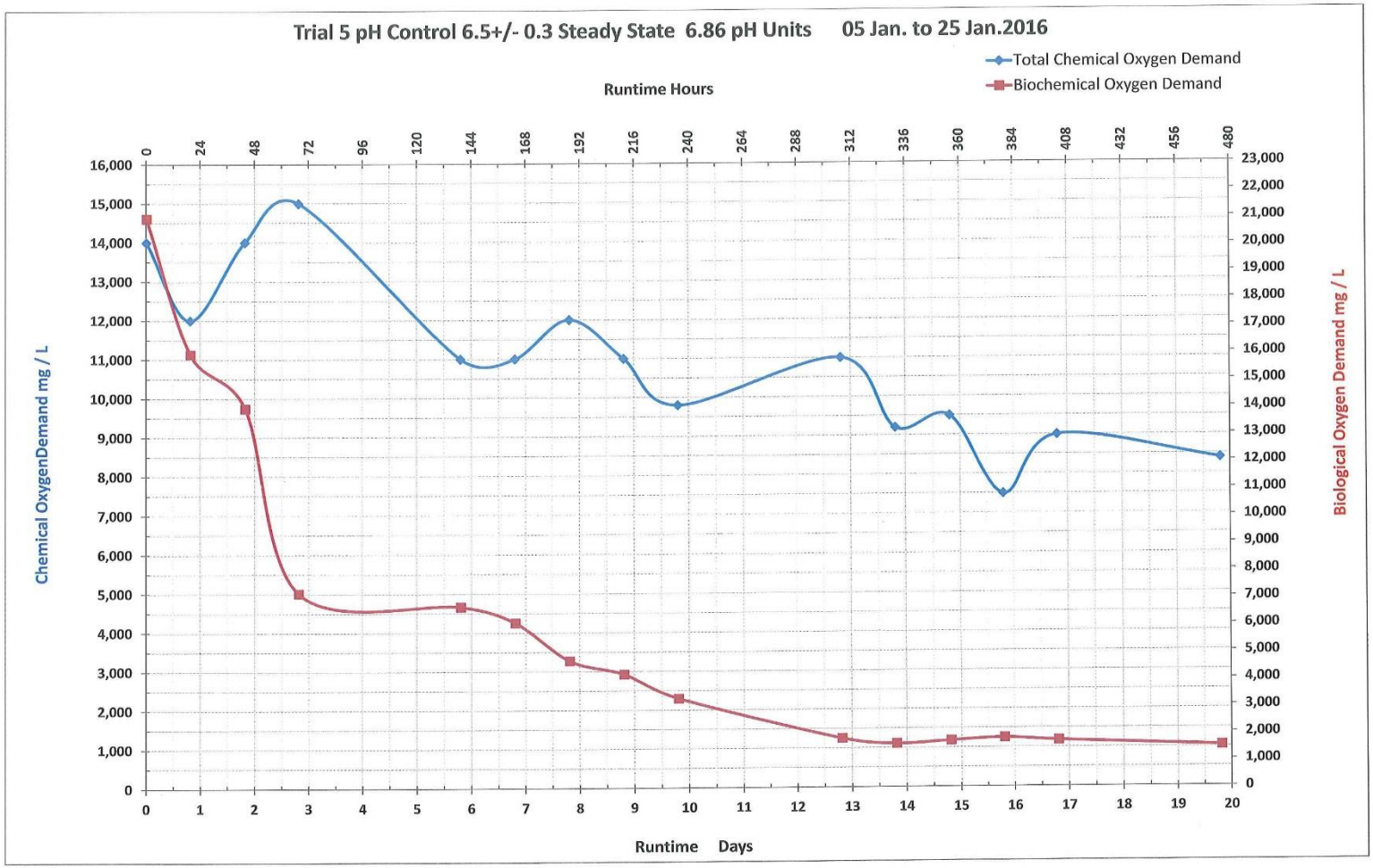
Chart 2


Alberta Agriculture and Forestry
ACIDF 2015 Aerobic Digestion of Poultry Manure Project
Nutrient Recycling Group
Trial 5 pH Control 6.5 +/- 0.3 Steady State 6.86 pH Units 05 Jan. to 25 Jan. 2016
Dissolved Potassium (K)
Dissolved Sodium (Na)
Hardness (CaCO3)
Dissolved Sulphate (SO4)
Total Dissolved Solids
Alkalinity (Total as CaCO3)
Runtime Hours
Runtime Days
Hardness (CaCO3) mg/L
Potassium mg/L Sodium mg/L Sulphur mg/L
Total Dissolved Solids mg / mL
Alkalinity Total as CACO3
\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015, 2016\Graph's and Data\Trial 5 Jan 2016\
Trial 5 4 Feb 2016 Maxxam B606164V1P-R2016-02-04_09-19-11_N001 (00000002).xlsChart 1



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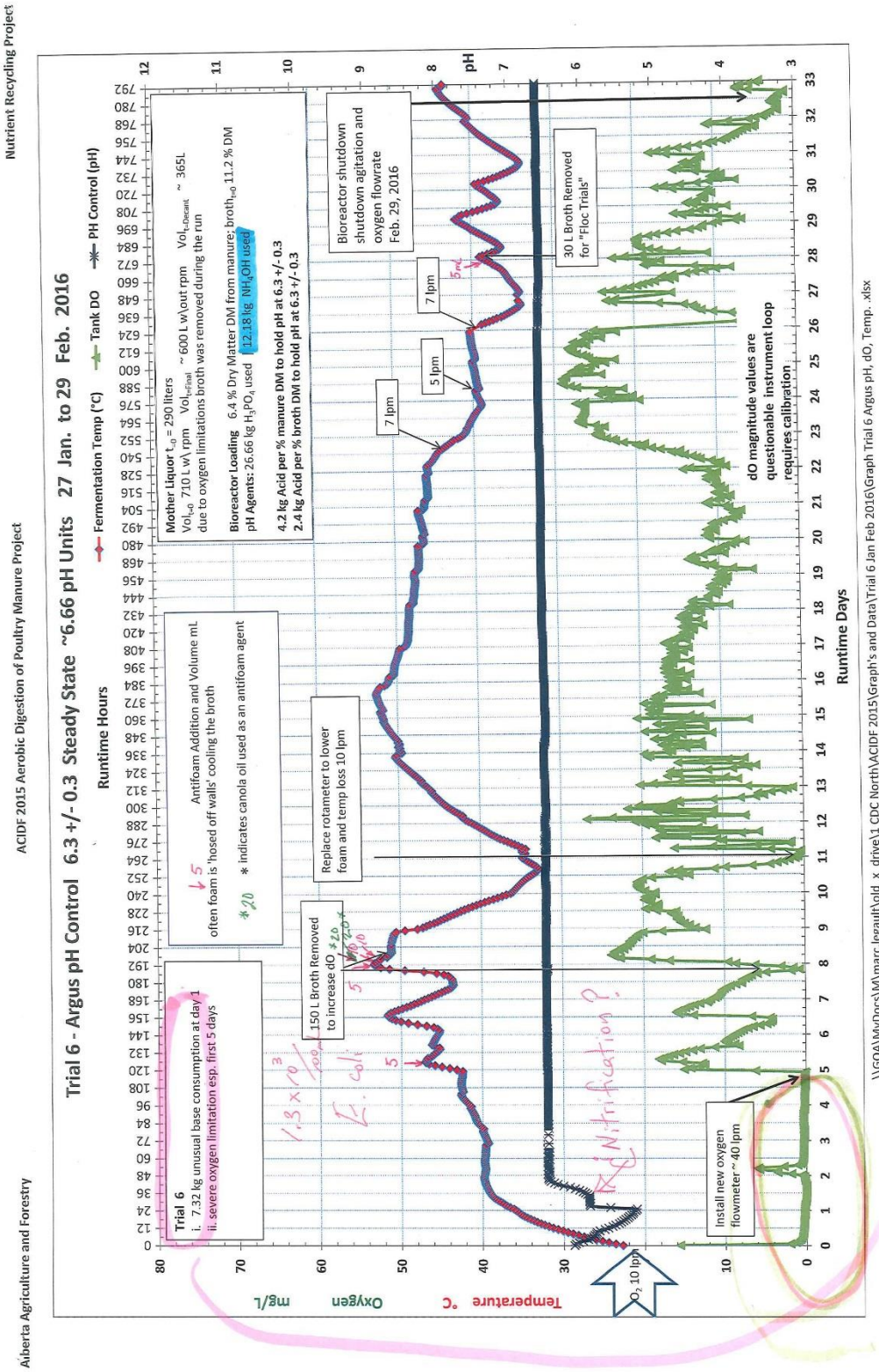
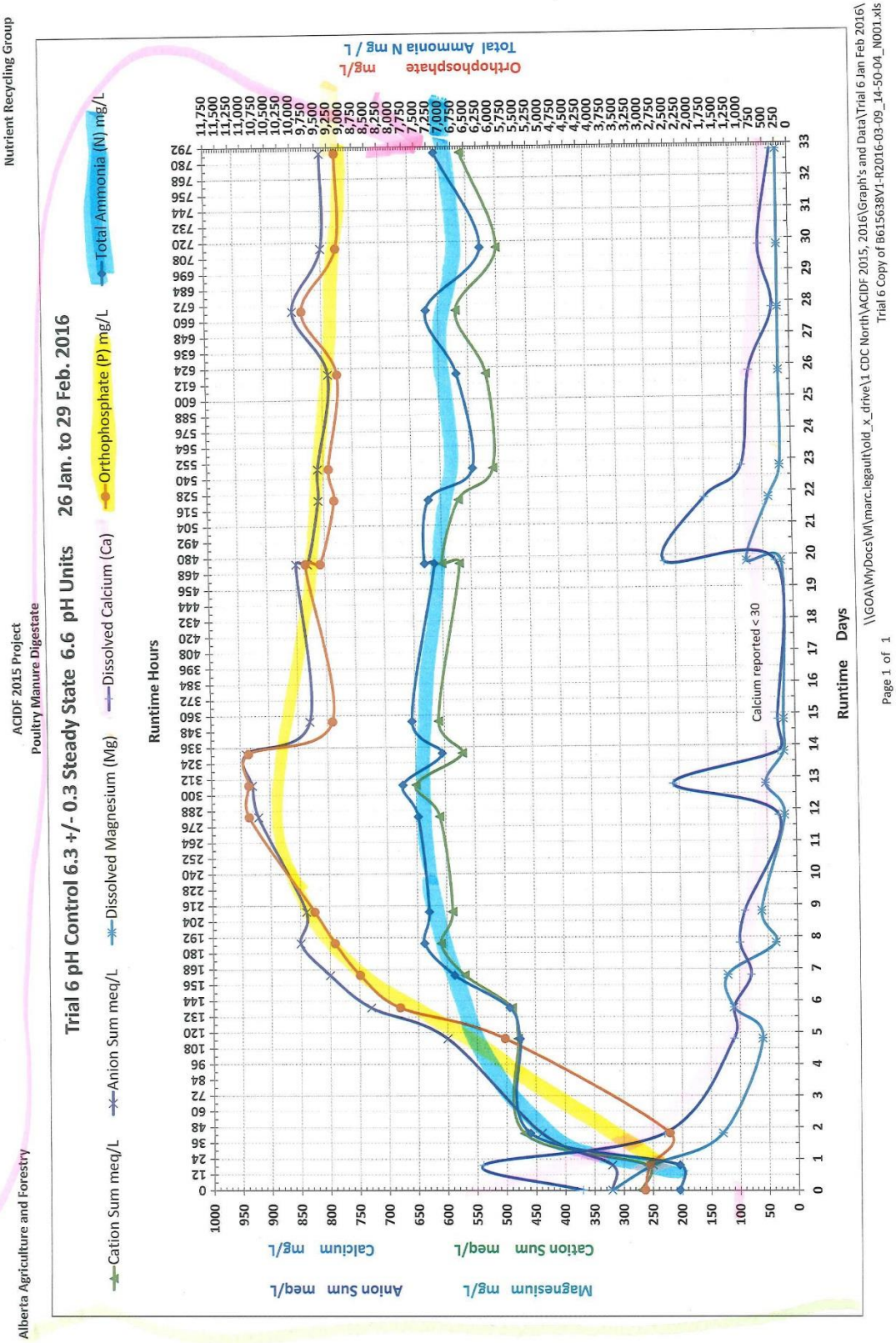
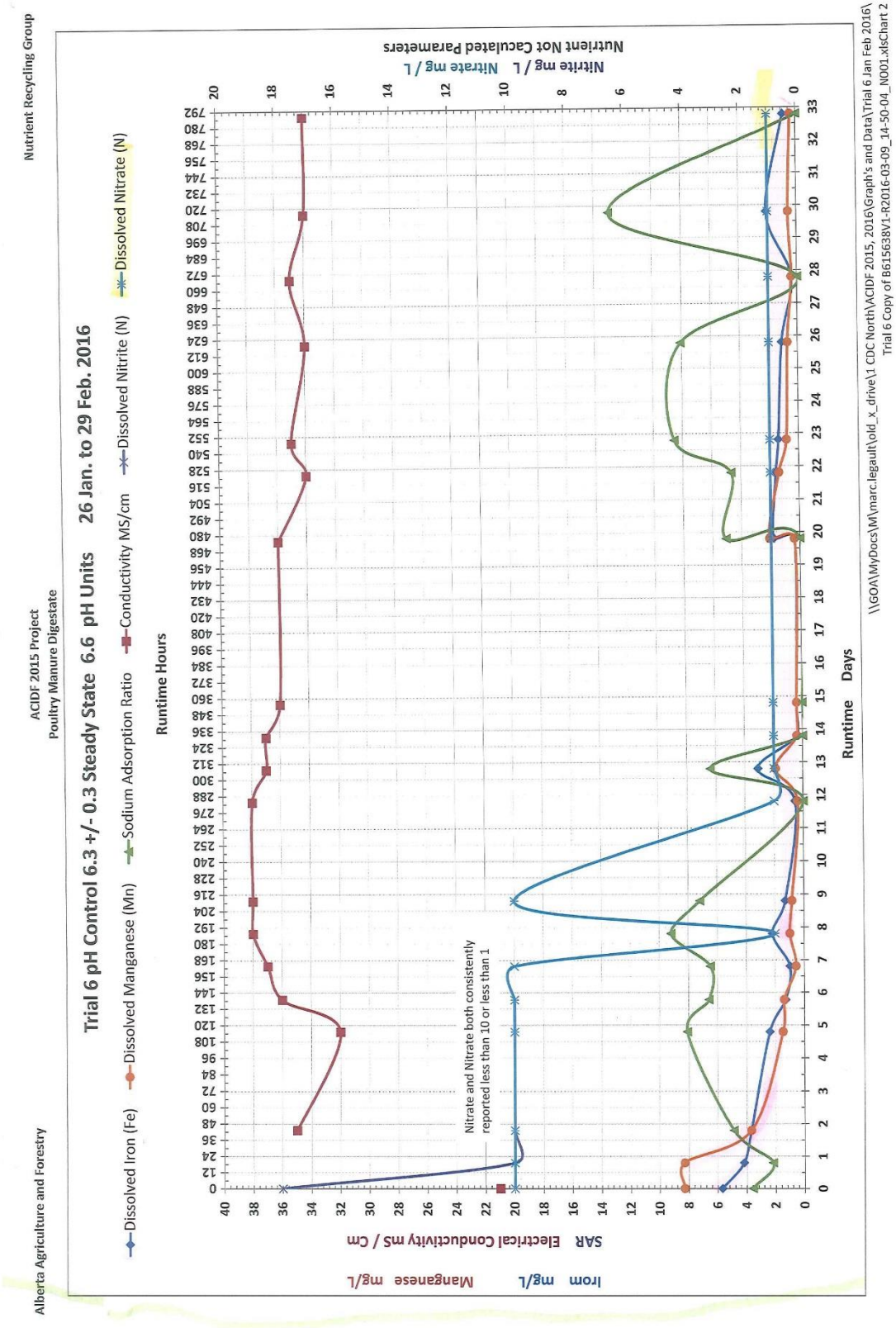
BOD f B606164V3-R2016-02-10_17-29-48_N001.xls BOD Graph


Nutrient Recycling Project
ACIDF 2015 Aerobic Digestion of Poultry Manure Project
Alberta Agriculture and Forestry
Trial 6 - Argus pH Control 6.3 +/- 0.3 Steady State ~6.66 pH Units 27 Jan. to 29 Feb. 2016
Fermentation Temp (°C)
Tank DO
PH Control (pH)
Runtime Hours
Runtime Days
Temperature °C
Oxygen mg/L
pH
Trial 6
i. 7.32 kg unusual base consumption at day 1
ii. severe oxygen limitation esp. first 5 days
150 L Broth Removed to increase dO
Antifoam Addition and Volume mL
often foam is 'hosed off walls' cooling the broth
* indicates canola oil used as an antifoam agent
Replace rotameter to lower foam and temp loss 10 lpm
Mother Liquor $t_{i,0}$ = 290 liters
$Vol_{t=0}$ 710 L w\ rpm $Vol_{t=Final}$ ~ 600 L w\out rpm $Vol_{t=Decant}$ ~ 365L
due to oxygen limitations broth was removed during the run
Bioreactor Loading 6.4 % Dry Matter DM from manure; $broth_{t=0}$ 11.2 % DM
pH Agents: 26.66 kg H_3PO_4 used 12.18 kg NH_4OH used
4.2 kg Acid per % manure DM to hold pH at 6.3 +/- 0.3
2.4 kg Acid per % broth DM to hold pH at 6.3 +/- 0.3
7 lpm
5 lpm
7 lpm
5 mL
Bioreactor shutdown shutdown agitation and oxygen flowrate Feb. 29, 2016
30 L Broth Removed for "Floc Trials"
dO magnitude values are questionable instrument loop requires calibration
Install new oxygen flowmeter ~ 40 lpm
O_2 10 lpm
\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015\Graph's and Data\Trial 6 Jan Feb 2016\Graph Trial 6 Argus pH, dO, Temp. .xlsx
Graph Trial 6 pH, dO, Temp

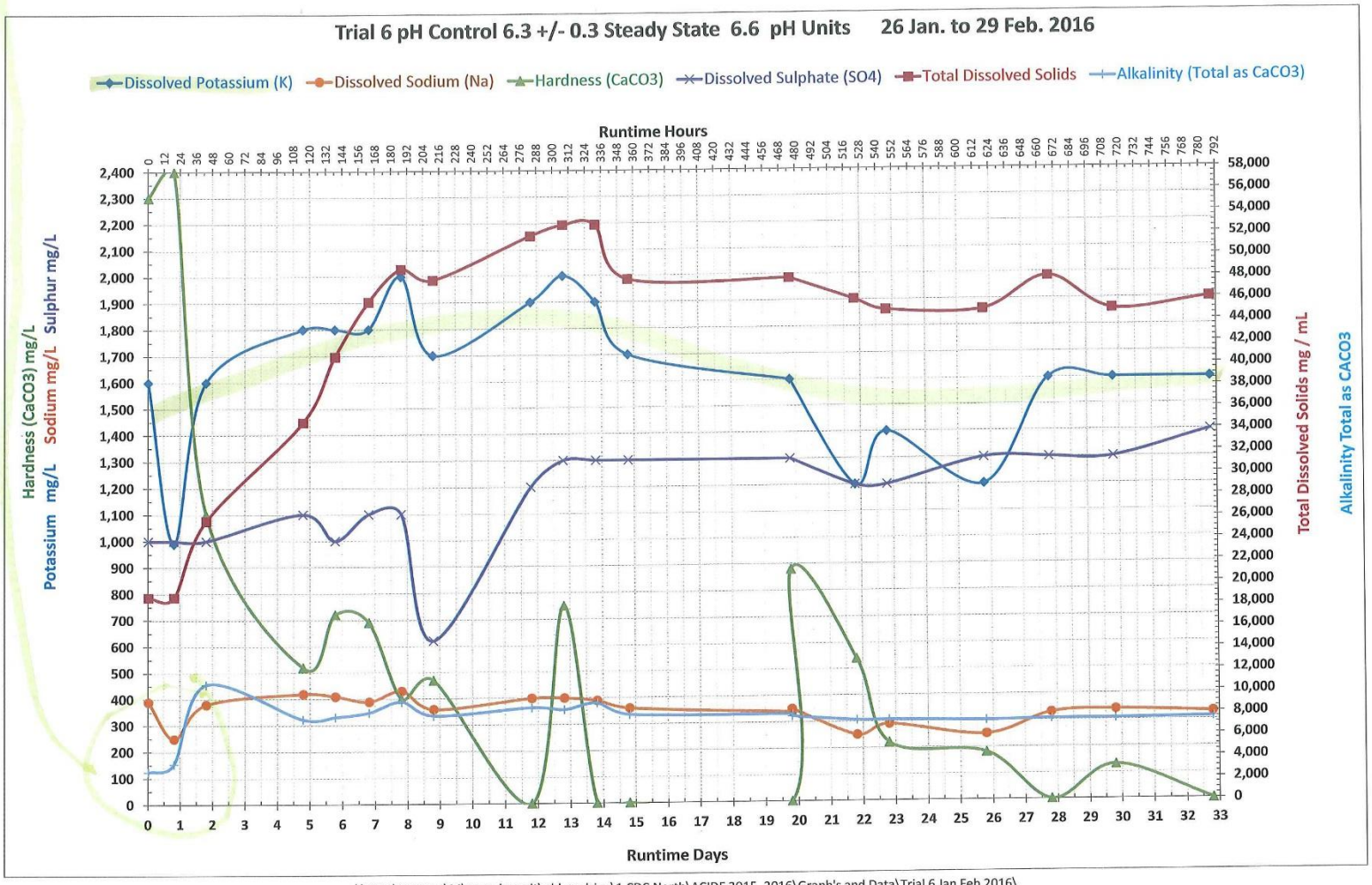

Nutrient Recycling Group
ACIDF 2015 Project
Poultry Manure Digestate
Alberta Agriculture and Forestry
Trial 6 pH Control 6.3 +/- 0.3 Steady State 6.6 pH Units 26 Jan. to 29 Feb. 2016
Cation Sum meq/L
Anion Sum meq/L
Dissolved Magnesium (Mg)
Dissolved Calcium (Ca)
Orthophosphate (P) mg/L
Total Ammonia (N) mg/L
Runtime Hours
Runtime Days
Anion Sum meq/L
Calcium mg/L
Cation Sum meq/L
Magnesium mg/L
Orthophosphate mg/L
Total Ammonia N mg / L
Calcium reported < 30
\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015, 2016\Graph's and Data\Trial 6 Jan Feb 2016\
Trial 6 Copy of B615638V1-R2016-03-09_14-50-04_N001.xls
Page 1 of 1


Nutrient Recycling Group
ACIDF 2015 Project
Poultry Manure Digestate
Alberta Agriculture and Forestry
Trial 6 pH Control 6.3 +/- 0.3 Steady State 6.6 pH Units 26 Jan. to 29 Feb. 2016
Dissolved Iron (Fe)
Dissolved Manganese (Mn)
Sodium Adsorption Ratio
Conductivity MS/cm
Dissolved Nitrite (N)
Dissolved Nitrate (N)
Runtime Hours
Nitrate and Nitrate both consistently reported less than 10 or less than 1
Nitrite mg / L Nitrate mg / L
Nutrient Not Cacluated Parameters
Iron mg/L
Manganese mg/L
SAR Electrical Conductivity mS / Cm
Runtime Days
\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015, 2016\Graph's and Data\Trial 6 Jan Feb 2016\
Trial 6 Copy of B615638V1-R2016-03-09_14-50-04_N001.xlsChart 2

Alberta Agriculture and Forestry

ACIDF 2015 Aerobic Digestion of Poultry Manure Project

Nutrient Recycling Group



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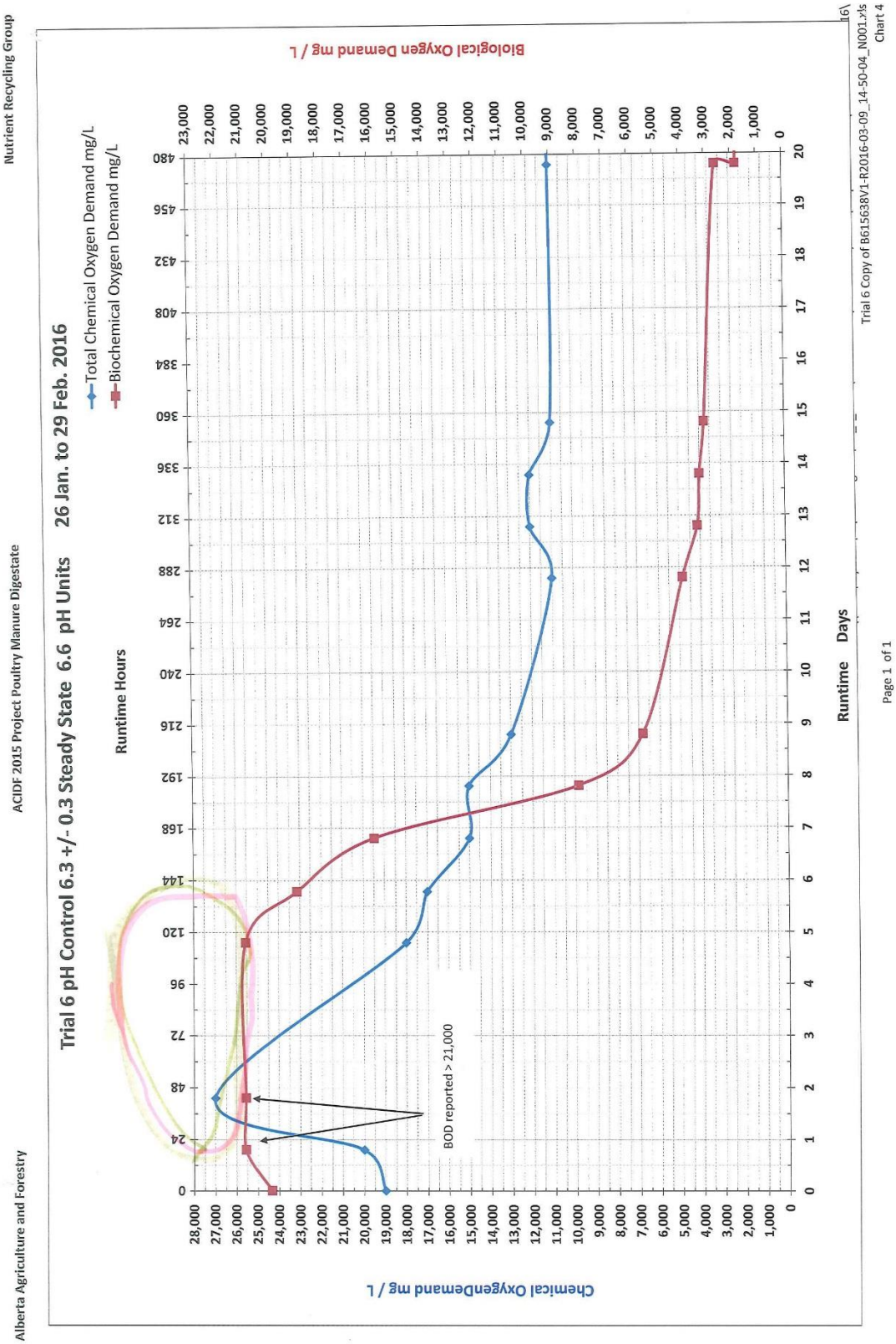
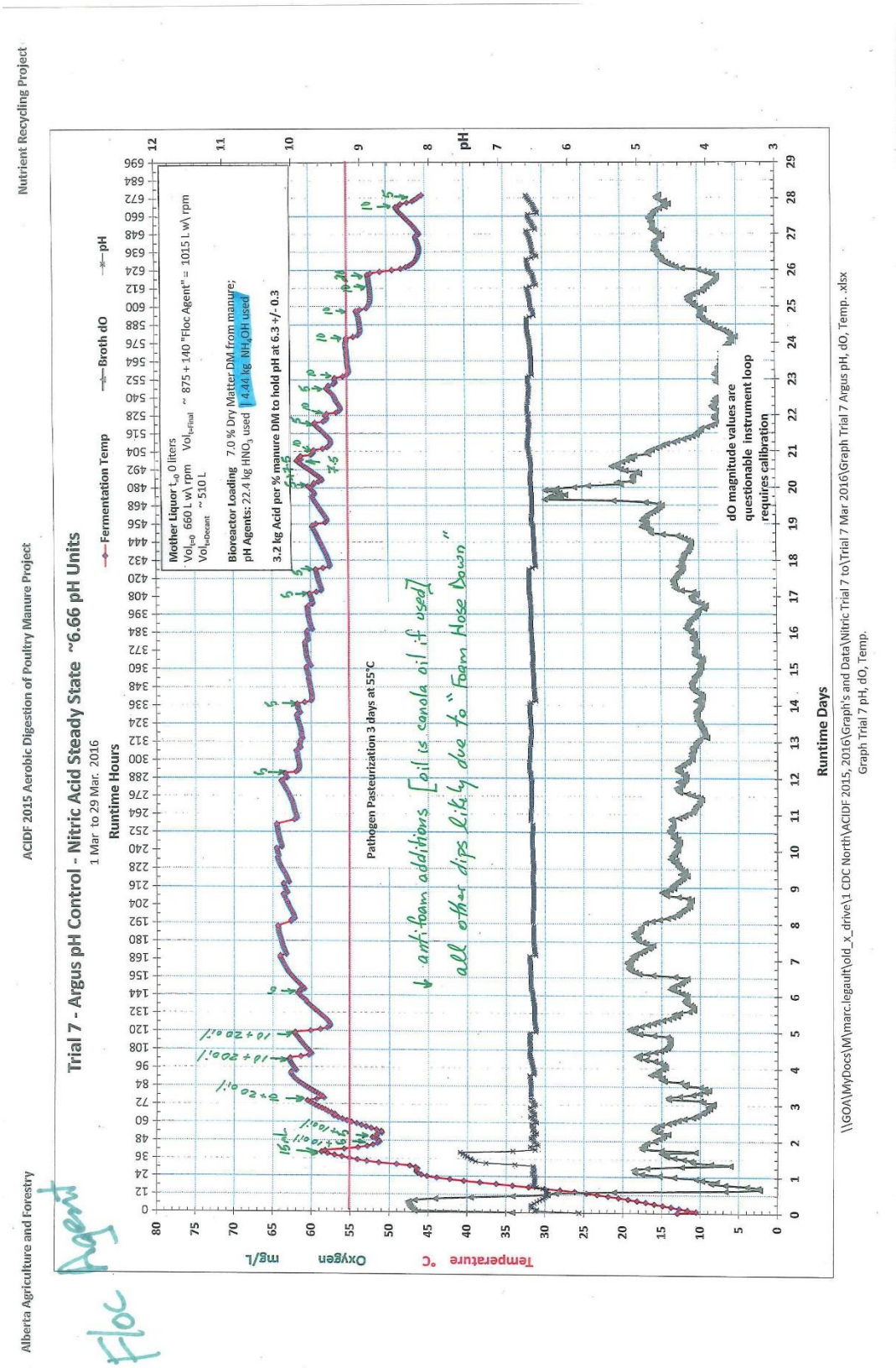
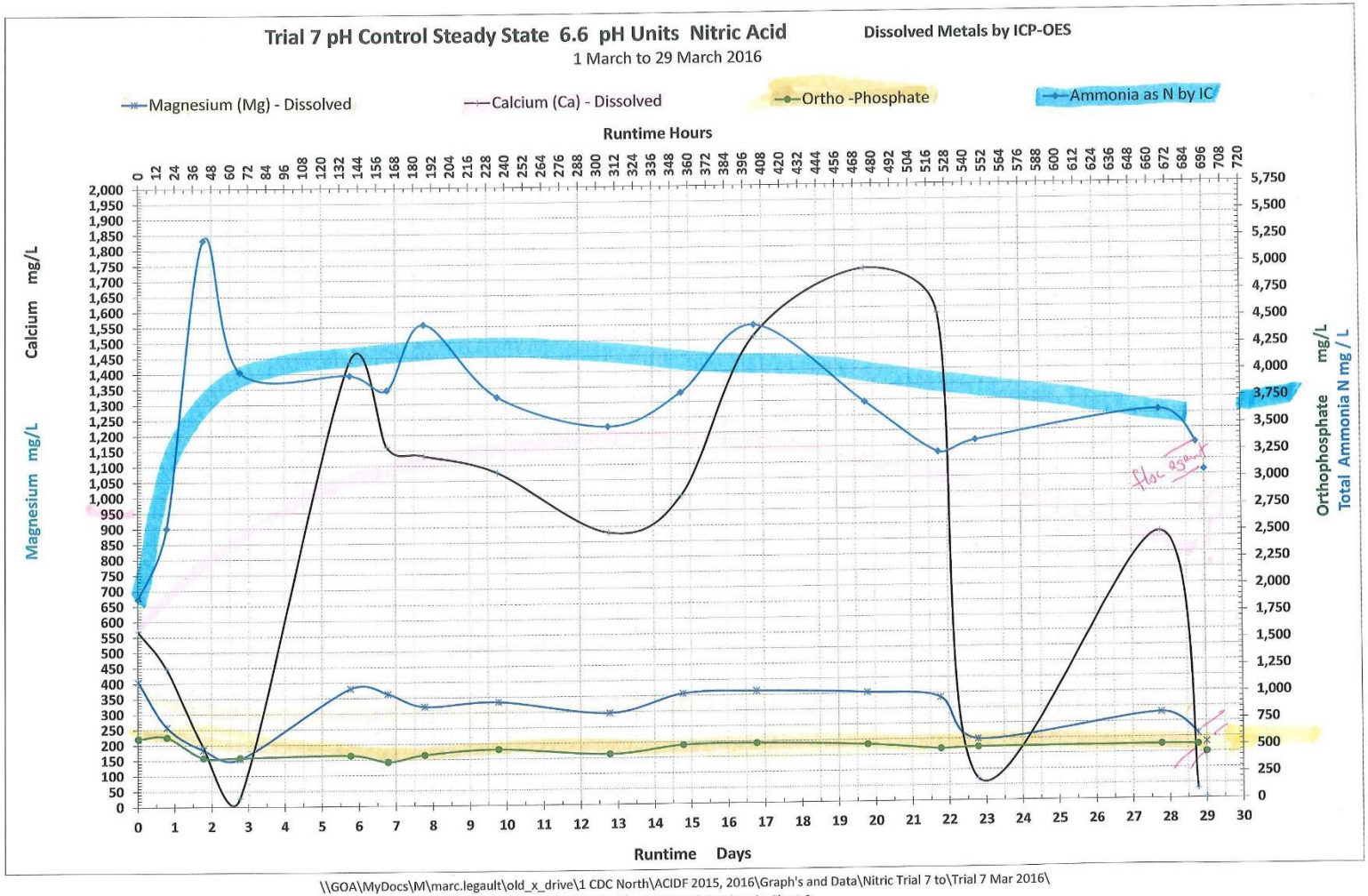
Trial 6 Copy of B615638V1-R2016-03-09_14-50-04_N001.xlsChart 1

Nitrification eats up alkalinity


Nutrient Recycling Group
ACIDF 2015 Project Poultry Manure Digestate
Alberta Agriculture and Forestry
Trial 6 pH Control 6.3 +/- 0.3 Steady State 6.6 pH Units 26 Jan. to 29 Feb. 2016
Total Chemical Oxygen Demand mg/L
Biochemical Oxygen Demand mg/L
Runtime Hours
Runtime Days
Chemical OxygenDemand mg / L
Biological Oxygen Demand mg / L
BOD reported > 21,000
Trial 6 Copy of B615638V1-R2016-03-09_14-50-04_N001.xls
Chart 4
Page 1 of 1


Alberta Agriculture and Forestry
ACIDF 2015 Aerobic Digestion of Poultry Manure Project
Nutrient Recycling Project
Trial 7 - Argus pH Control - Nitric Acid Steady State ~6.66 pH Units
1 Mar to 29 Mar. 2016
Runtime Hours
Fermentation Temp
Broth dO
pH
Mother Liquor L=0 0 liters
Vol L=0 660 L w\ rpm Vol L=final ~ 875 + 140 "Floc Agent" = 1015 L w\ rpm
Vol L=Decant ~ 510 L
Bioreactor Loading 7.0 % Dry Matter DM from manure;
pH Agents: 22.4 kg HNO3 used 14.44 kg NH4OH used
3.2 kg Acid per % manure DM to hold pH at 6.3 +/- 0.3
Pathogen Pasteurization 3 days at 55°C
dO magnitude values are questionable instrument loop requires calibration
Runtime Days
Temperature °C
Oxygen mg/L
pH
Agent
Floc
\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015, 2016\Graph's and Data\Nitric Trial 7 to\Trial 7 Mar 2016\Graph Trial 7 Argus pH, dO, Temp. .xlsx
Graph Trial 7 pH, dO, Temp.



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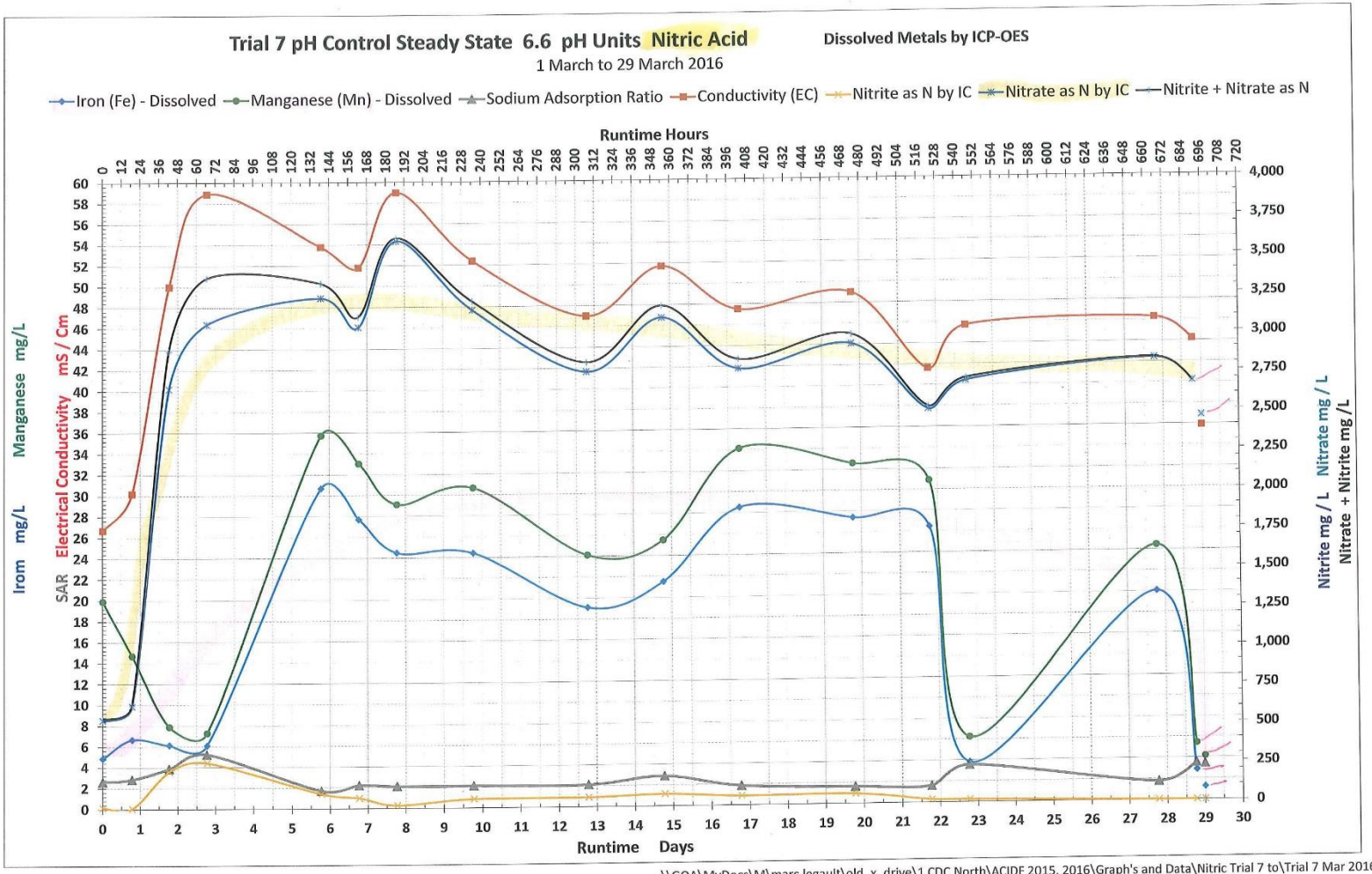
Trial 7 Nutrient Data and Graphs .xlsxChart 3



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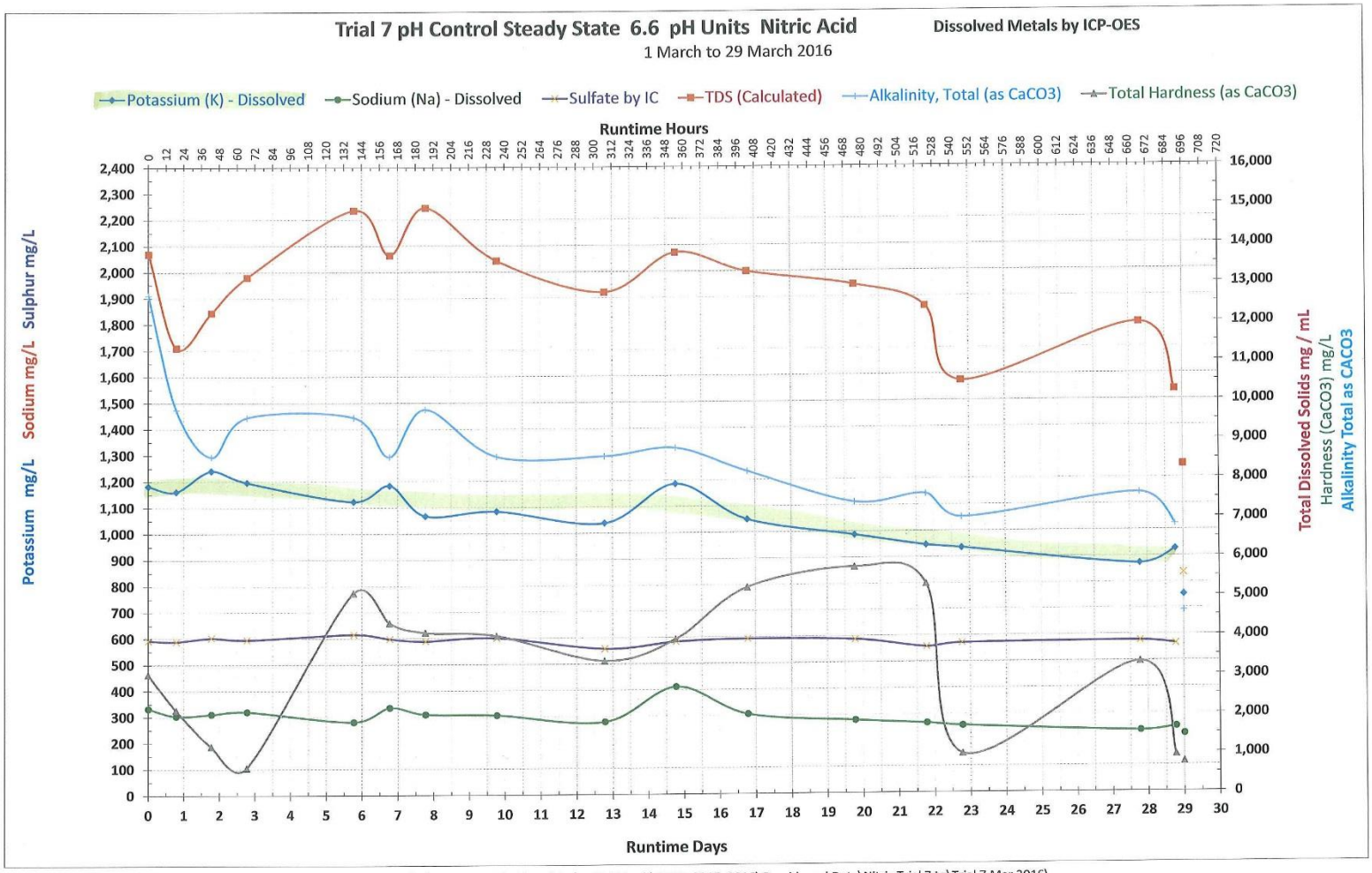
Trial 7 Nutrient Data and Graphs .xlsxChart 2



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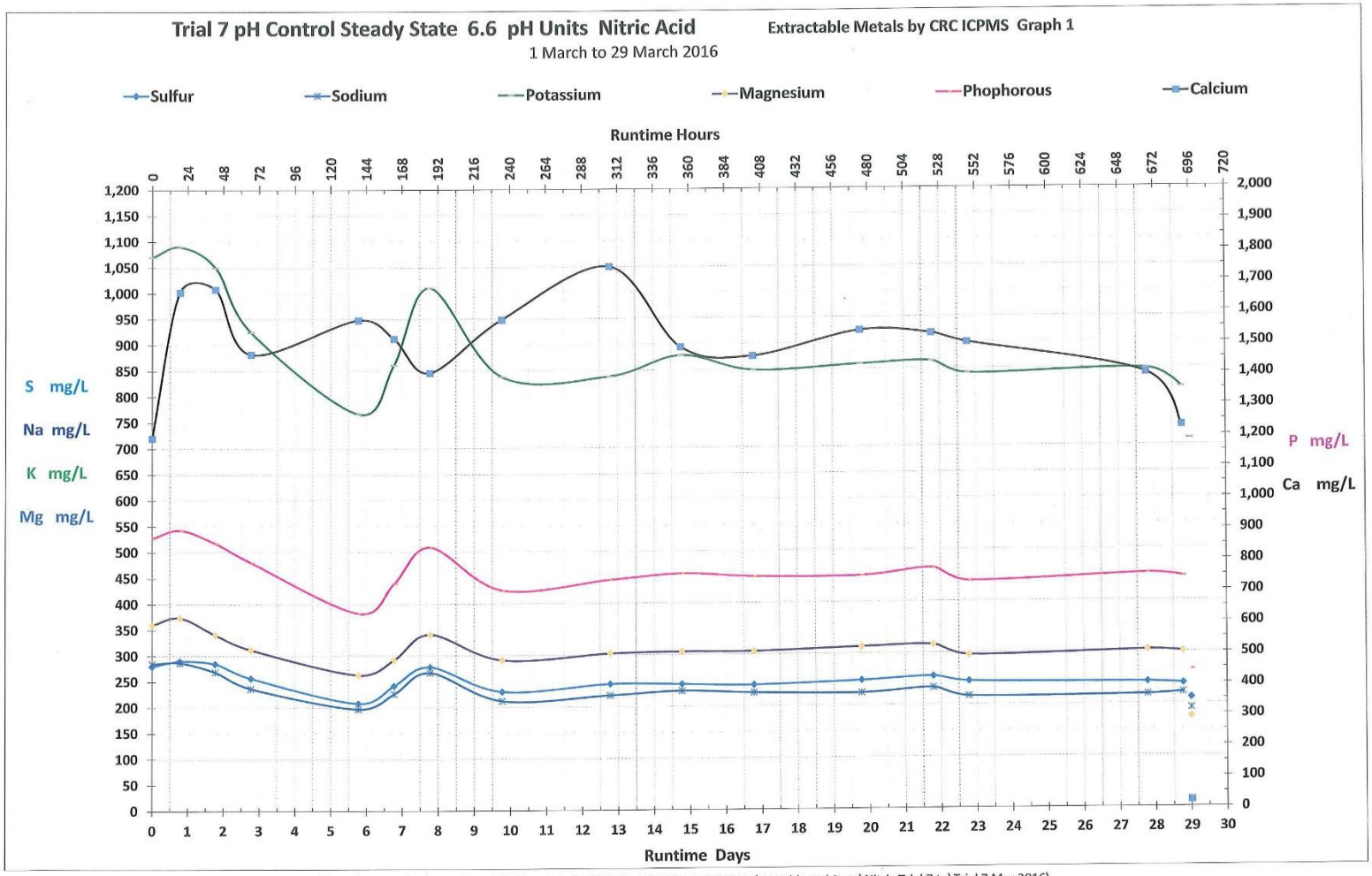
Trial 7 Nutrient Data and Graphs .xlsxChart 1



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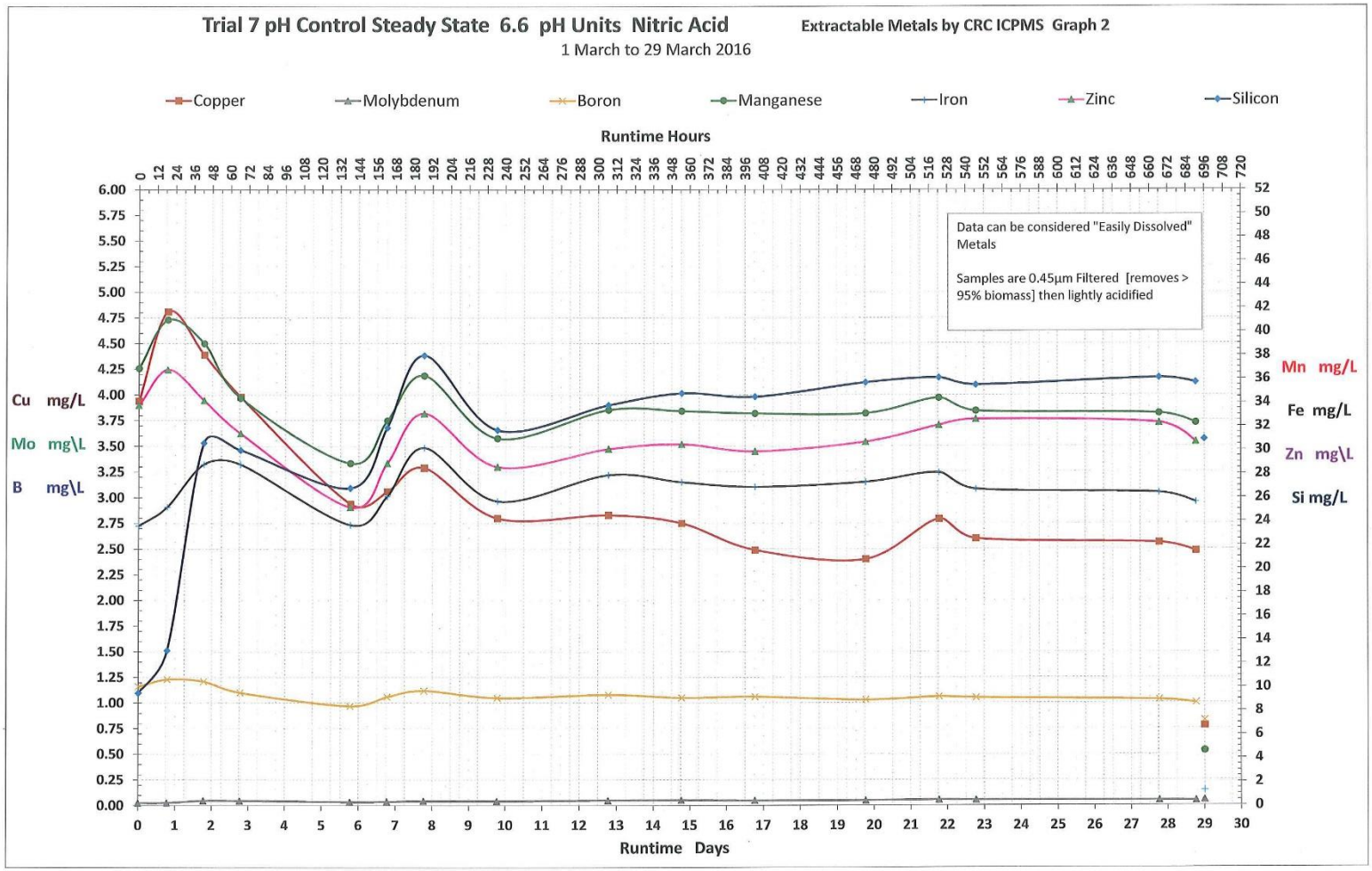
Trial 7 Nutrient Data and Graphs .xlsx Easily Dissolved Metals Graph 1



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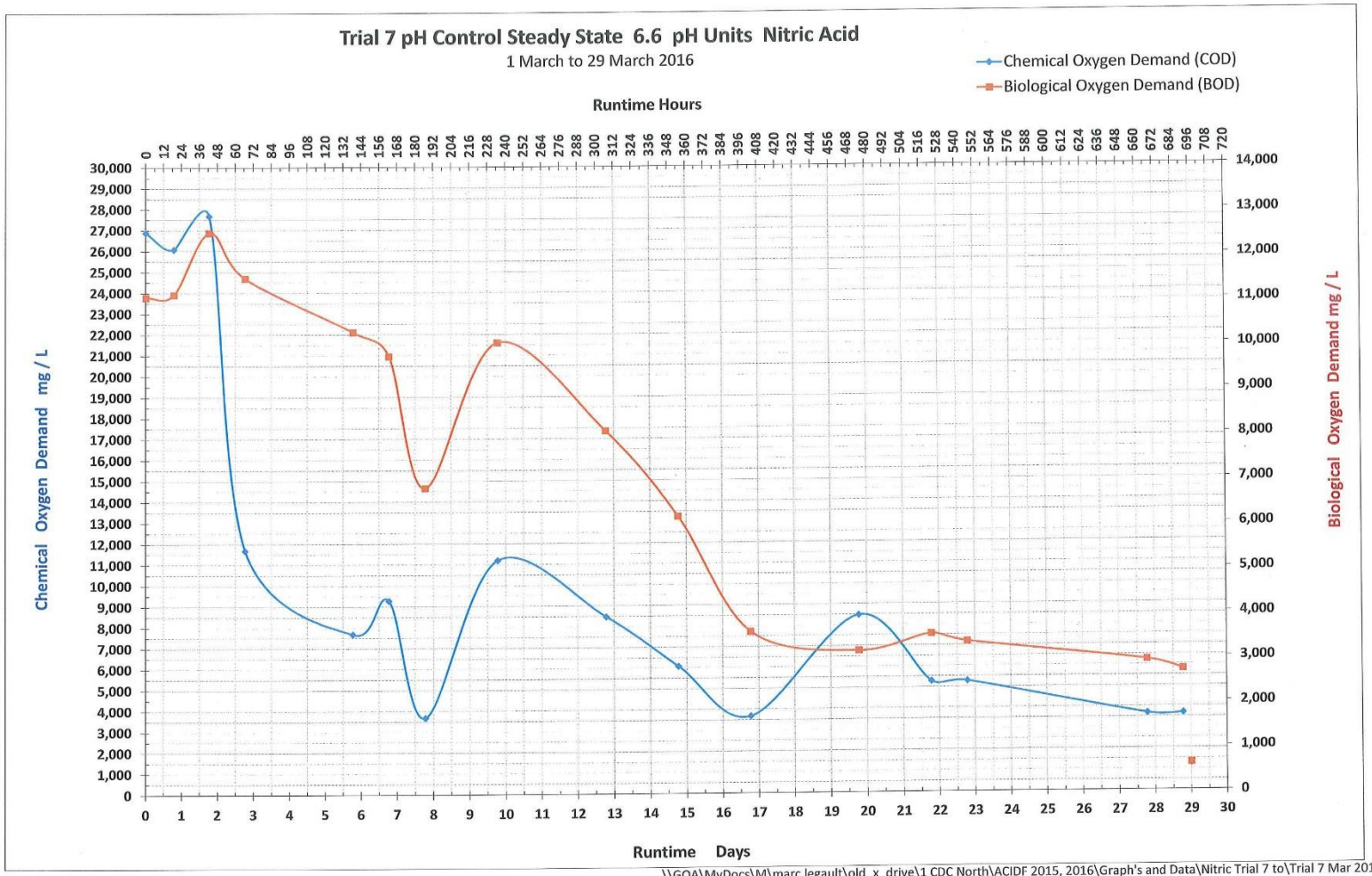

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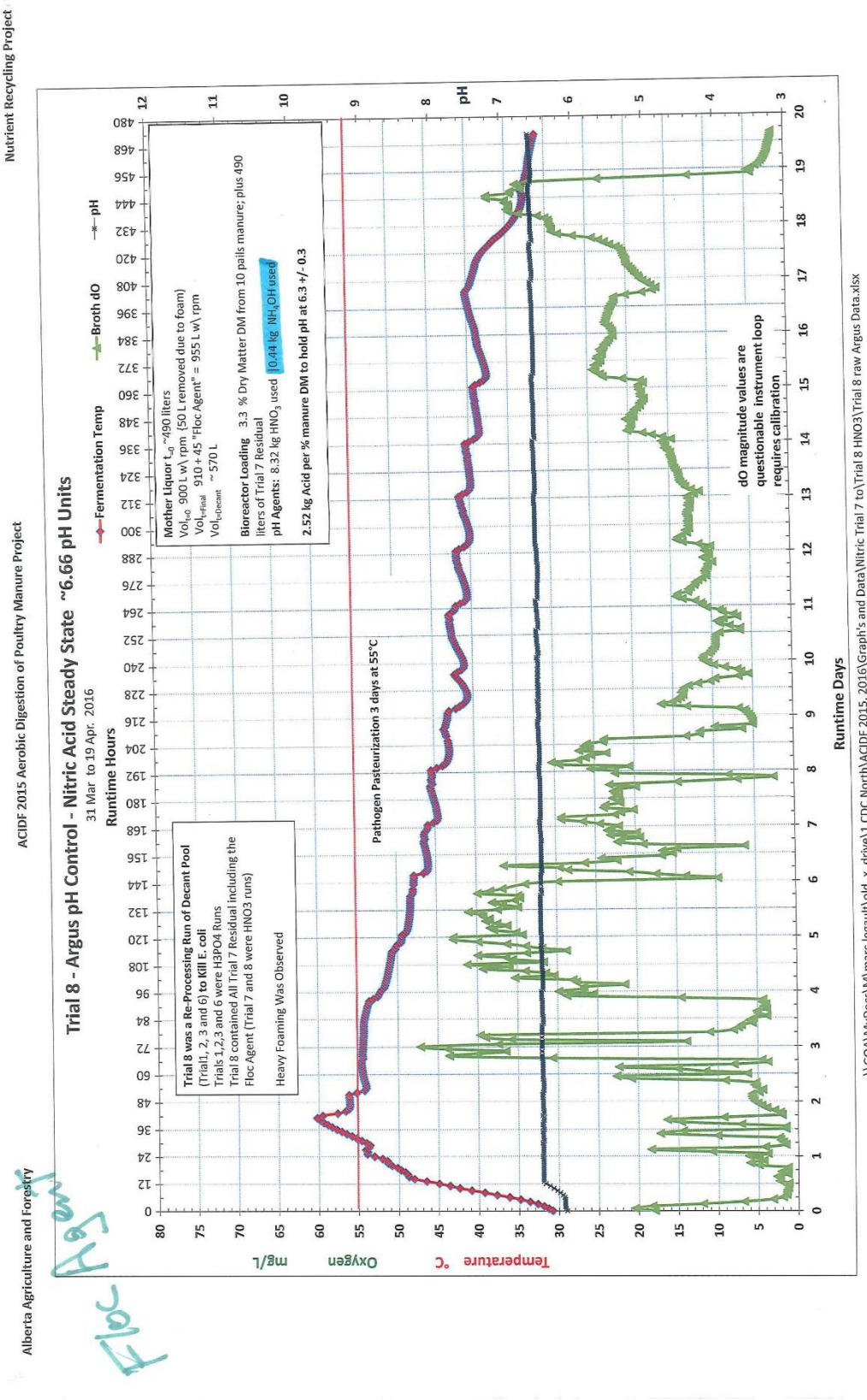
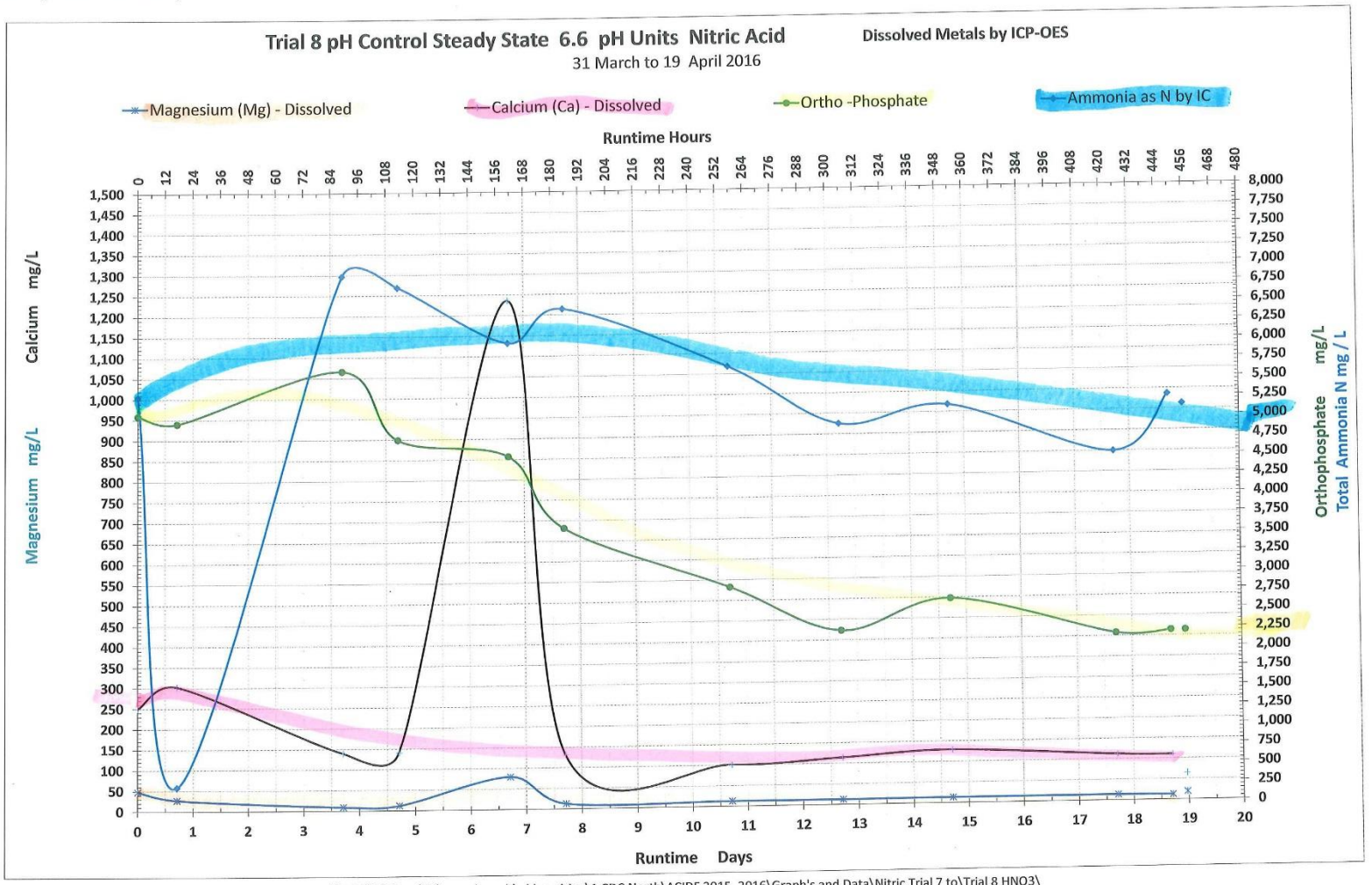
Trial 7 Nutrient Data and Graphs .xlsx
Chart 4


Nutrient Recycling Project
ACIDF 2015 Aerobic Digestion of Poultry Manure Project
Alberta Agriculture and Forestry
Trial 8 - Argus pH Control - Nitric Acid Steady State ~6.66 pH Units
31 Mar to 19 Apr. 2016
Runtime Hours
Fermentation Temp
Broth dO
pH
Trial 8 was a Re-Processing Run of Decant Pool (Trial1, 2, 3 and 6) to Kill E. coli
Trials 1,2,3 and 6 were H3PO4 Runs
Trial 8 contained All Trial 7 Residual including the Floc Agent (Trial 7 and 8 were HNO3 runs)
Heavy Foaming Was Observed
Mother Liquor t_0 ~490 liters
$Vol_{t=0}$ 900 L w\ rpm (50 L removed due to foam)
$Vol_{t=Final}$ 910 + 45 "Floc Agent" = 955 L w\ rpm
$Vol_{t=Decant}$ ~ 570 L
Bioreactor Loading 3.3 % Dry Matter DM from 10 pails manure; plus 490 liters of Trial 7 Residual
pH Agents: 8.32 kg HNO_3 used 0.44 kg NH_4OH used
2.52 kg Acid per % manure DM to hold pH at 6.3 +/- 0.3
Pathogen Pasteurization 3 days at 55°C
dO magnitude values are questionable instrument loop requires calibration
Temperature °C
Oxygen mg/L
pH
Runtime Days
\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015, 2016\Graph's and Data\Nitric Trial 7 to\Trial 8 HNO3\Trial 8 raw Argus Data.xlsx
Graph Trial 8 pH, dO, Temp



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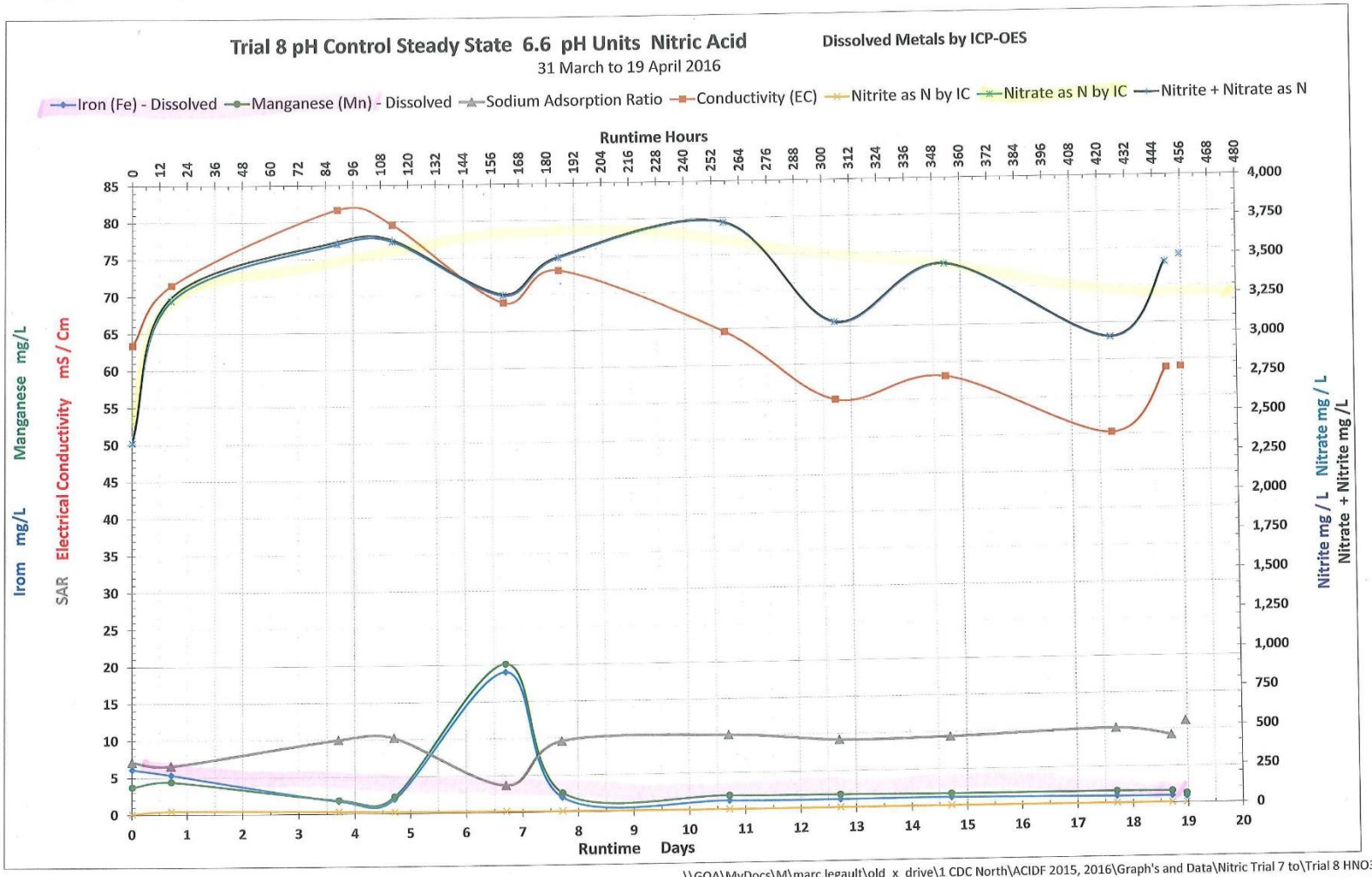
Trial 8 Nutrient Data and Graphs .xlsxChart 3

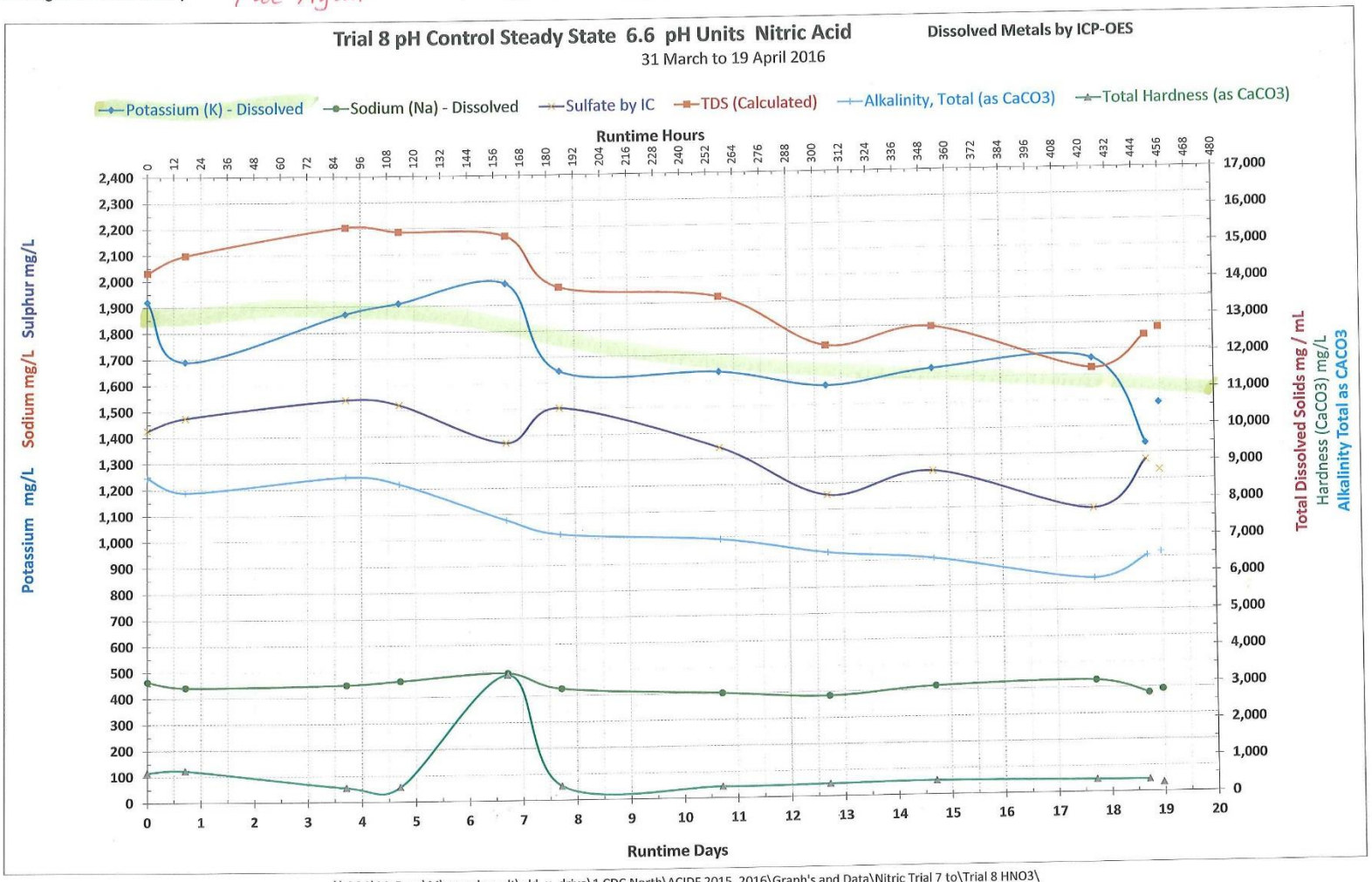

Trial 8 pH Control Steady State 6.6 pH Units Nitric Acid
31 March to 19 April 2016
Dissolved Metals by ICP-OES
Iron (Fe) - Dissolved
Manganese (Mn) - Dissolved
Sodium Adsorption Ratio
Conductivity (EC)
Nitrite as N by IC
Nitrate as N by IC
Nitrite + Nitrate as N
Runtime Hours
Irom mg/L
Manganese mg/L
SAR
Electrical Conductivity mS / Cm
Nitrite mg / L
Nitrate mg / L
Nitrate + Nitrite mg /L
Runtime Days
\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015, 2016\Graph's and Data\Nitric Trial 7 to\Trial 8 HNO3\
Trial 8 Nutrient Data and Graphs .xlsxChart 2

Alberta Agriculture and Forestry

Floc Agent

ACIDF 2015 Aerobic Digestion of Poultry Manure Project

Nutrient Recycling Group



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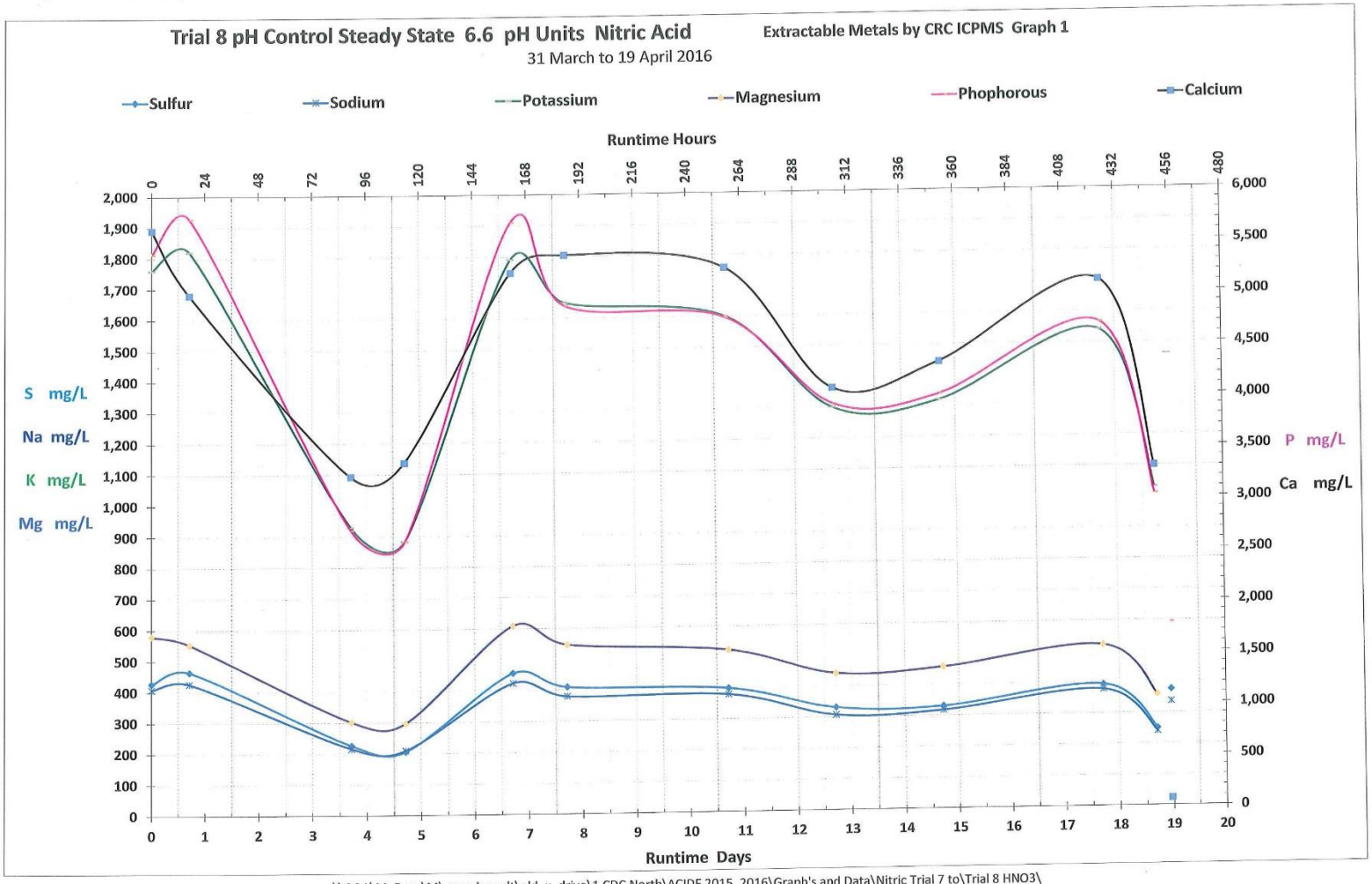
Trial 8 Nutrient Data and Graphs .xlsxChart 1



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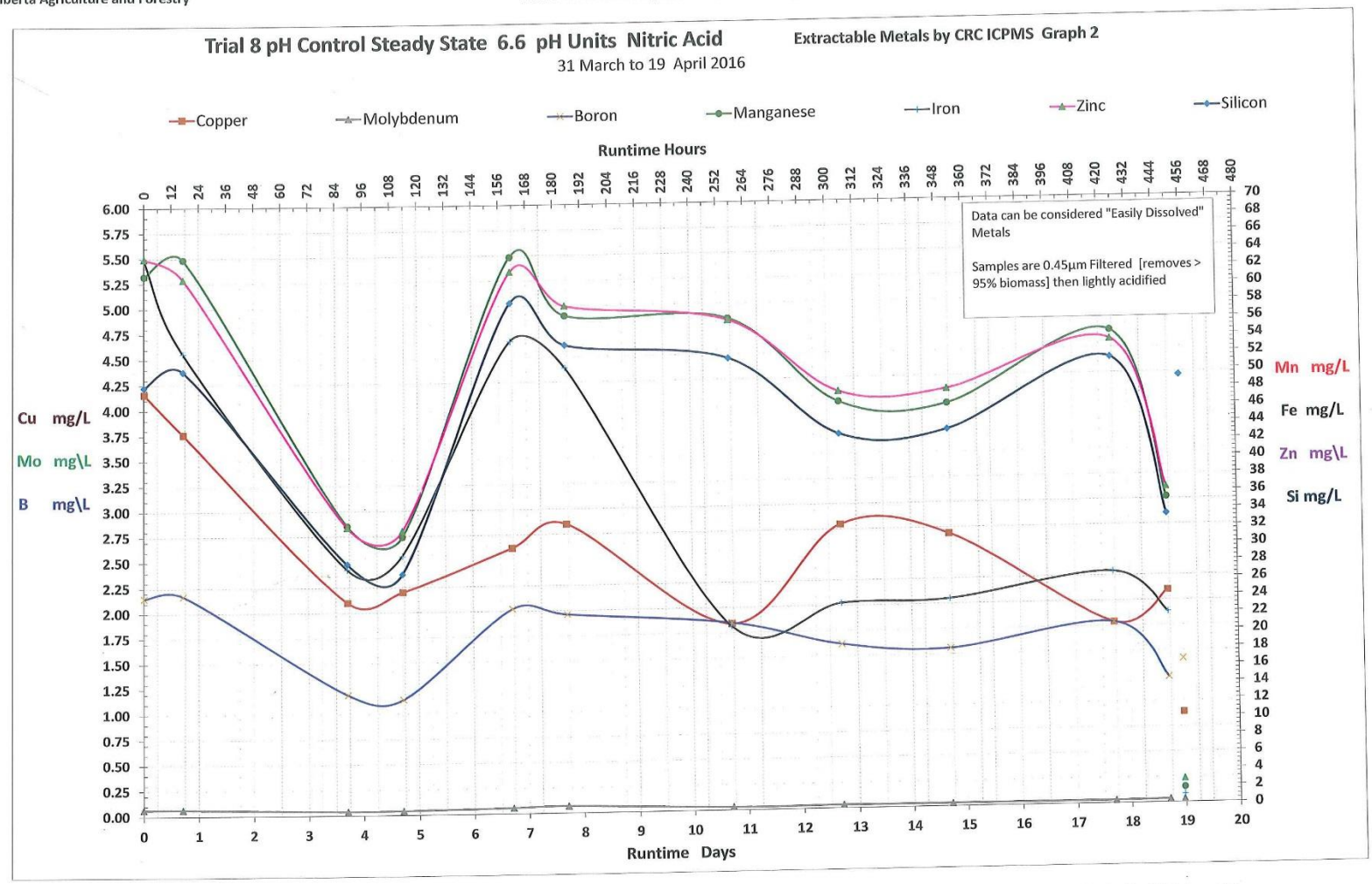
Trial 8 Nutrient Data and Graphs .xlsx Easily Dissolved Metals Graph 1



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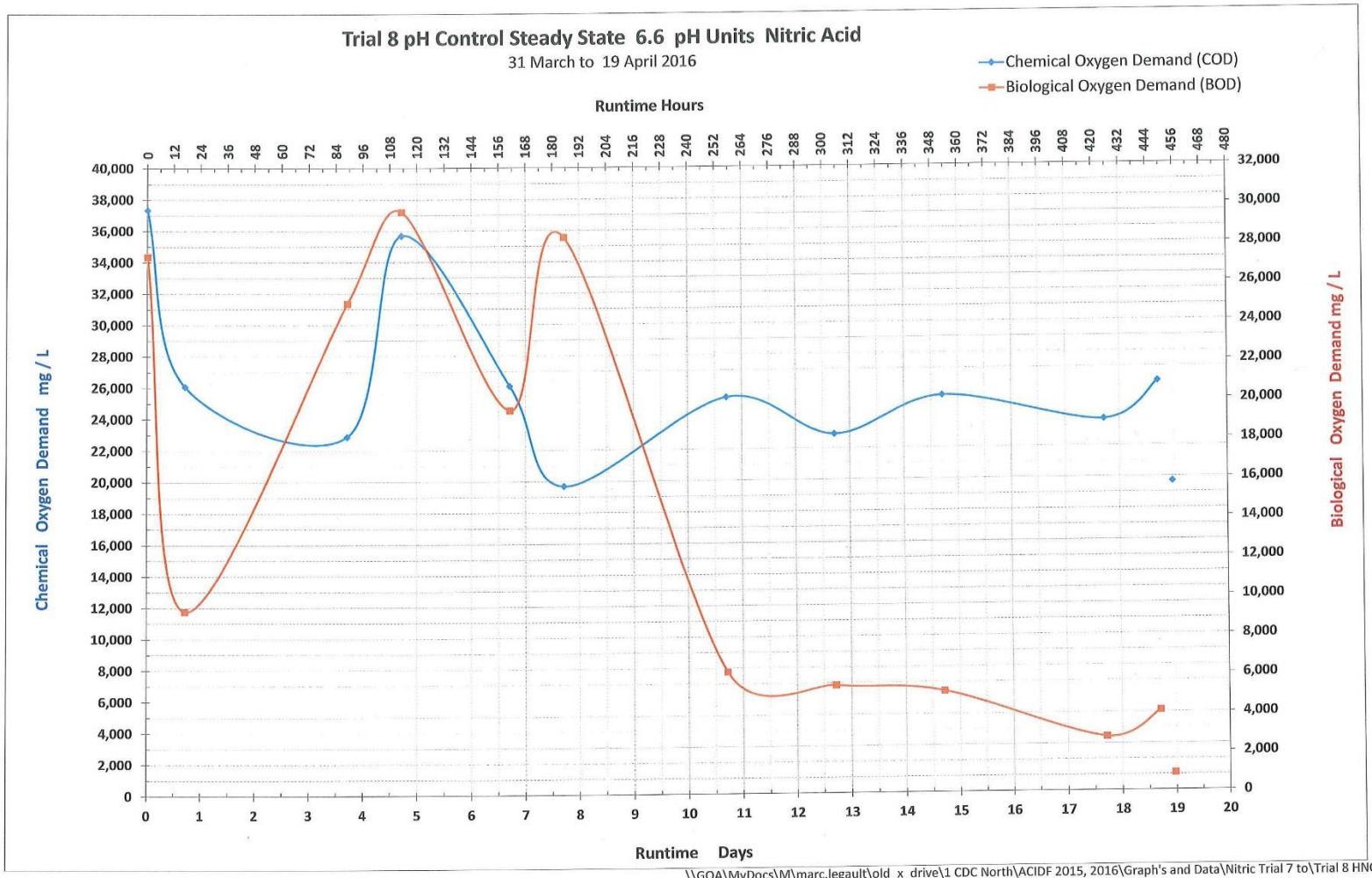

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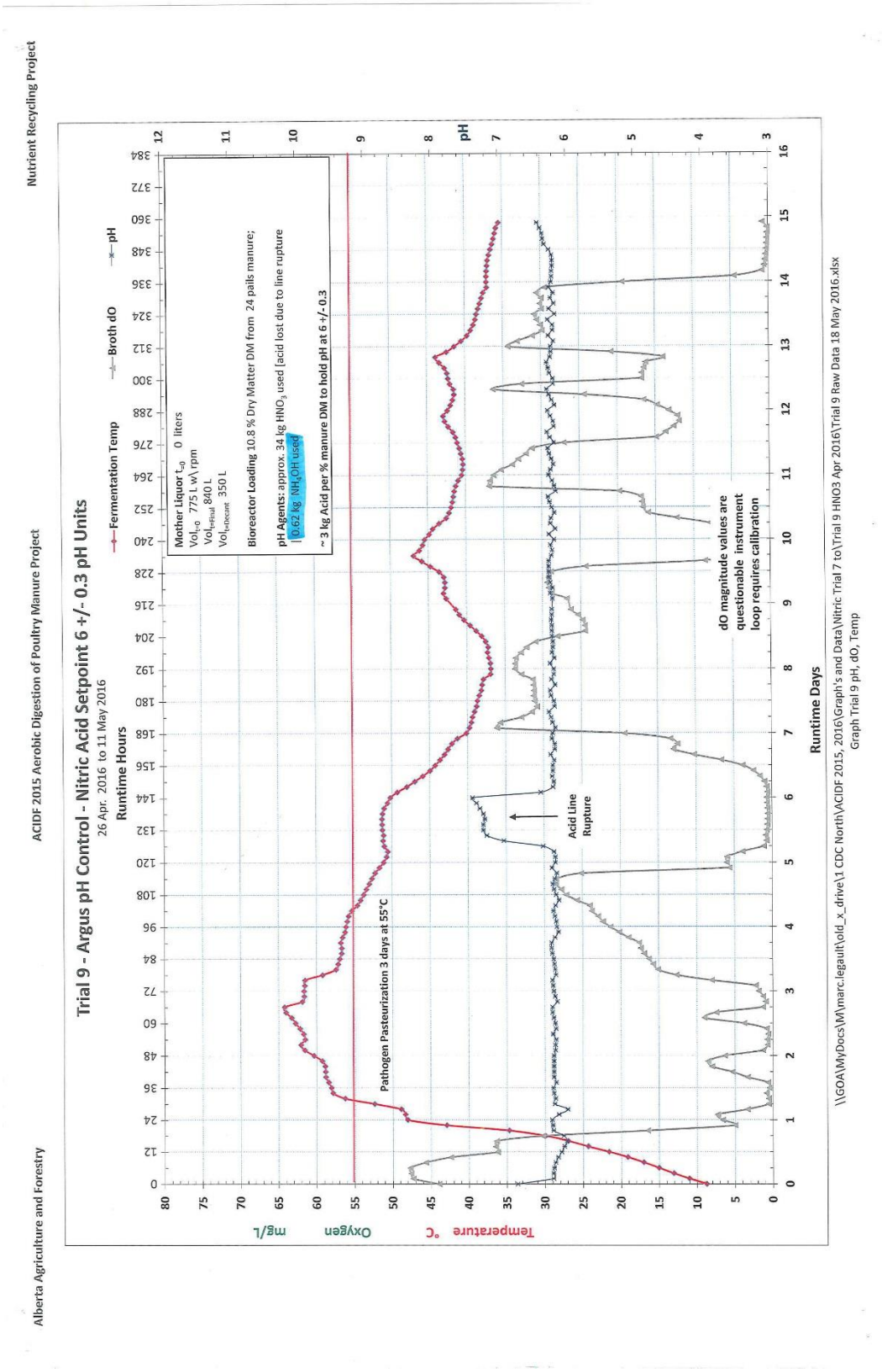
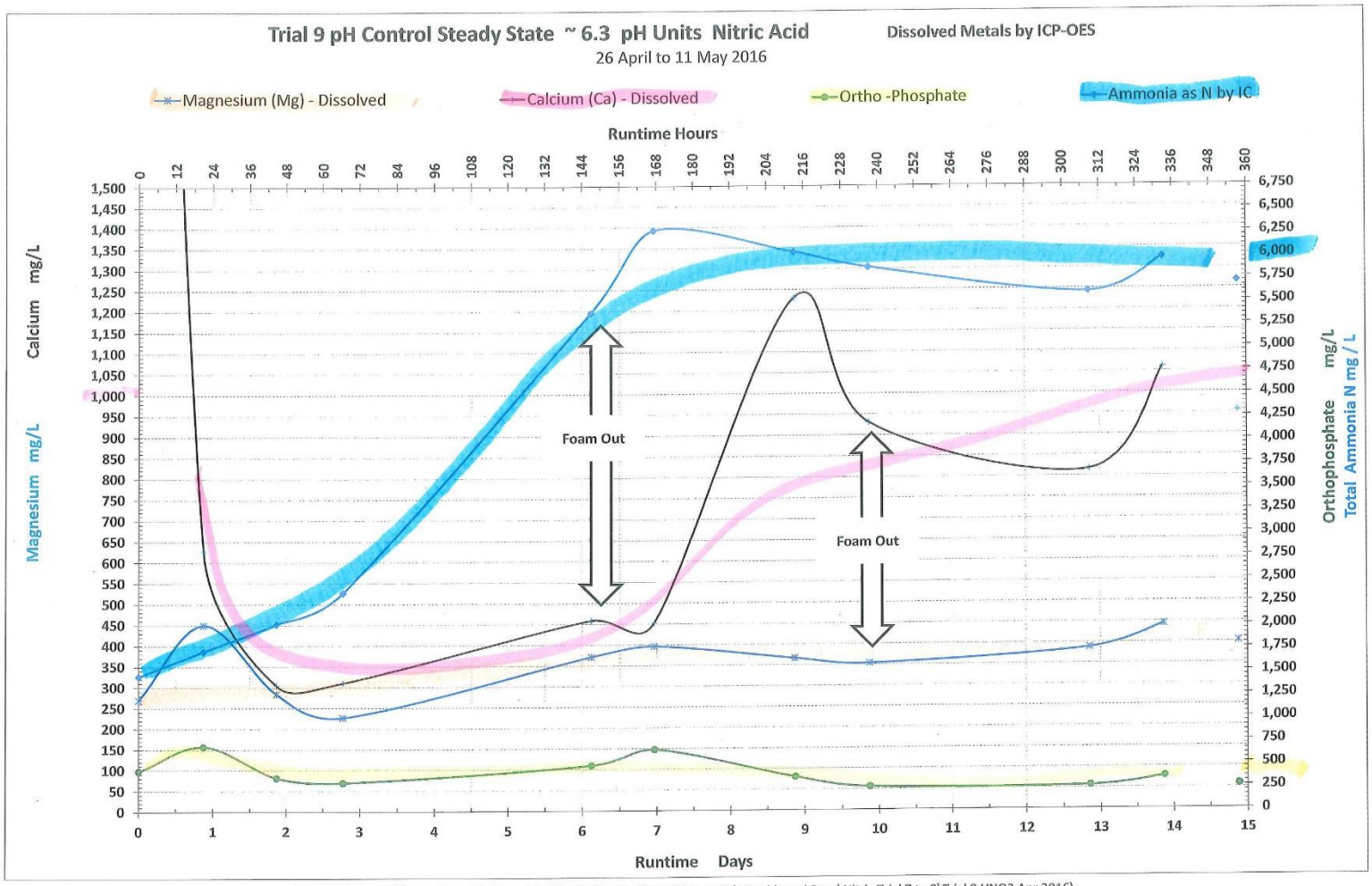
Trial 8 Nutrient Data and Graphs .xlsx
Chart 4


Nutrient Recycling Project
ACIDF 2015 Aerobic Digestion of Poultry Manure Project
Alberta Agriculture and Forestry
Trial 9 - Argus pH Control - Nitric Acid Setpoint 6 +/- 0.3 pH Units
26 Apr. 2016 to 11 May 2016
Runtime Hours
Fermentation Temp
Broth dO
pH
Mother Liquor $t_{=0}$ 0 liters
$Vol_{t=0}$ 775 l w\ rpm
Vol_{tFinal} 840 L
$Vol_{t=Decant}$ 350 L
Bioreactor Loading 10.8 % Dry Matter DM from 24 pails manure;
pH Agents: approx. 34 kg HNO_3 used [acid lost due to line rupture
0.62 kg NH_4OH used
~ 3 kg Acid per % manure DM to hold pH at 6 +/- 0.3
Pathogen Pasteurization 3 days at 55°C
Acid Line Rupture
dO magnitude values are questionable instrument loop requires calibration
Temperature °C
Oxygen mg/L
pH
Runtime Days
\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015, 2016\Graph's and Data\Nitric Trial 7 to\Trial 9 HNO3 Apr 2016\Trial 9 Raw Data 18 May 2016.xlsx
Graph Trial 9 pH, dO, Temp



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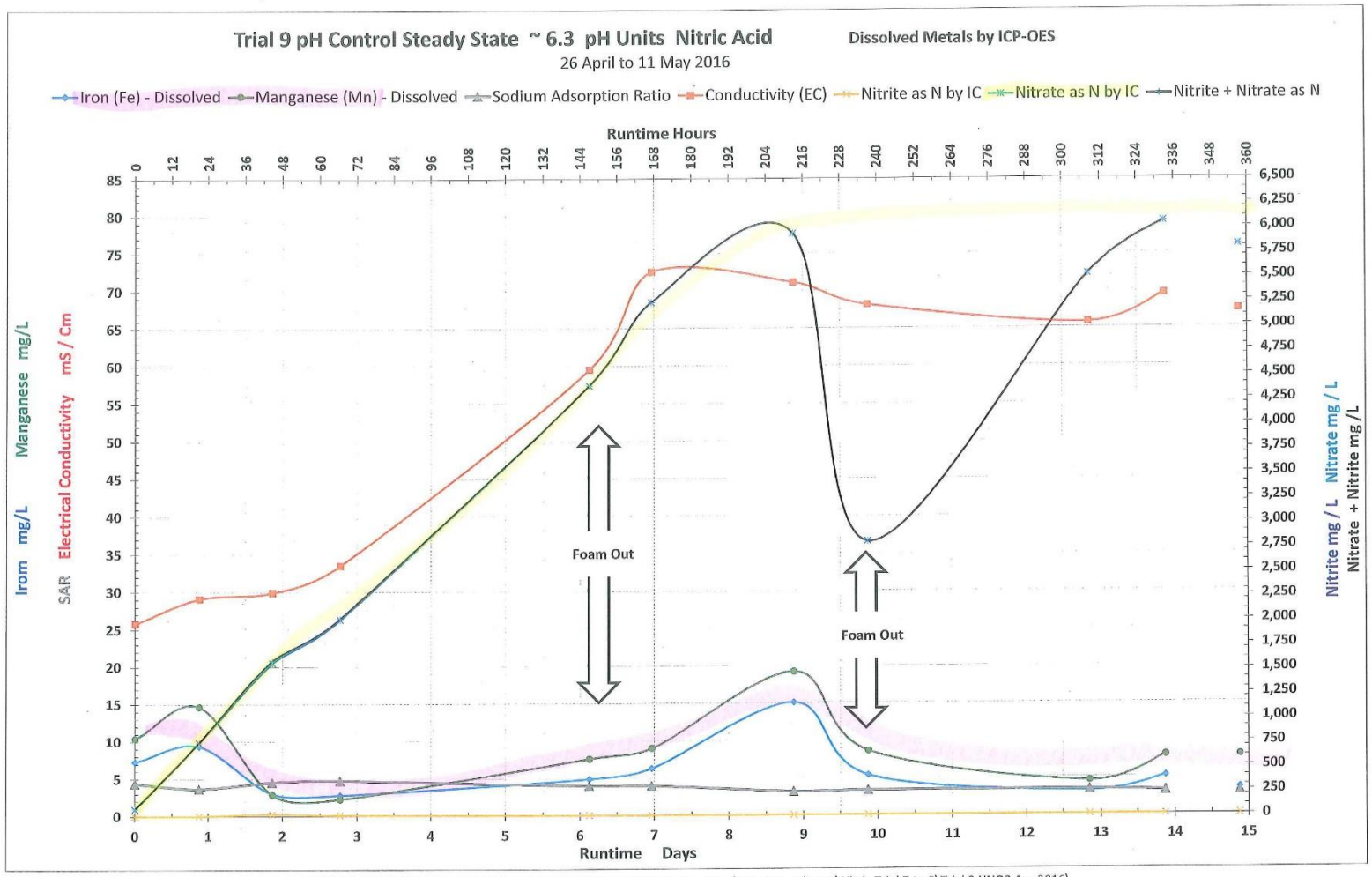
Trial 9 Nutrient Data and Graphs .xlsxChart 3



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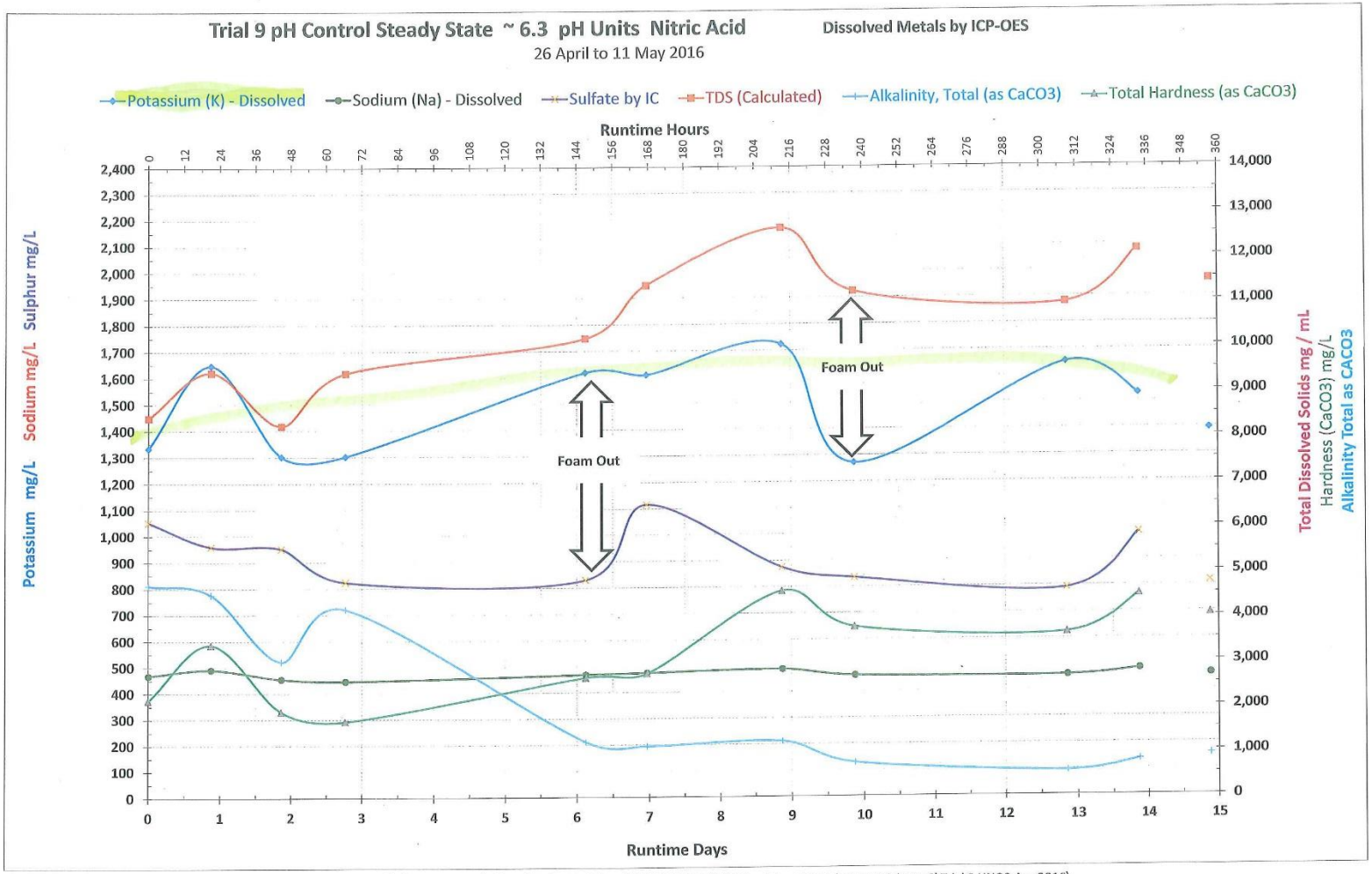
Trial 9 Nutrient Data and Graphs .xlsxChart 2



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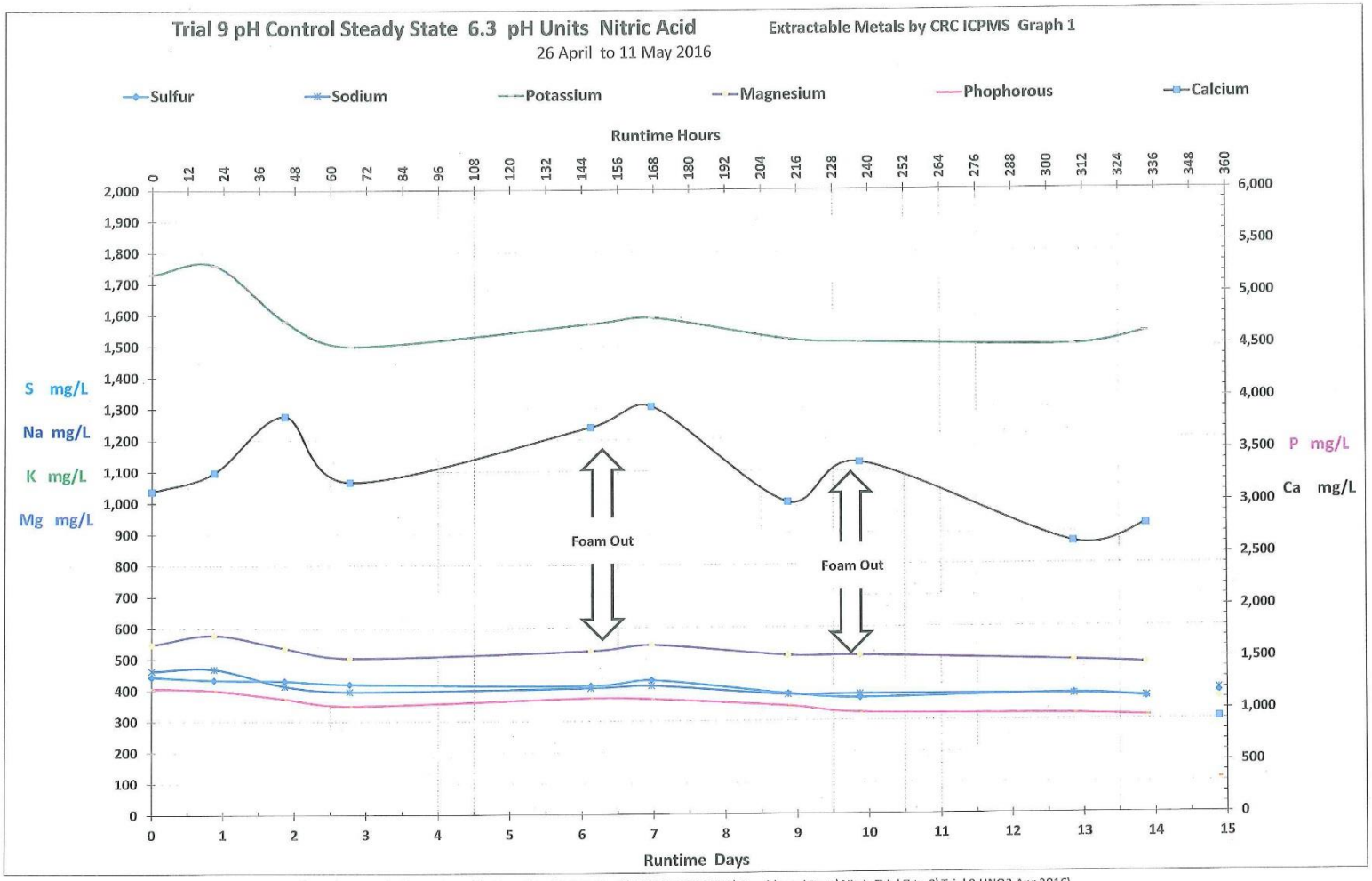
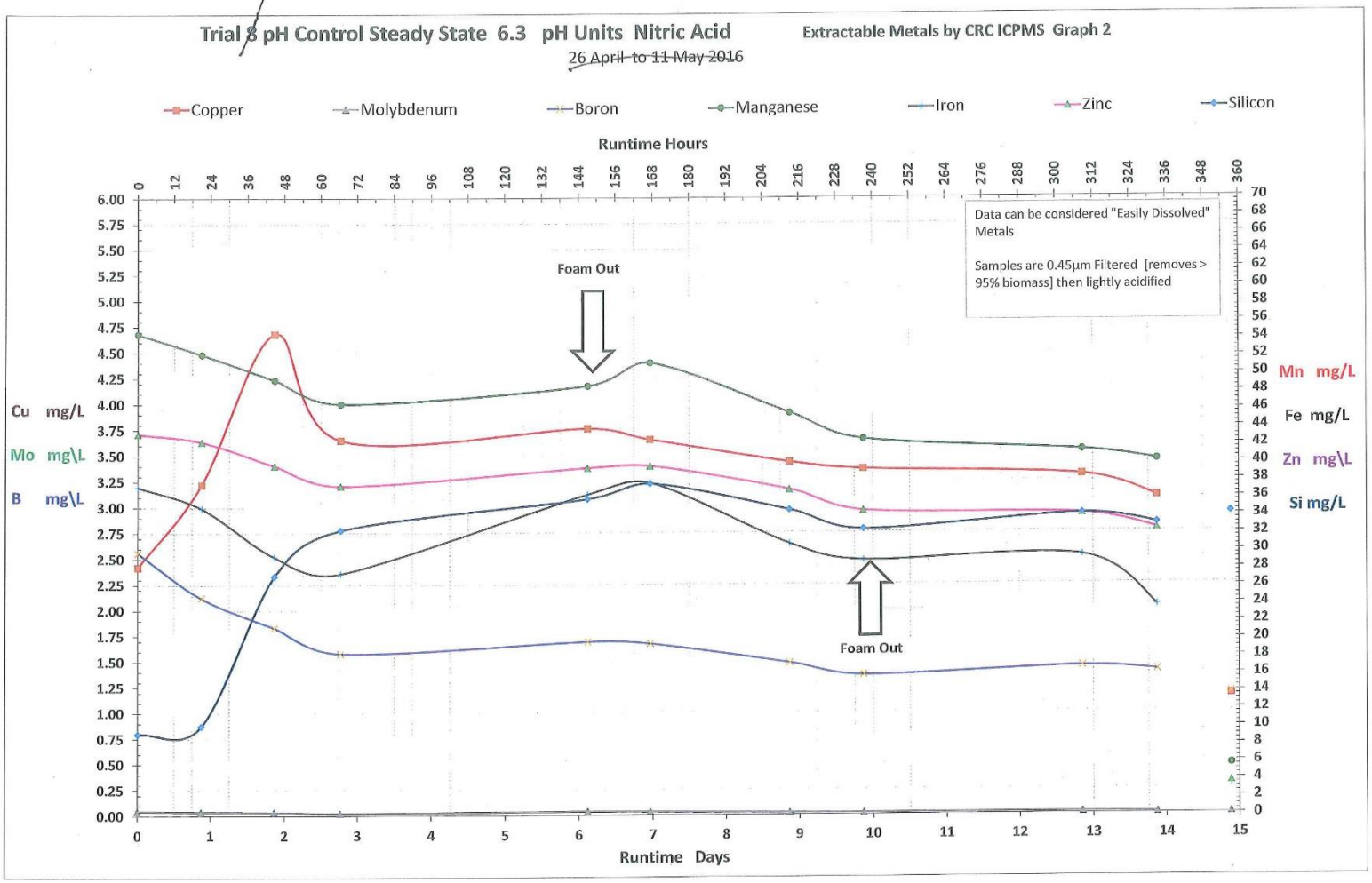
Trial 9 Nutrient Data and Graphs .xlsxChart 1



\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015, 2016\Graph's and Data\Nitric Trial 7 to 9\Trial 9 HNO3 Apr 2016\
Trial 9 Nutrient Data and Graphs .xlsx Easily Dissolved Metals Graph 1



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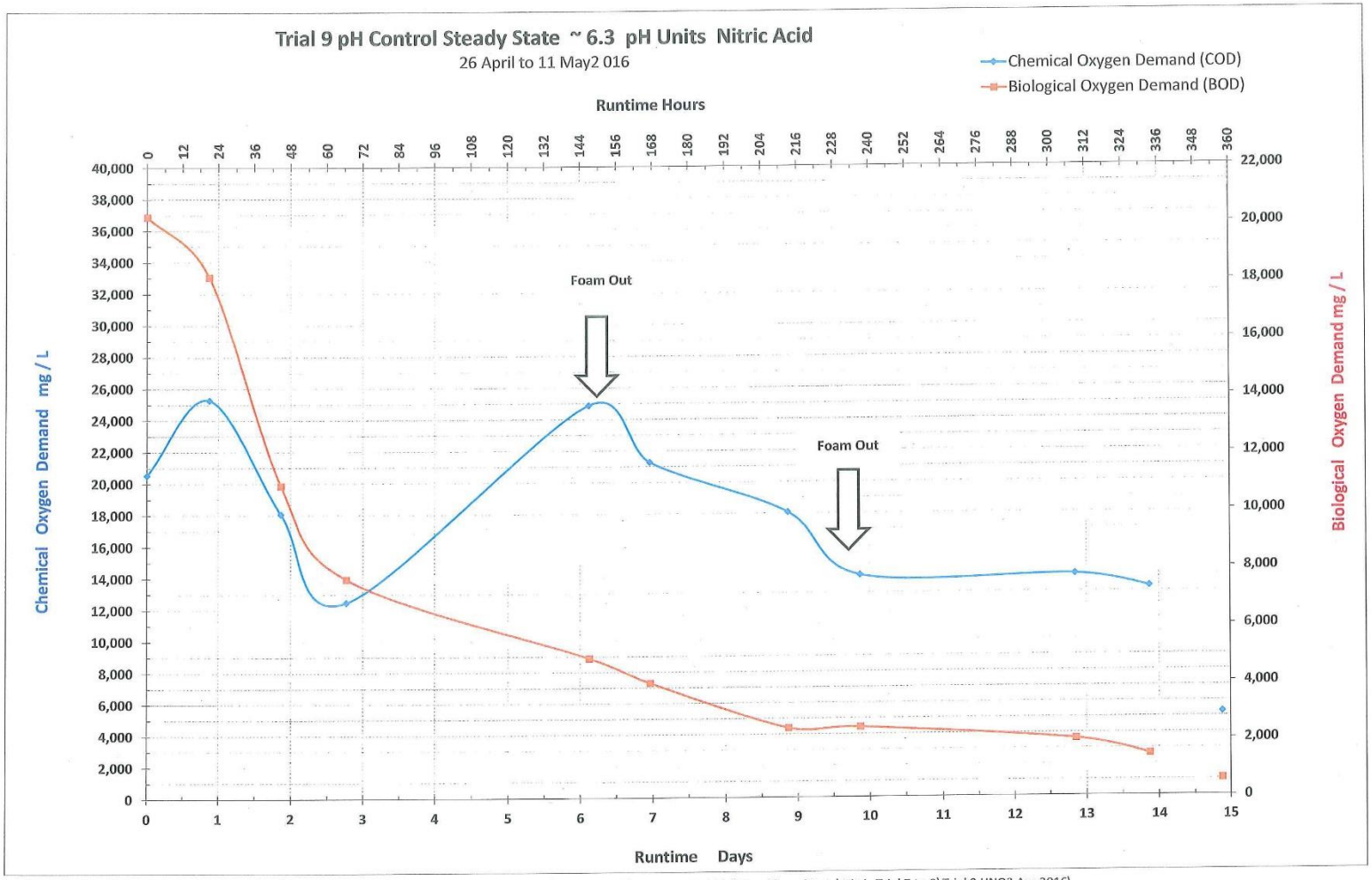

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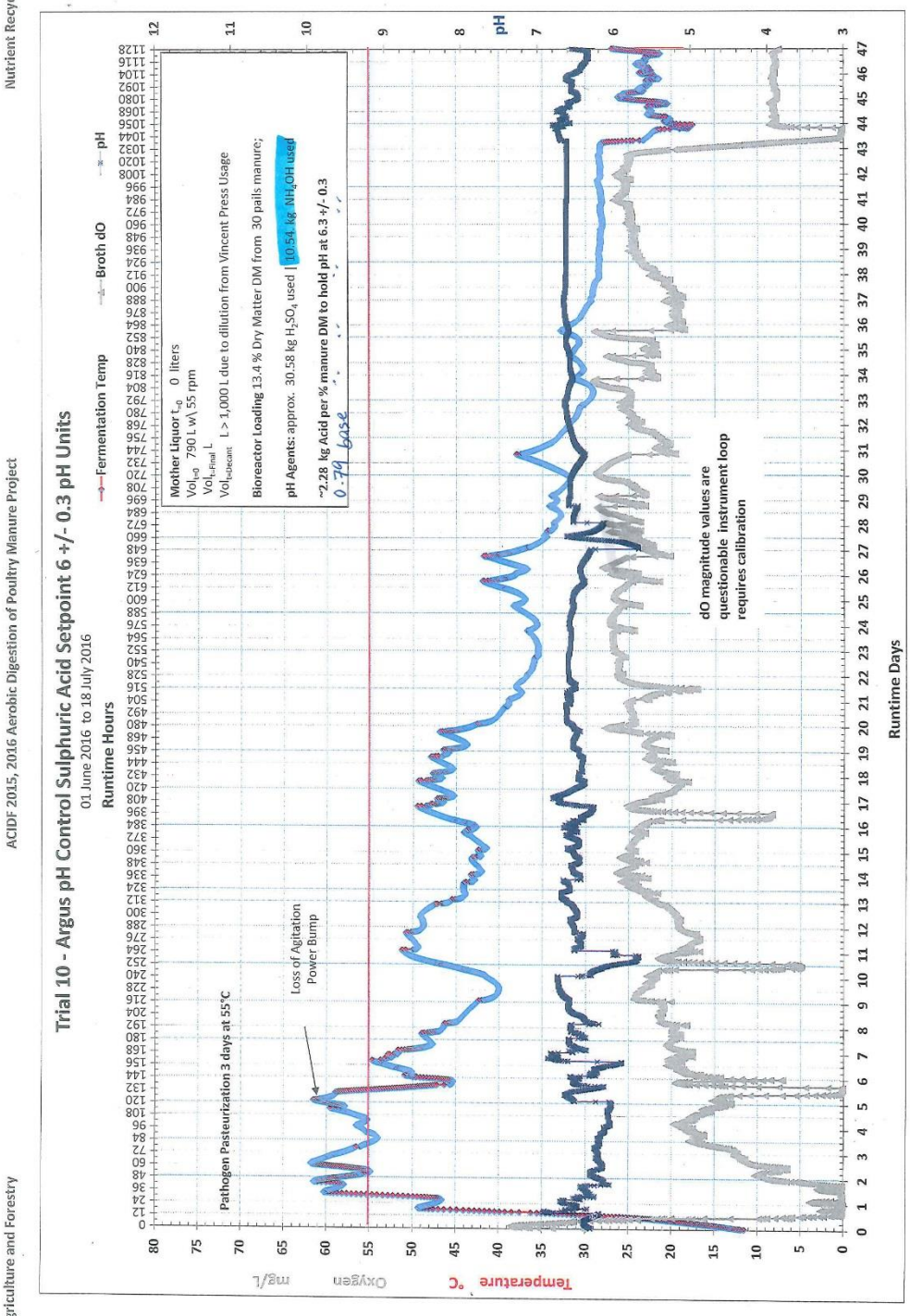
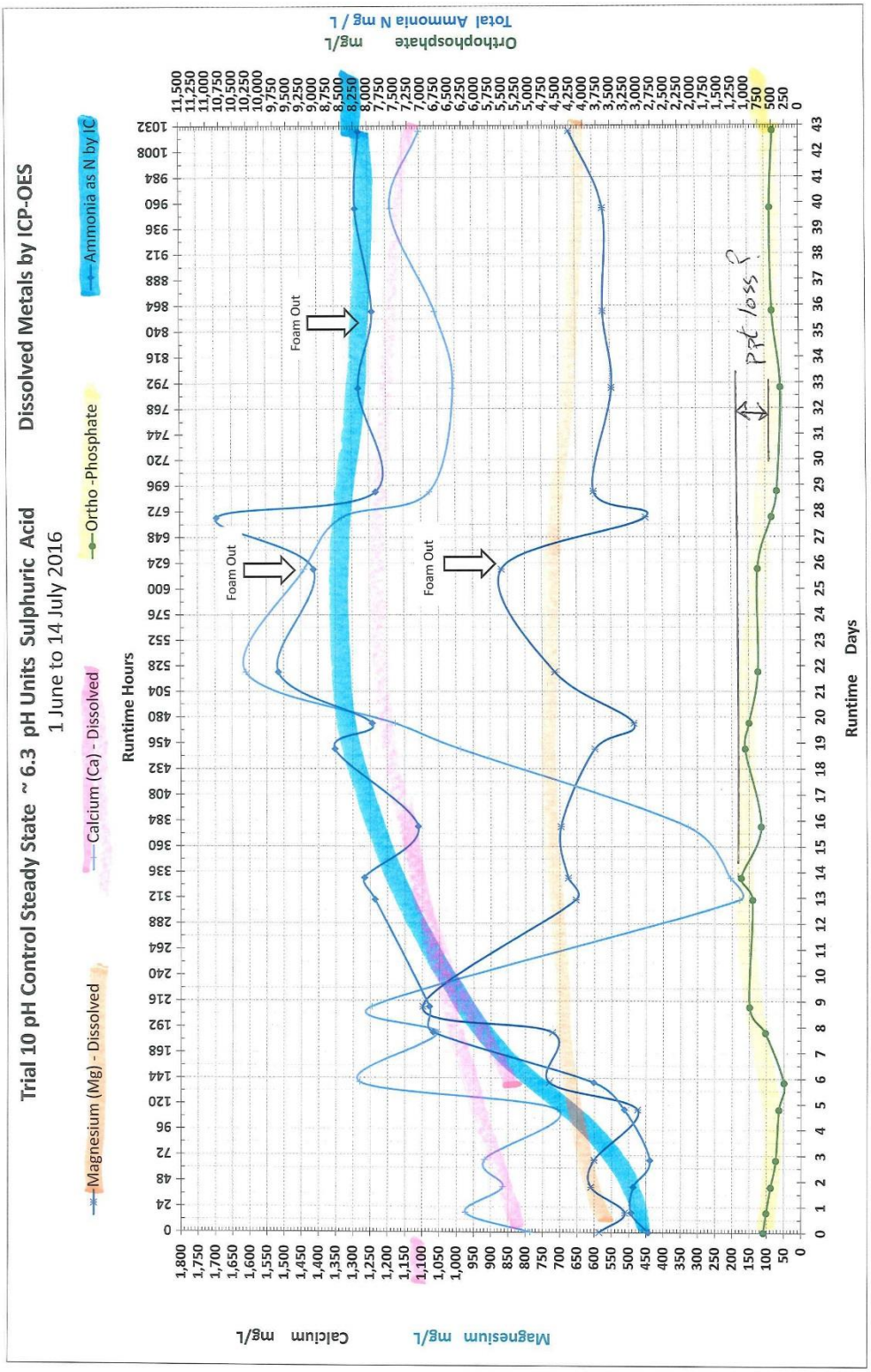
Trial 9 Nutrient Data and Graphs .xlsxChart 4


Alberta Agriculture and Forestry
ACIDF 2015, 2016 Aerobic Digestion of Poultry Manure Project
Nutrient Recycling Project
Trial 10 - Argus pH Control Sulphuric Acid Setpoint 6 +/- 0.3 pH Units
01 June 2016 to 18 July 2016
Runtime Hours
Fermentation Temp
Broth dO
pH
Mother Liquor $t_{=0}$ 0 liters
$Vol_{t=0}$ 790 L w\ 55 rpm
$Vol_{t=Final}$ L
$Vol_{t=Decant}$ L > 1,000 L due to dilution from Vincent Press Usage
Bioreactor Loading 13.4 % Dry Matter DM from 30 pails manure;
pH Agents: approx. 30.58 kg H_2SO_4 used | 10.54 kg NH_4OH used
~2.28 kg Acid per % manure DM to hold pH at 6.3 +/- 0.3
0.79 base
Pathogen Pasteurization 3 days at 55°C
Loss of Agitation Power Bump
dO magnitude values are questionable instrument loop requires calibration
Temperature °C
Oxygen mg/L
pH
Runtime Days
C:\Users\marc.legault\AppData\Roaming\Microsoft\Excel\Trial 10 Raw Argus Data (version 1).xlsb
Graph Trial 10 pH, dO Temp



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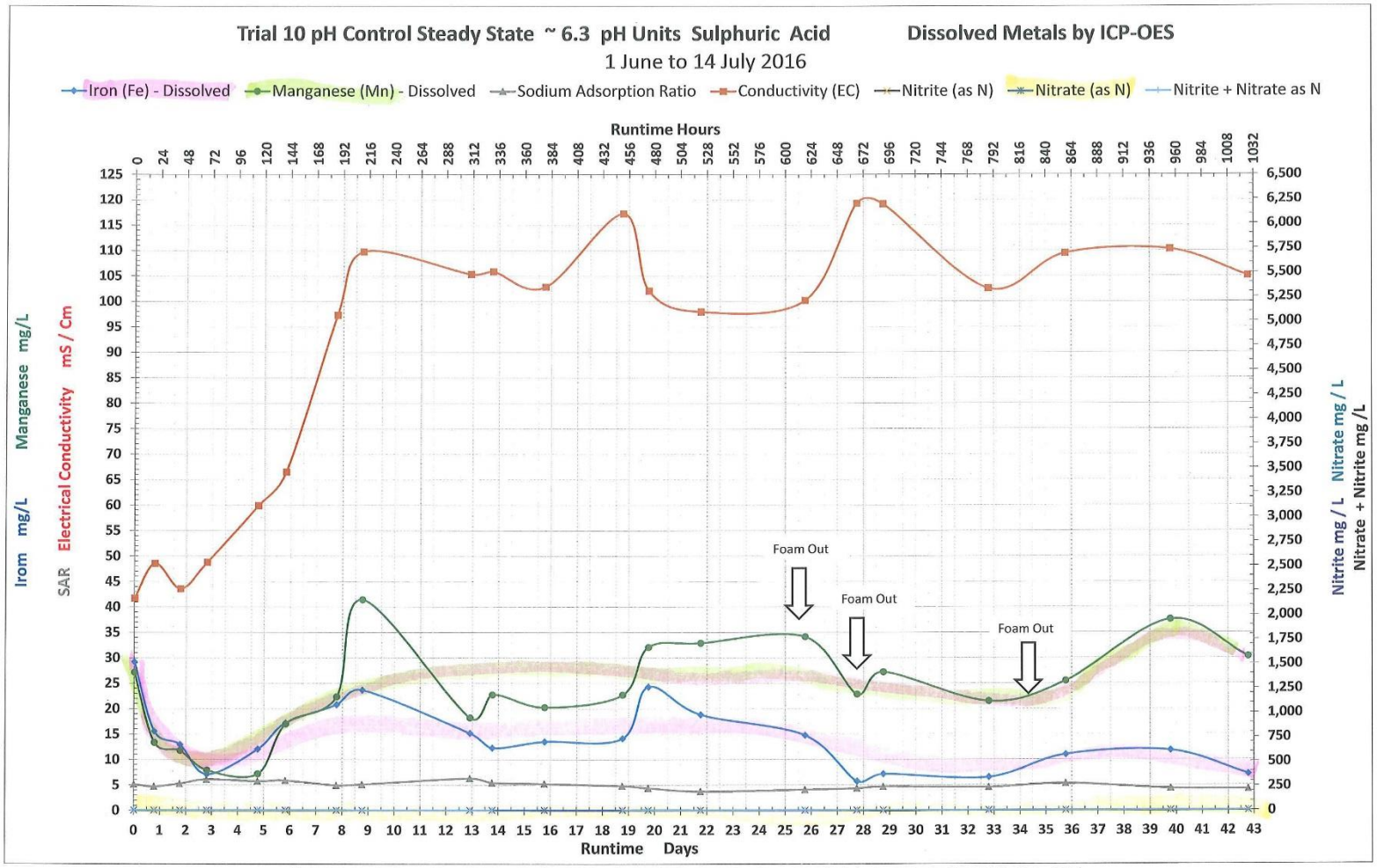
Trial 10 Nutrient Data and Graphs .xlsxChart 3



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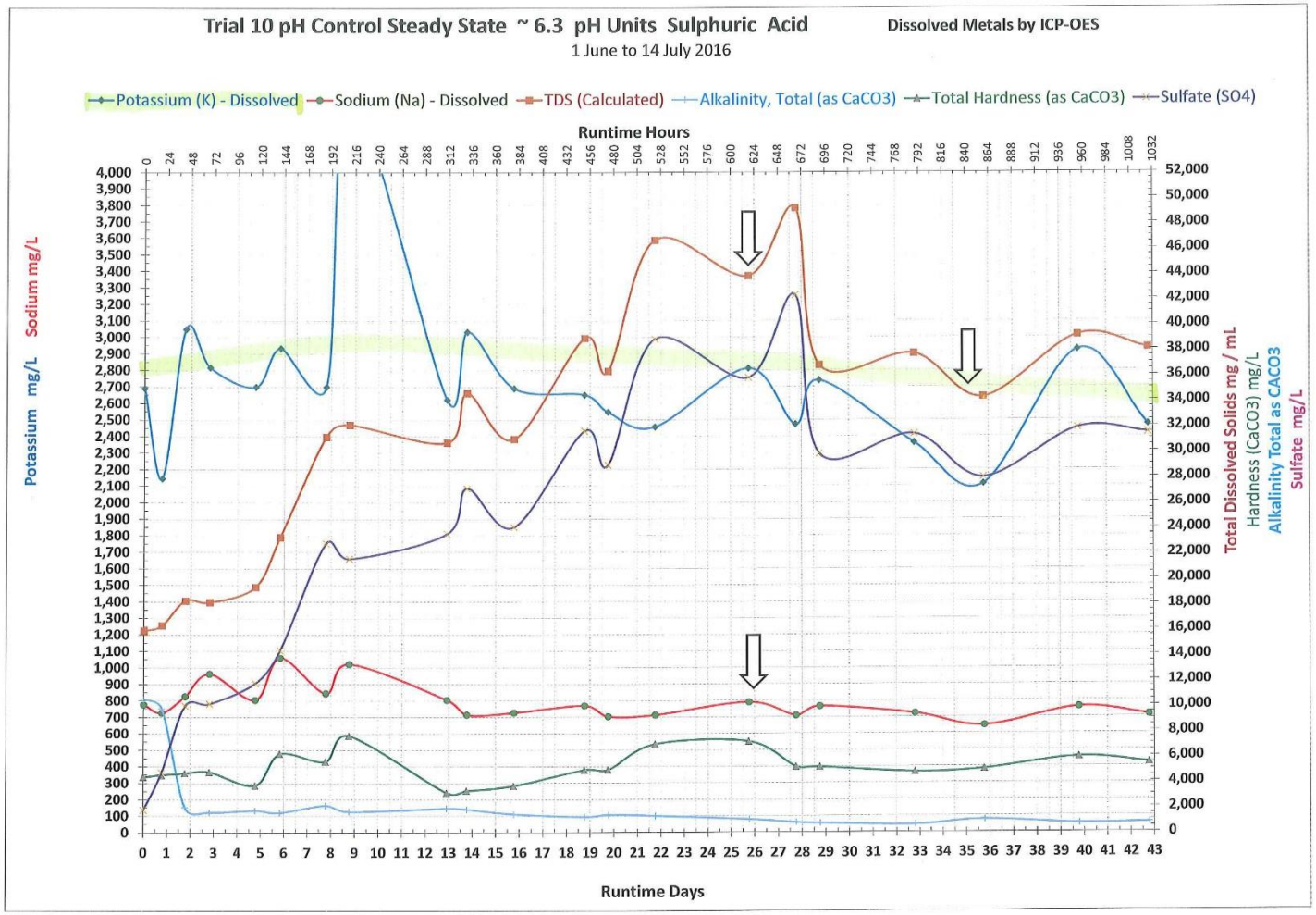
Trial 10 Nutrient Data and Graphs .xlsxChart 2



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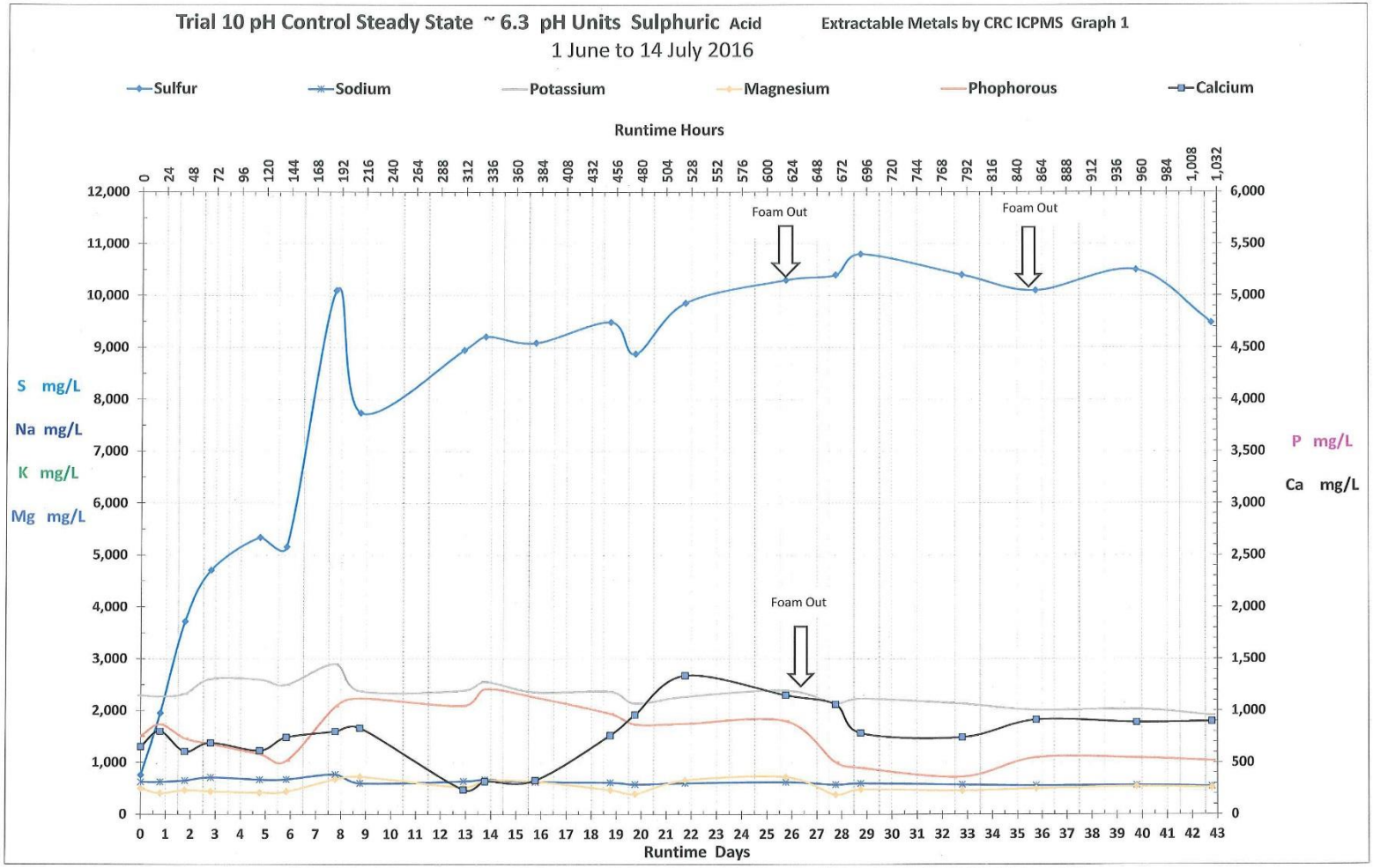
Trial 10 Nutrient Data and Graphs .xlsxChart 1



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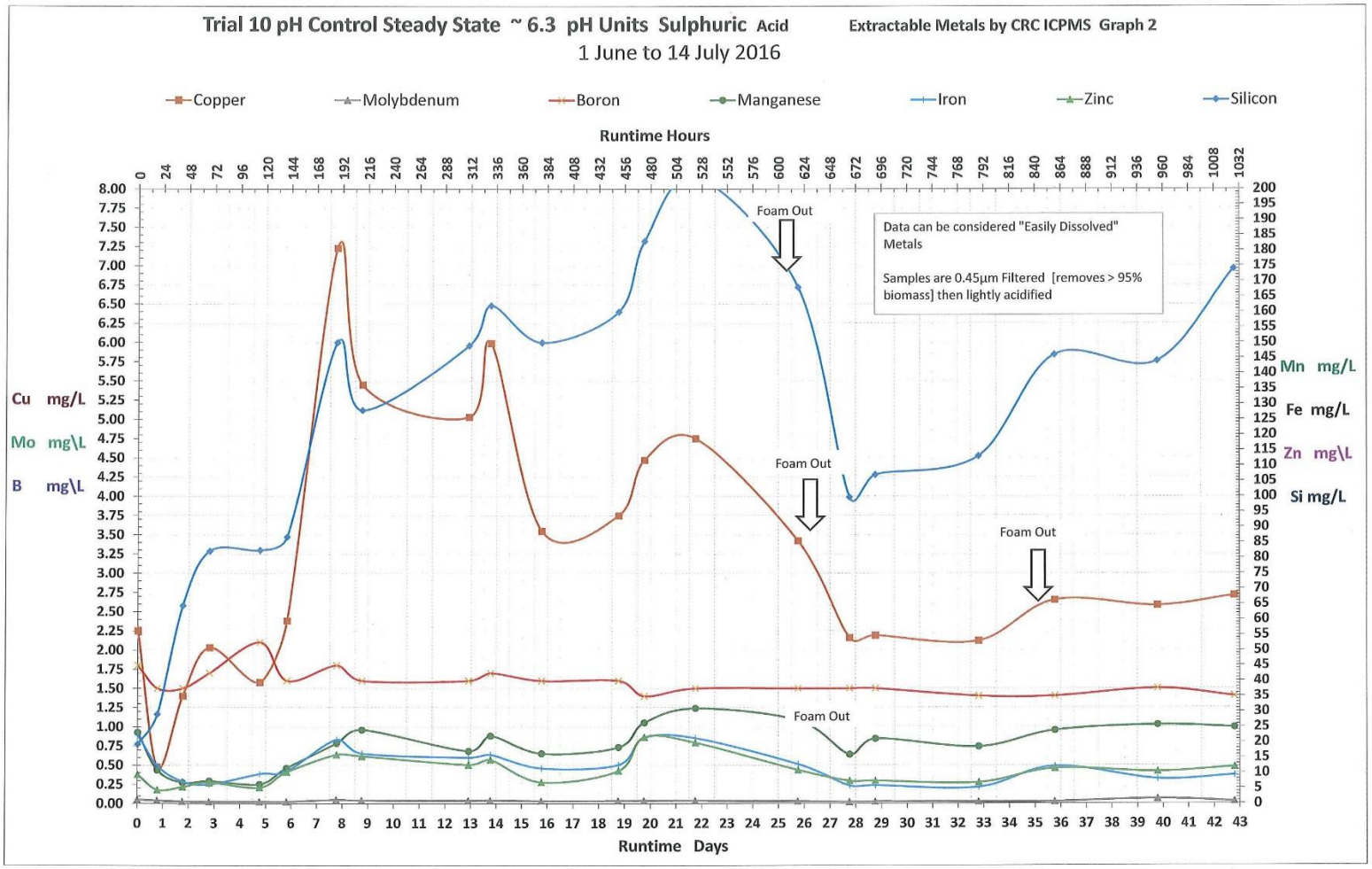
Trial 10 Nutrient Data and Graphs .xlsx Easily Dissolved Metals Graph 1



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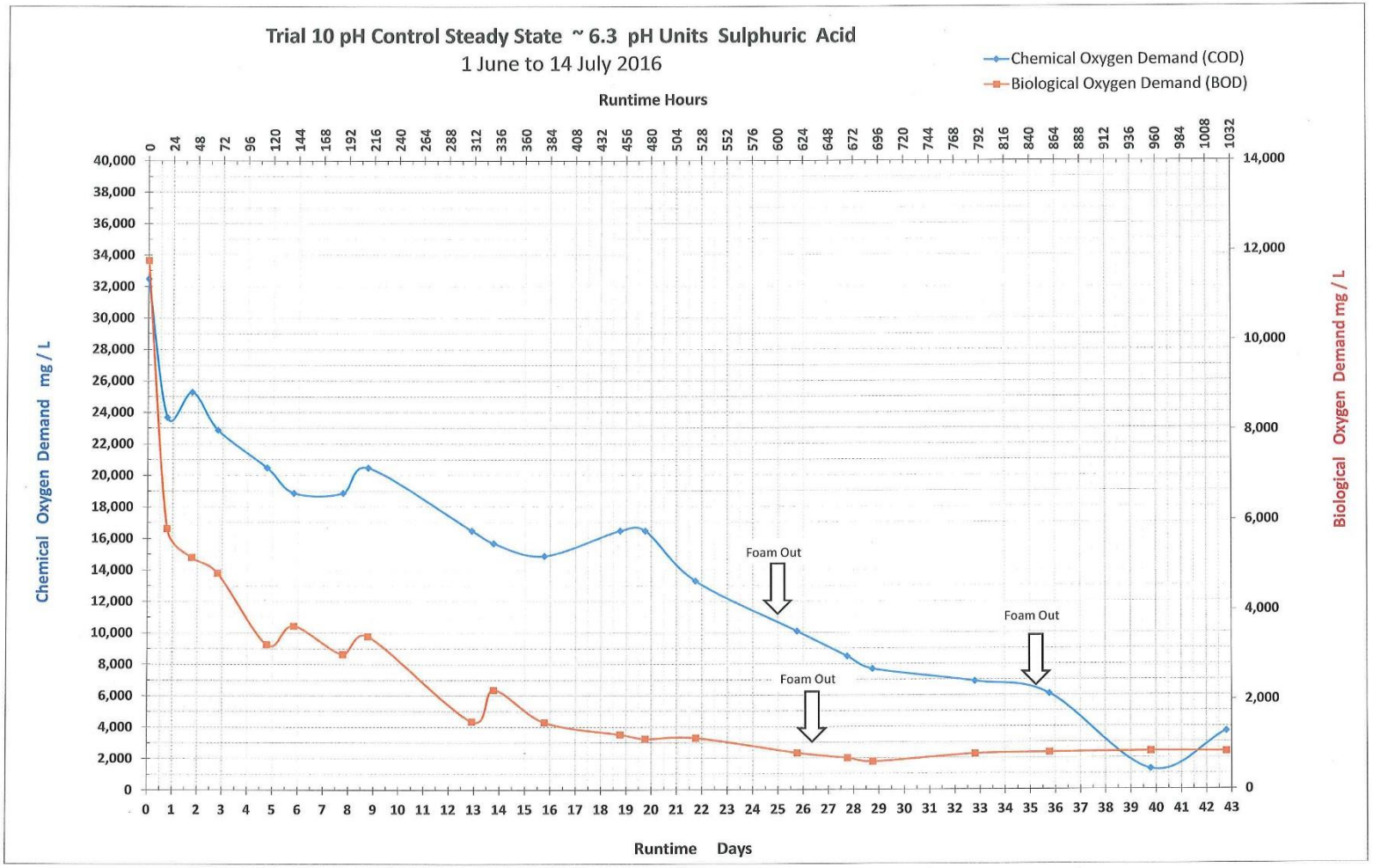

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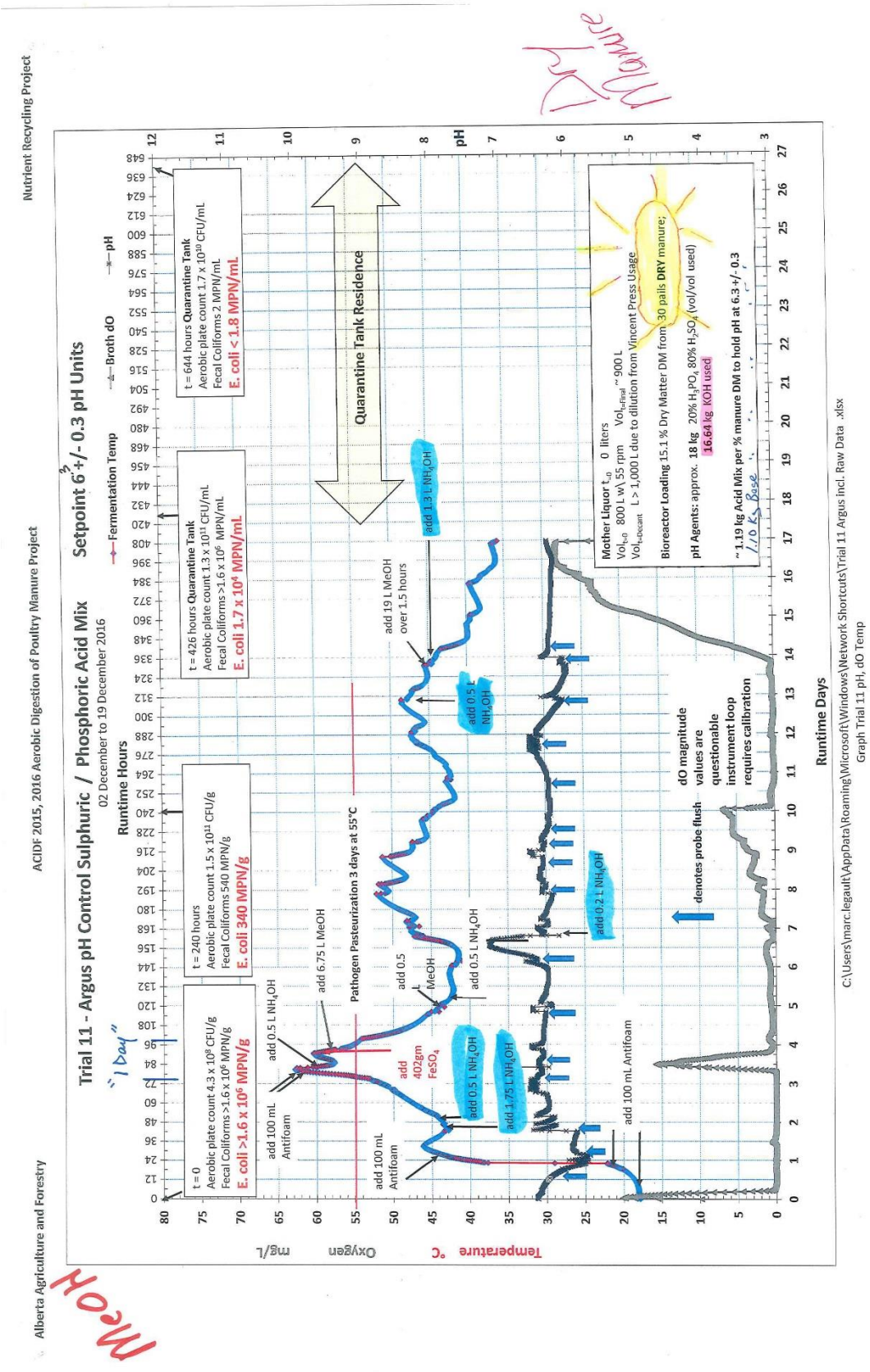
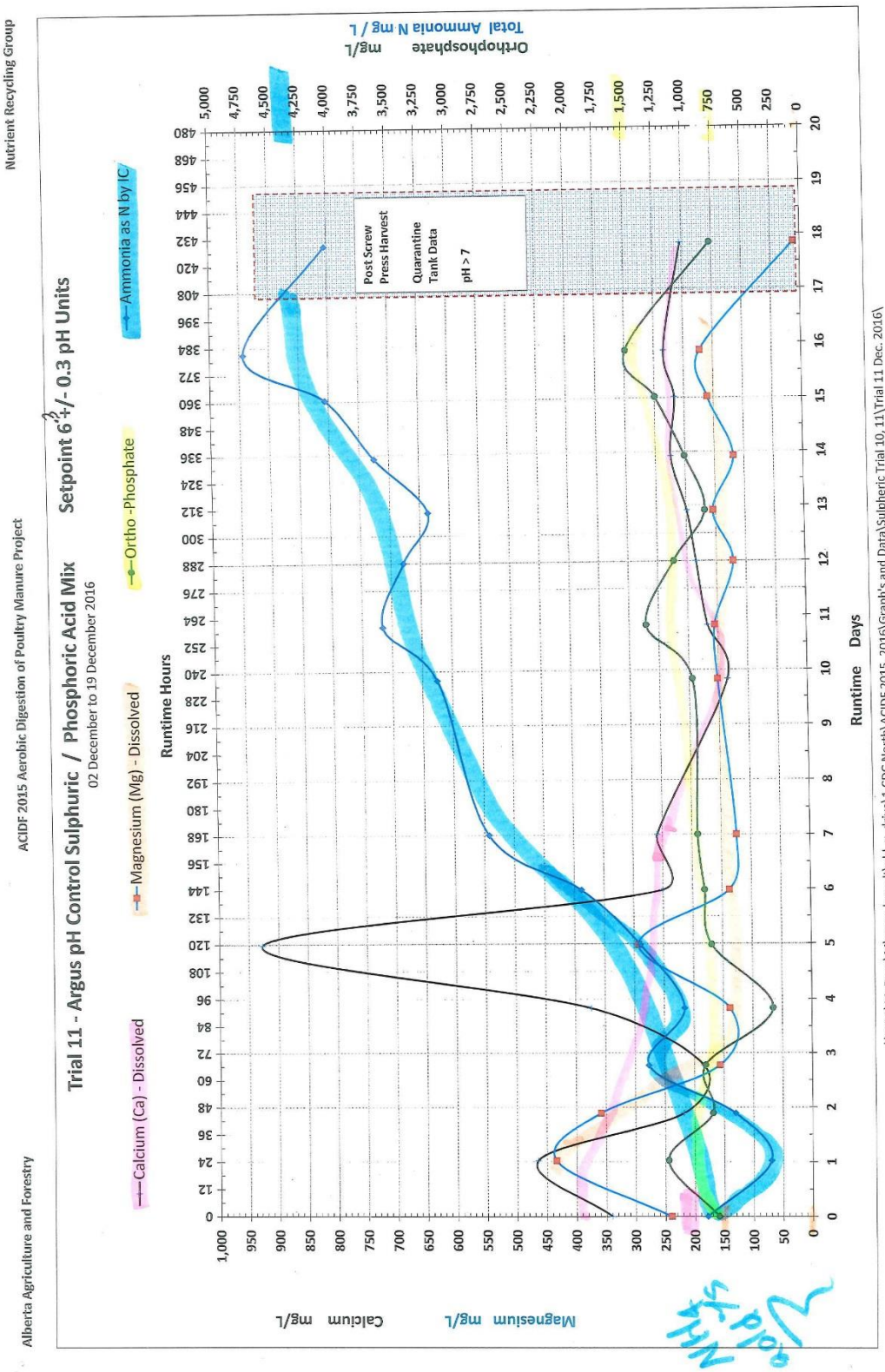
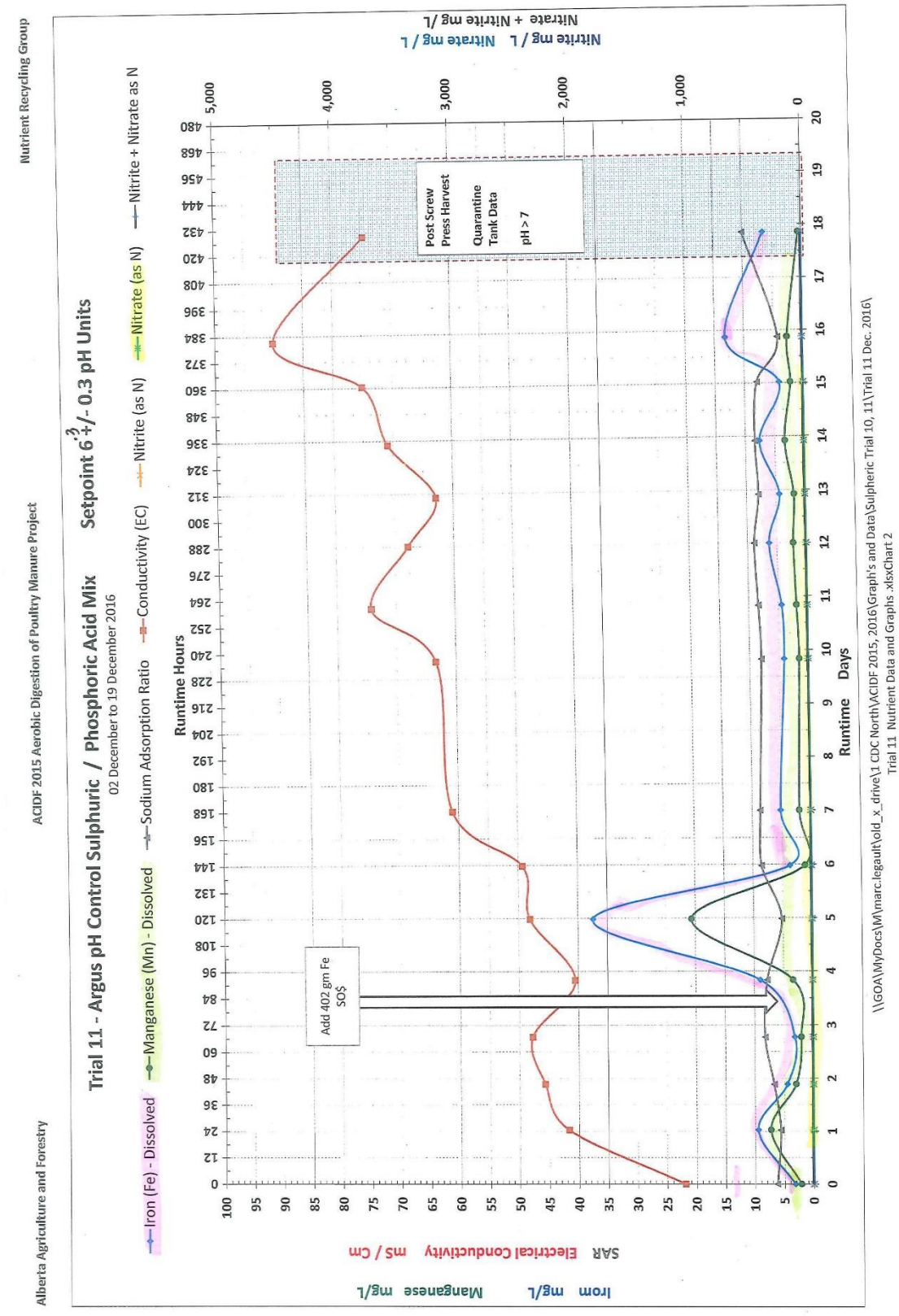
Trial 10 Nutrient Data and Graphs .xlsxChart 4


Nutrient Recycling Project
ACIDF 2015, 2016 Aerobic Digestion of Poultry Manure Project
Alberta Agriculture and Forestry
Trial 11 - Argus pH Control Sulphuric / Phosphoric Acid Mix
02 December to 19 December 2016
Setpoint 6.3 +/- 0.3 pH Units
Fermentation Temp
Broth dO
pH
Runtime Hours
Runtime Days
Temperature °C
Oxygen mg/L
pH
t = 0
Aerobic plate count 4.3 x 10^8 CFU/g
Fecal Coliforms >1.6 x 10^6 MPN/g
E. coli >1.6 x 10^6 MPN/g
t = 240 hours
Aerobic plate count 1.5 x 10^11 CFU/g
Fecal Coliforms 540 MPN/g
E. coli 340 MPN/g
t = 426 hours Quarantine Tank
Aerobic plate count 1.3 x 10^11 CFU/mL
Fecal Coliforms >1.6 x 10^6 MPN/mL
E. coli 1.7 x 10^4 MPN/mL
t = 644 hours Quarantine Tank
Aerobic plate count 1.7 x 10^10 CFU/mL
Fecal Coliforms 2 MPN/mL
E. coli < 1.8 MPN/mL
Quarantine Tank Residence
Pathogen Pasteurization 3 days at 55°C
add 100 mL Antifoam
add 0.5 L NH4OH
add 6.75 L MeOH
add 0.5 L MeOH
add 402gm FeSO4
add 0.5 L NH4OH
add 1.75 L NH4OH
add 0.5 L NH4OH
add 19 L MeOH over 1.5 hours
add 1.3 L NH4OH
add 0.5 L NH4OH
add 0.2 L NH4OH
add 100 mL Antifoam
denotes probe flush
dO magnitude values are questionable instrument loop requires calibration
Mother Liquor t_i,0 0 liters
Vol_t=0 800 L w\ 55 rpm Vol_t=Final ~ 900 L
Vol_t=Decant L > 1,000 L due to dilution from Vincent Press Usage
Bioreactor Loading 15.1 % Dry Matter DM from 30 pails DRY manure;
pH Agents: approx. 18 kg 20% H3PO4 80% H2SO4 (vol/vol used)
16.64 kg KOH used
~1.19 kg Acid Mix per % manure DM to hold pH at 6.3 +/- 0.3
1.10 KOH Base
C:\Users\marc.legault\AppData\Roaming\Microsoft\Windows\Network Shortcuts\Trial 11 Argus incl. Raw Data .xlsx
Graph Trial 11 pH, dO Temp


Nutrient Recycling Group
ACIDF 2015 Aerobic Digestion of Poultry Manure Project
Alberta Agriculture and Forestry
Trial 11 - Argus pH Control Sulphuric / Phosphoric Acid Mix
Setpoint 6.2 +/- 0.3 pH Units
02 December to 19 December 2016
Calcium (Ca) - Dissolved
Magnesium (Mg) - Dissolved
Ortho -Phosphate
Ammonia as N by IC
Runtime Hours
Runtime Days
Post Screw Press Harvest
Quarantine Tank Data
pH > 7
Calcium mg/L
Magnesium mg/L
Orthophosphate mg/L
Total Ammonia N mg / L
\\GOA\MyDocs\W\marc.legault\old_x_drive\1 CDC North\ACIDF 2015, 2016\Graph's and Data\Sulpheric Trial 10, 11\Trial 11 Dec. 2016\
Trial 11 Nutrient Data and Graphs .xlsxChart 3

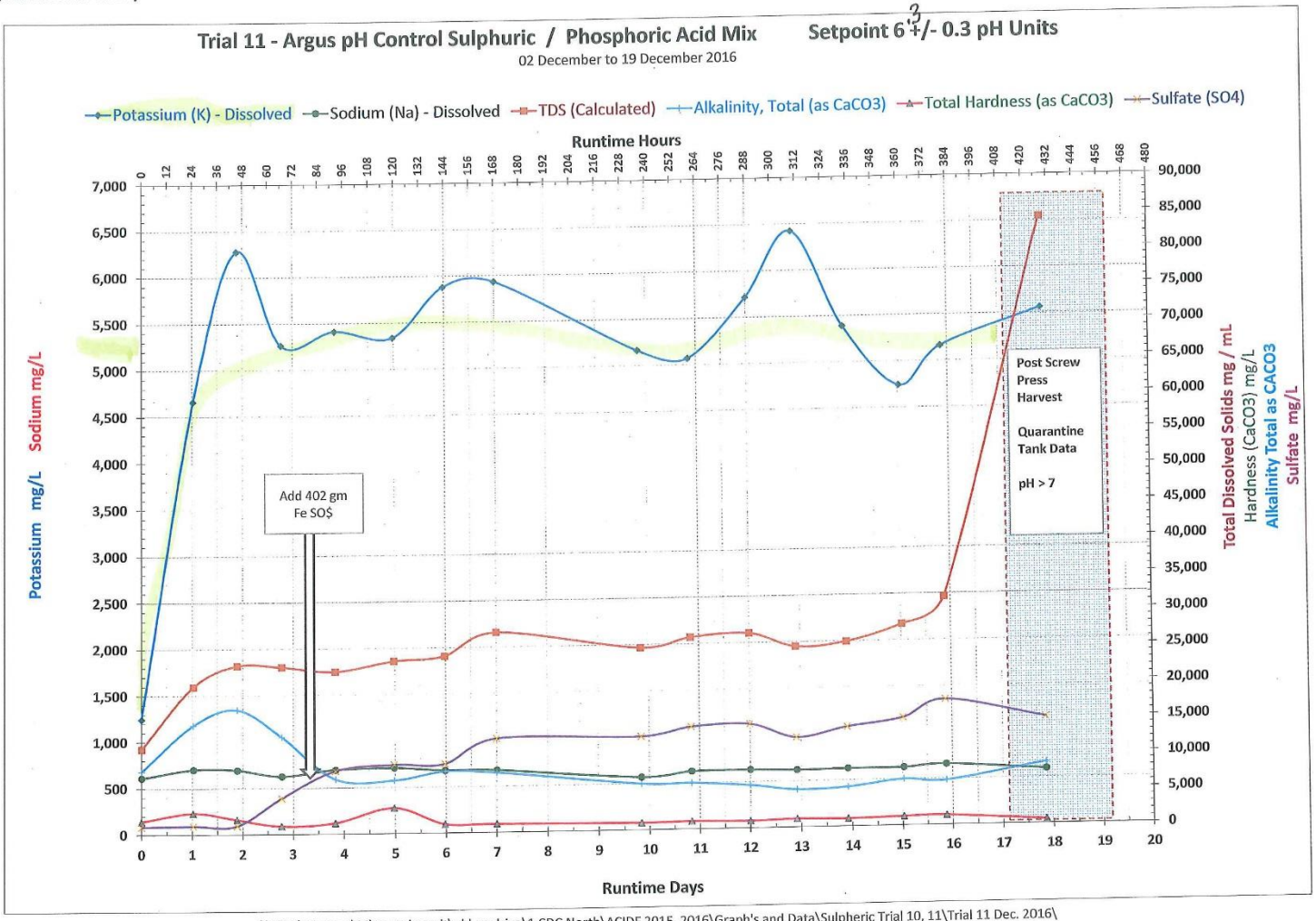

Nutrient Recycling Group
ACIDF 2015 Aerobic Digestion of Poultry Manure Project
Alberta Agriculture and Forestry
Trial 11 - Argus pH Control Sulphuric / Phosphoric Acid Mix
Setpoint 6.3 +/- 0.3 pH Units
02 December to 19 December 2016
Iron (Fe) - Dissolved
Manganese (Mn) - Dissolved
Sodium Adsorption Ratio
Conductivity (EC)
Nitrite (as N)
Nitrate (as N)
Nitrite + Nitrate as N
Runtime Hours
Runtime Days
Nitrite mg / L
Nitrate mg / L
Nitrate + Nitrite mg /L
Iron mg/L
Manganese mg/L
SAR
Electrical Conductivity mS / Cm
Add 402 gm Fe SO$
Post Screw Press Harvest
Quarantine Tank Data
pH > 7
\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015, 2016\Graph's and Data\Sulpheric Trial 10, 11\Trial 11 Dec. 2016\
Trial 11 Nutrient Data and Graphs .xlsxChart 2

Alberta Agriculture and Forestry

ACIDF 2015 Aerobic Digestion of Poultry Manure Project

Nutrient Recycling Group

KOH



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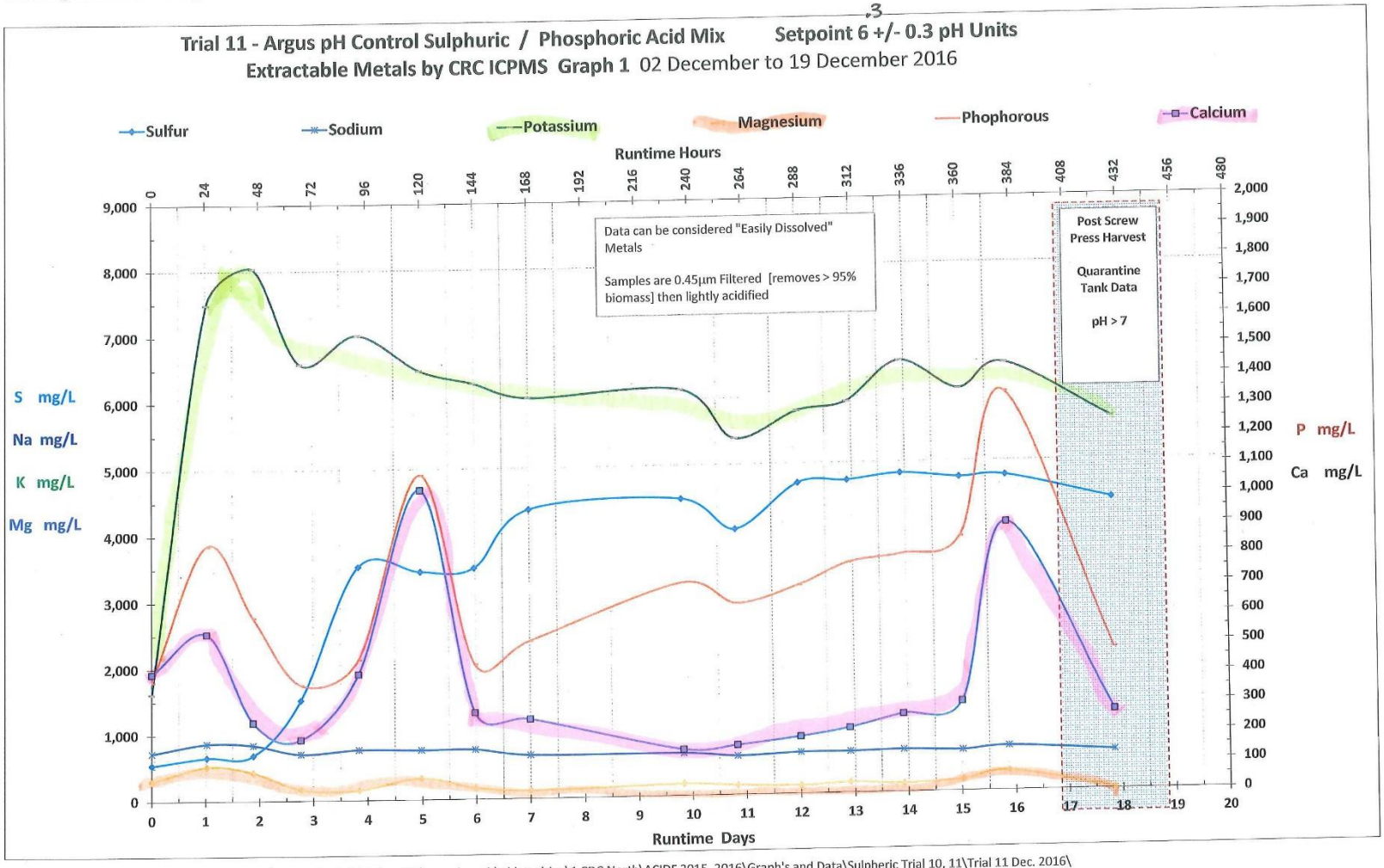
Trial 11 Nutrient Data and Graphs .xlsxChart 1

3

Trial 11 - Argus pH Control Sulphuric / Phosphoric Acid Mix Setpoint 6 +/- 0.3 pH Units
Extractable Metals by CRC ICPMS Graph 1 02 December to 19 December 2016



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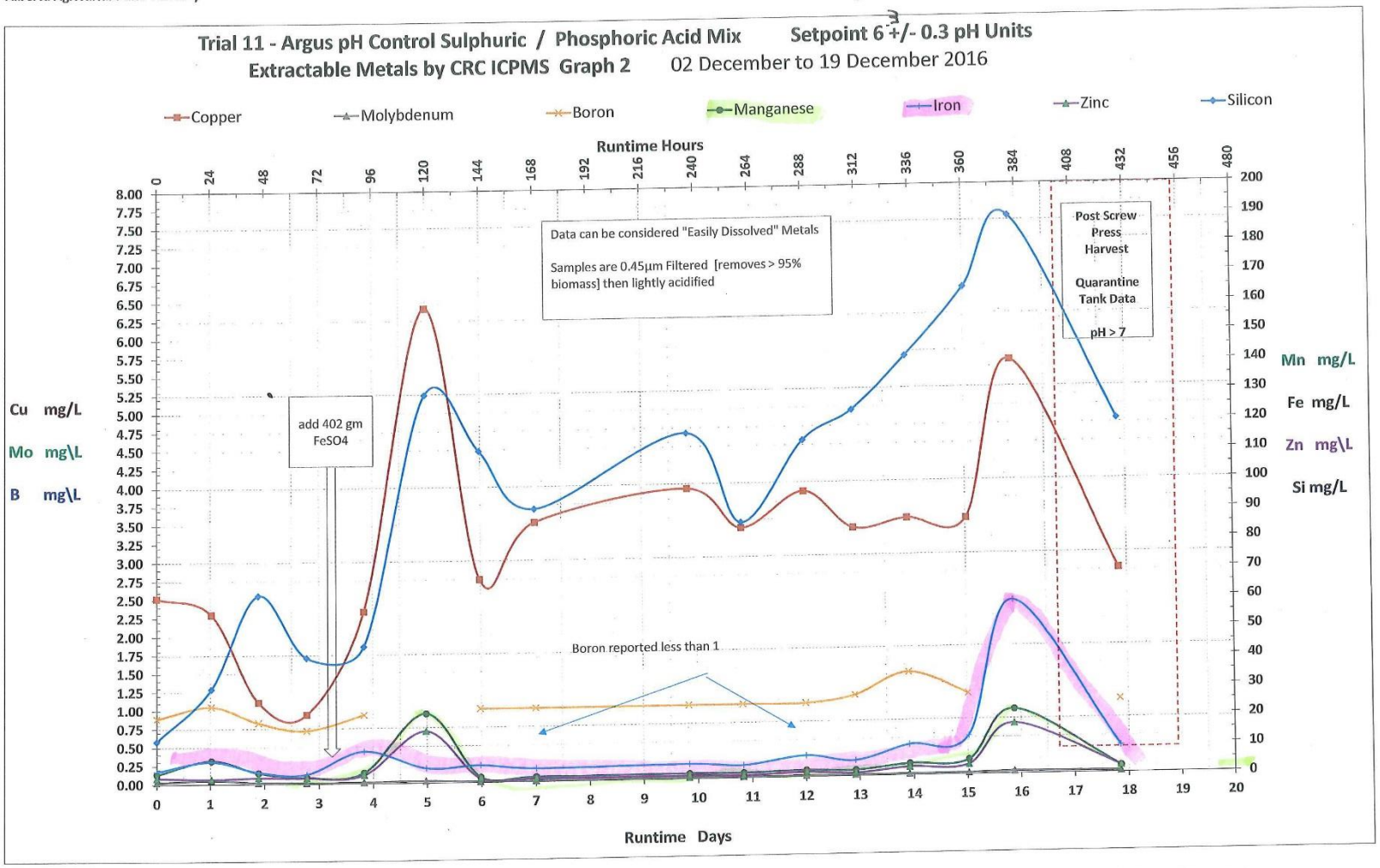
Trial 11 Nutrient Data and Graphs .xlsx Easily Dissolved Metals Graph 1

Alberta Agriculture and Forestry

ACIDF 2015 2016 Project Poultry Manure Digestate

Nutrient Recycling Group



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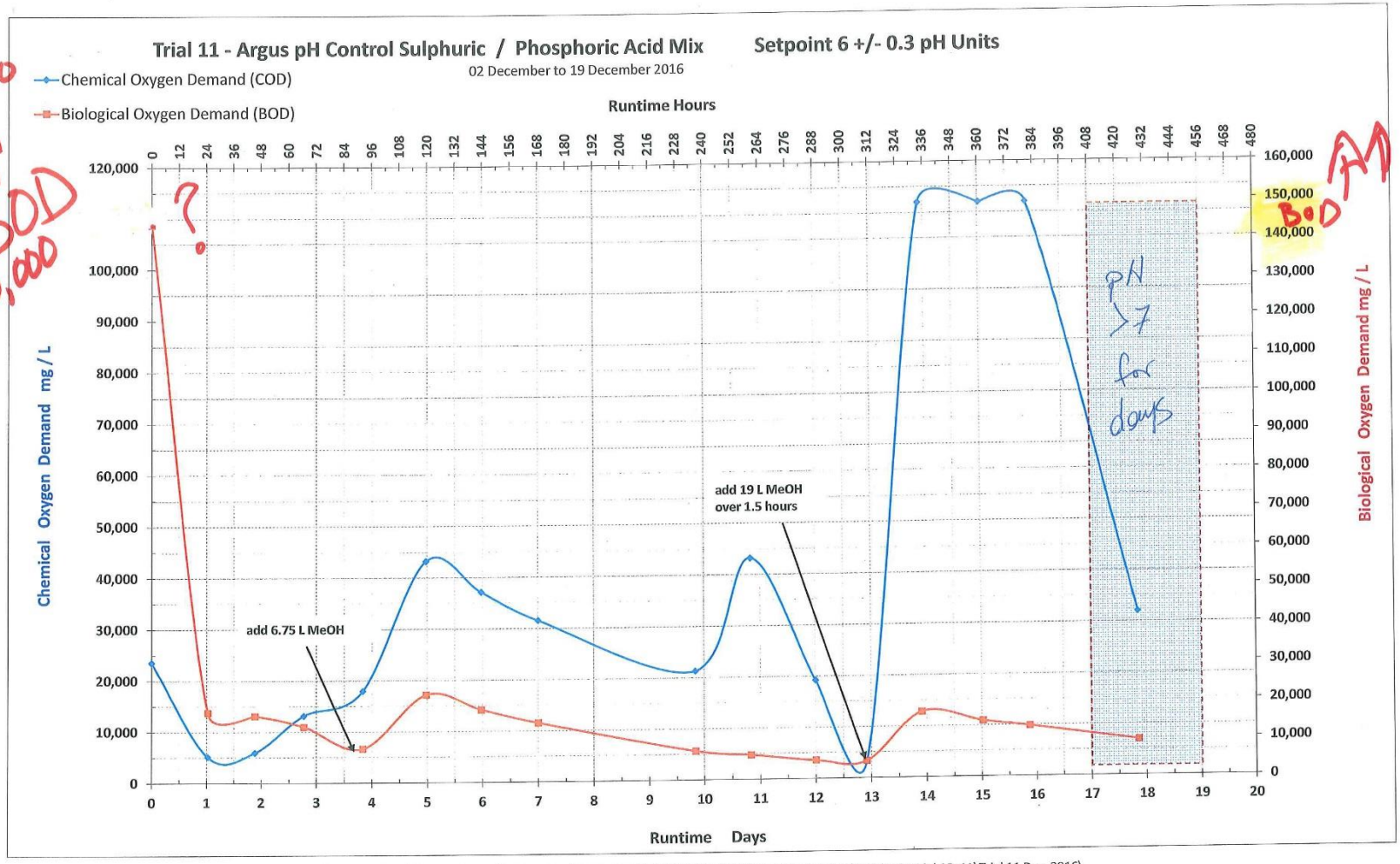

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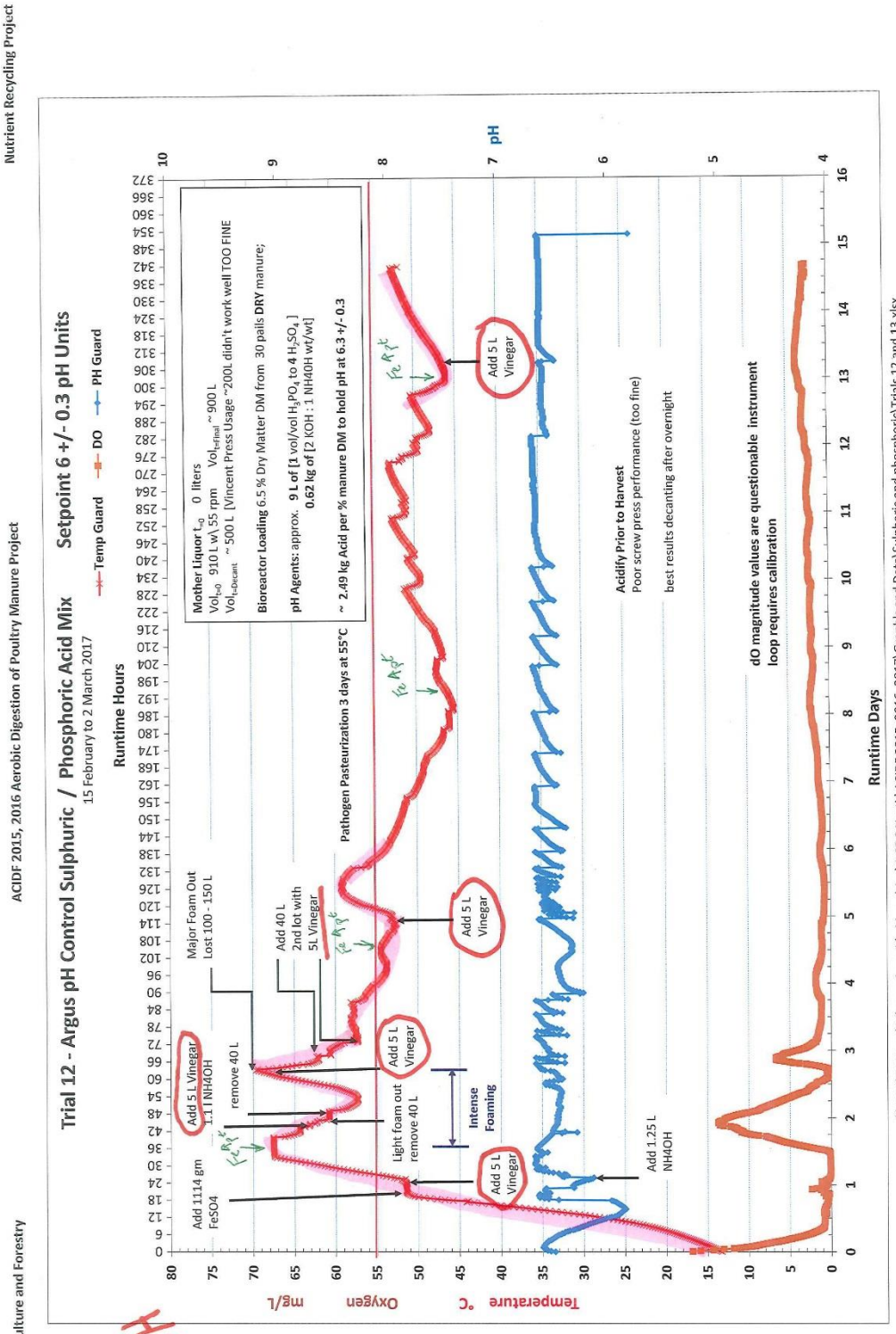
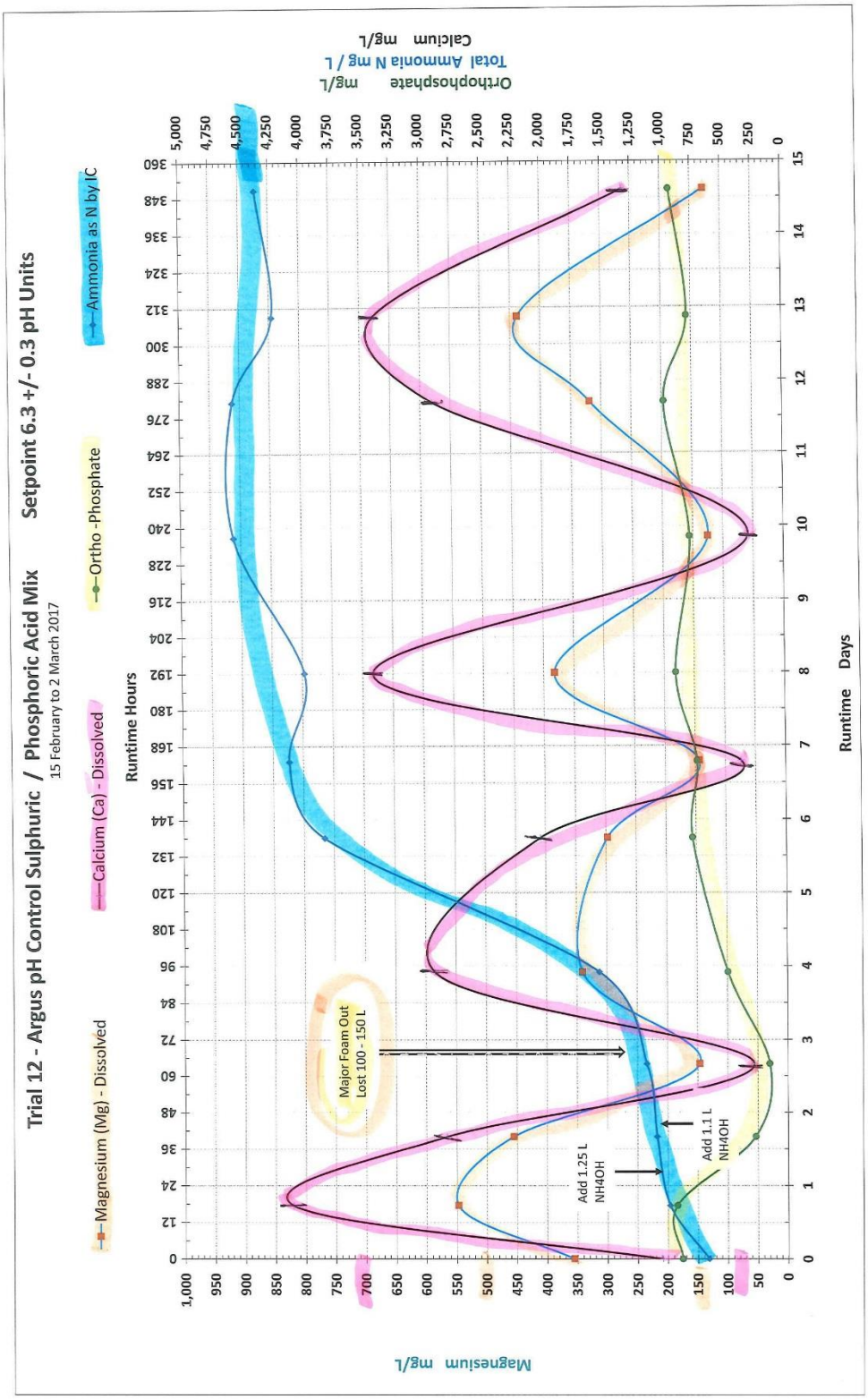
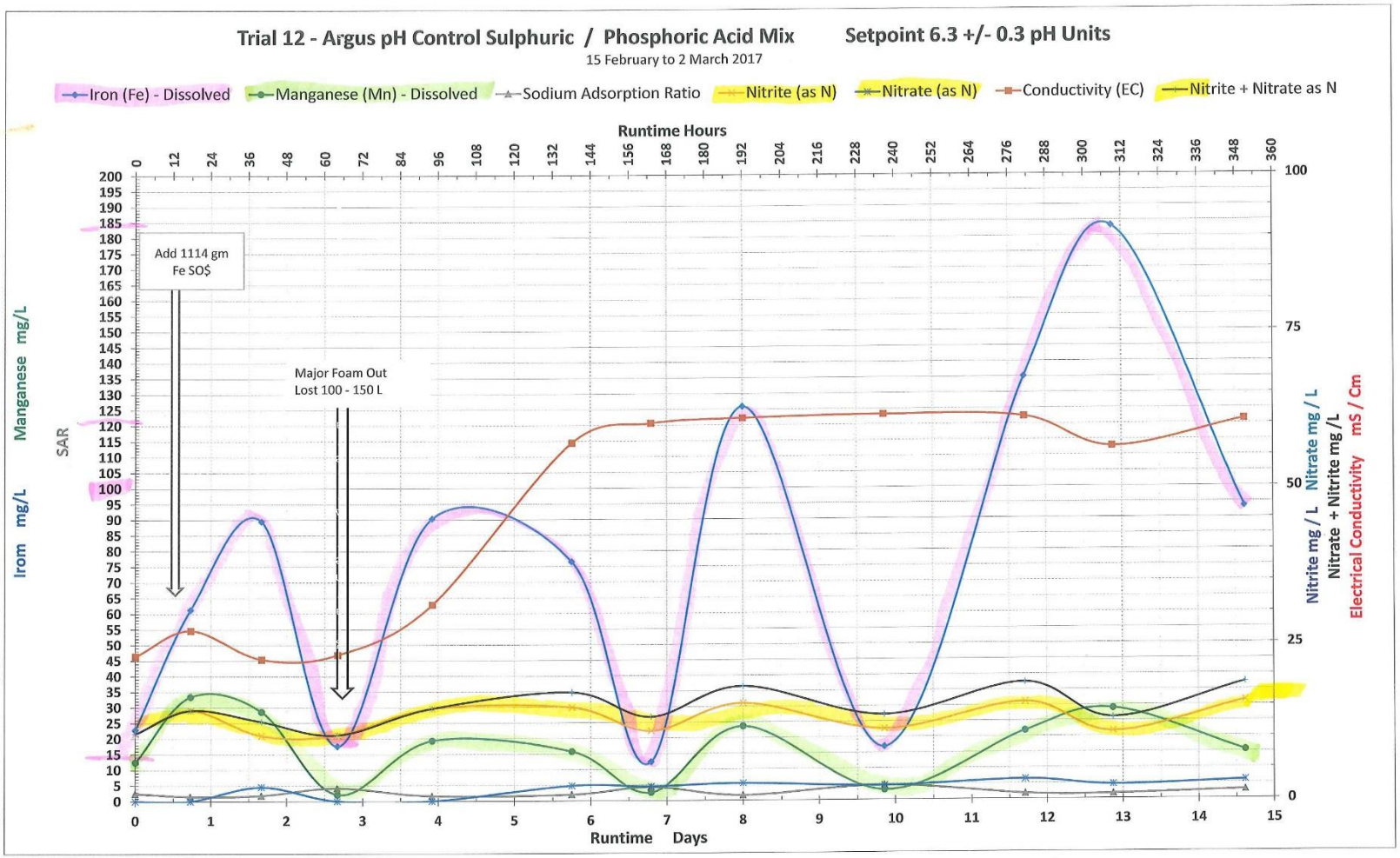
Trial 11 Nutrient Data and Graphs .xlsxChart 4


Alberta Agriculture and Forestry
ACIDF 2015, 2016 Aerobic Digestion of Poultry Manure Project
Nutrient Recycling Project
Trial 12 - Argus pH Control Sulphuric / Phosphoric Acid Mix
Setpoint 6 +/- 0.3 pH Units
15 February to 2 March 2017
Temp Guard
DO
PH Guard
Runtime Hours
Runtime Days
Oxygen mg/L
Temperature °C
pH
Mother Liquor $t_{=0}$ 0 liters
$Vol_{t=0}$ 910 L w\ 55 rpm $Vol_{t=final}$ ~ 900 L
$Vol_{t=Decant}$ ~ 500 L [Vincent Press Usage ~200L didn't work well TOO FINE
Bioreactor Loading 6.5 % Dry Matter DM from 30 pails DRY manure;
pH Agents: approx. 9 L of [1 vol/vol H_3PO_4 to 4 H_2SO_4]
0.62 kg of [2 KOH : 1 NH4OH wt/wt]
~ 2.49 kg Acid per % manure DM to hold pH at 6.3 +/- 0.3
Pathogen Pasteurization 3 days at 55°C
Add 1114 gm FeSO4
Add 5 L Vinegar 1:1 NH4OH
Major Foam Out Lost 100 - 150 L
remove 40 L
Add 40 L 2nd lot with 5L Vinegar
Add 5 L Vinegar
Light foam out remove 40 L
Add 5 L Vinegar
Intense Foaming
Add 5 L Vinegar
Add 5 L Vinegar
Add 1.25 L NH4OH
Acidify Prior to Harvest
Poor screw press performance (too fine)
best results decanting after overnight
dO magnitude values are questionable instrument loop requires calibration
\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015, 2016, 2017\Graph's and Data\Sulpheric and phosphoric\Trials 12 and 13.xlsx
Graph Runtime Trial 12


Trial 12 - Argus pH Control Sulphuric / Phosphoric Acid Mix
15 February to 2 March 2017
Setpoint 6.3 +/- 0.3 pH Units
Magnesium (Mg) - Dissolved
Calcium (Ca) - Dissolved
Ortho -Phosphate
Ammonia as N by IC
Runtime Hours
Runtime Days
Magnesium mg/L
Orthophosphate mg/L
Total Ammonia N mg / L
Calcium mg/L
Major Foam Out
Lost 100 - 150 L
Add 1.25 L
NH4OH
Add 1.1 L
NH4OH



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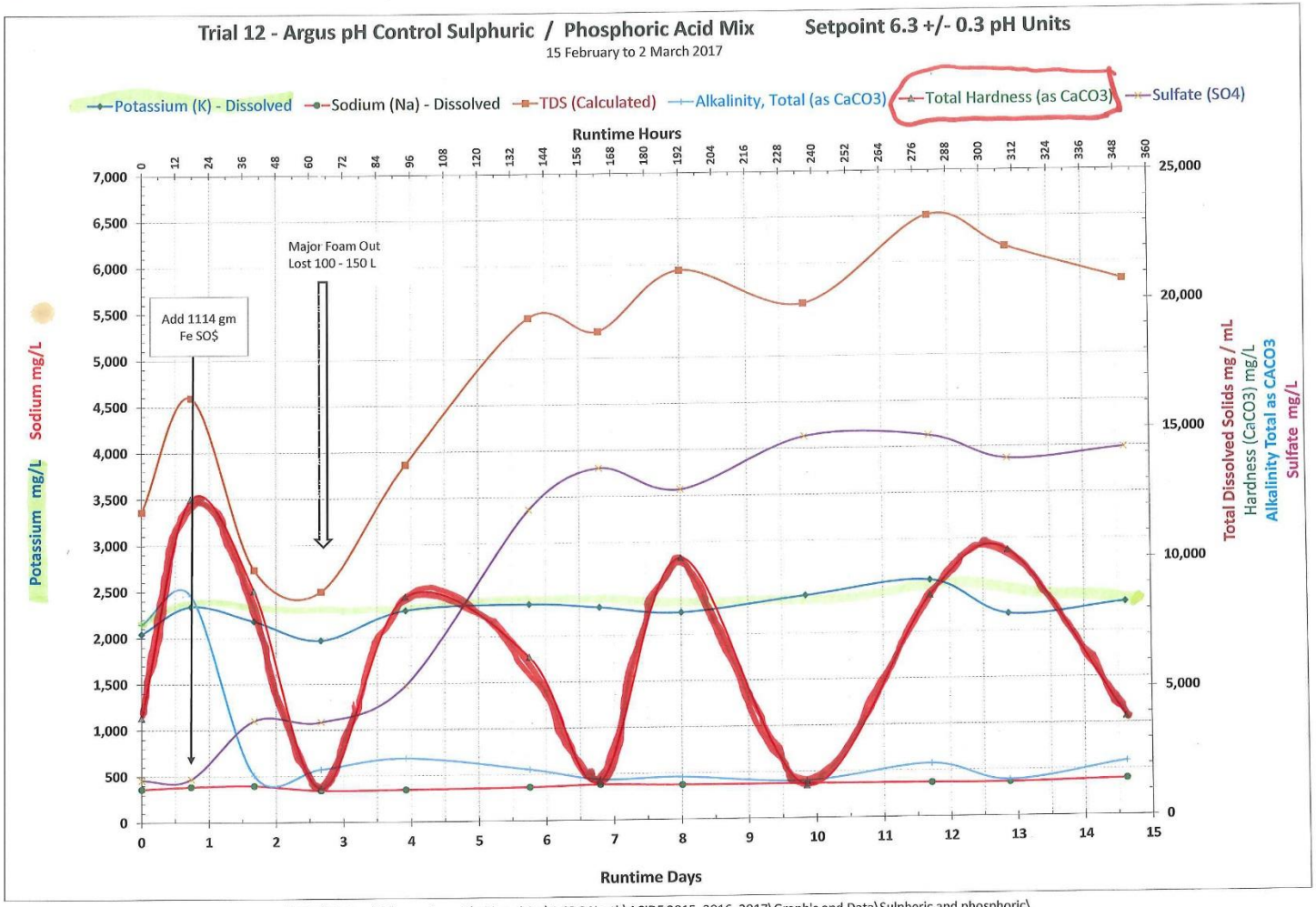
Trials 12 runtime and nutrient.xlsxChart 2



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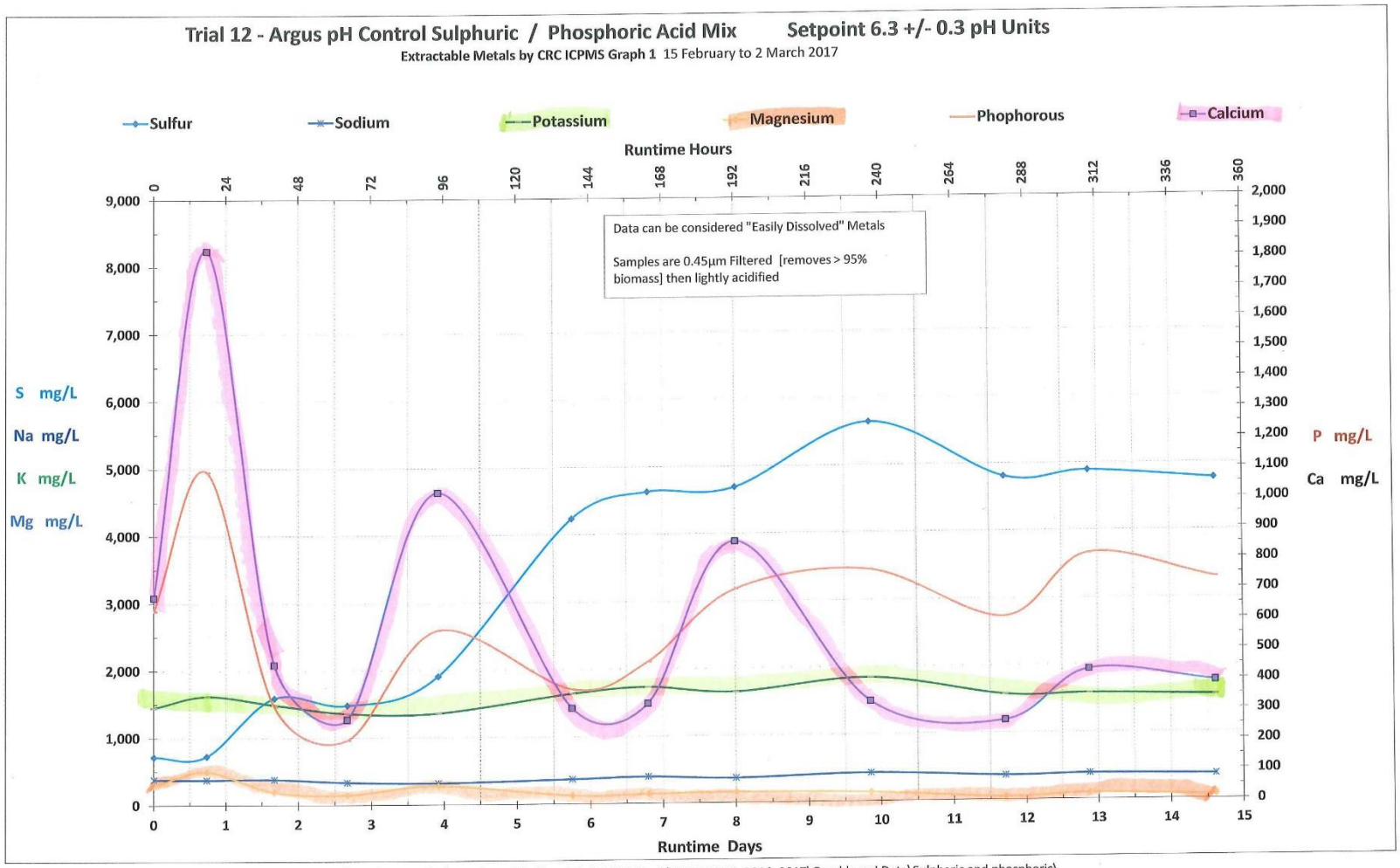
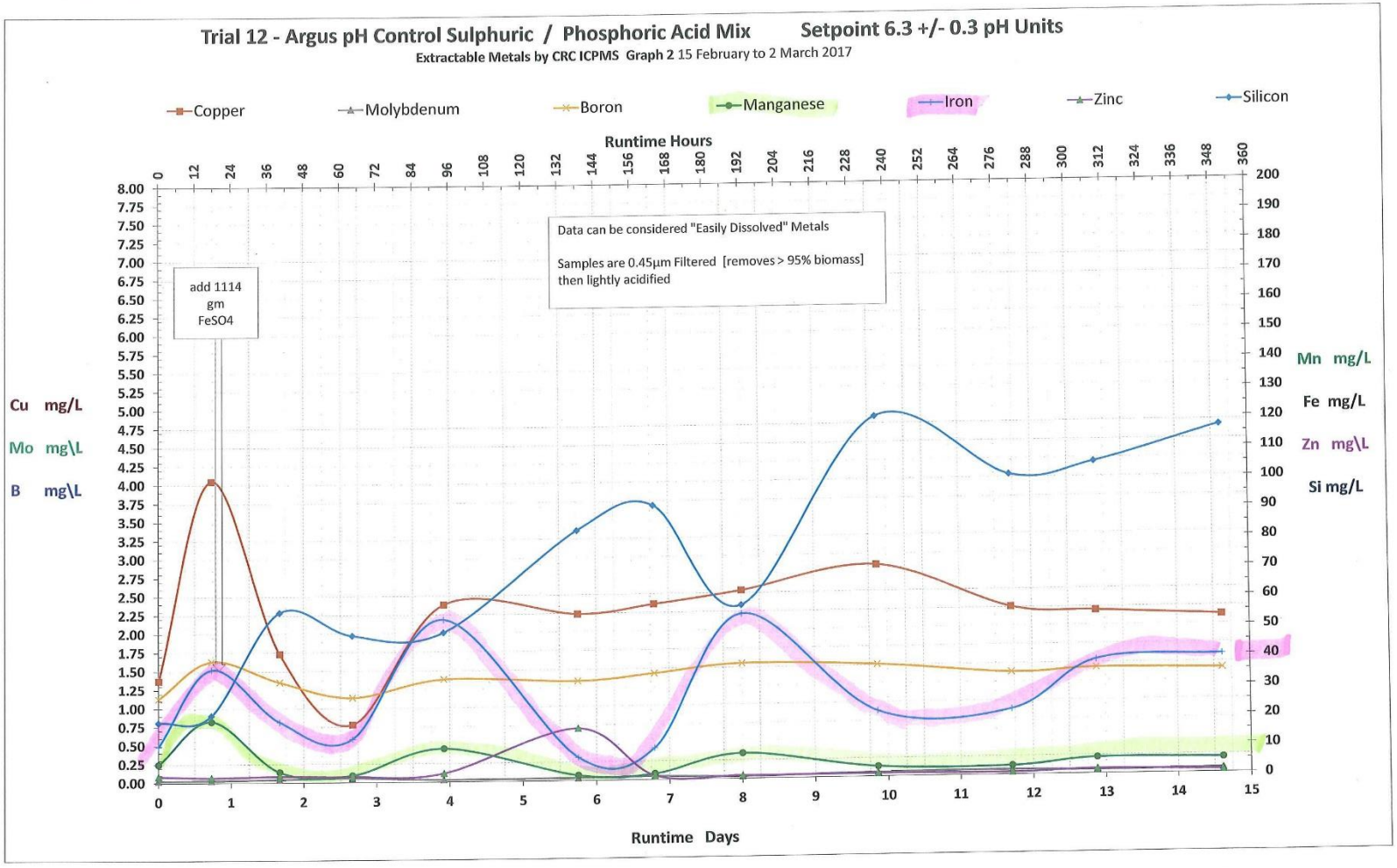
Trials 12 runtime and nutrient.xlsxChart 1



\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015, 2016, 2017\Graph's and Data\Sulpheric and phosphoric\
Trials 12 runtime and nutrient.xlsx Easily Dissolved Metals Graph 1



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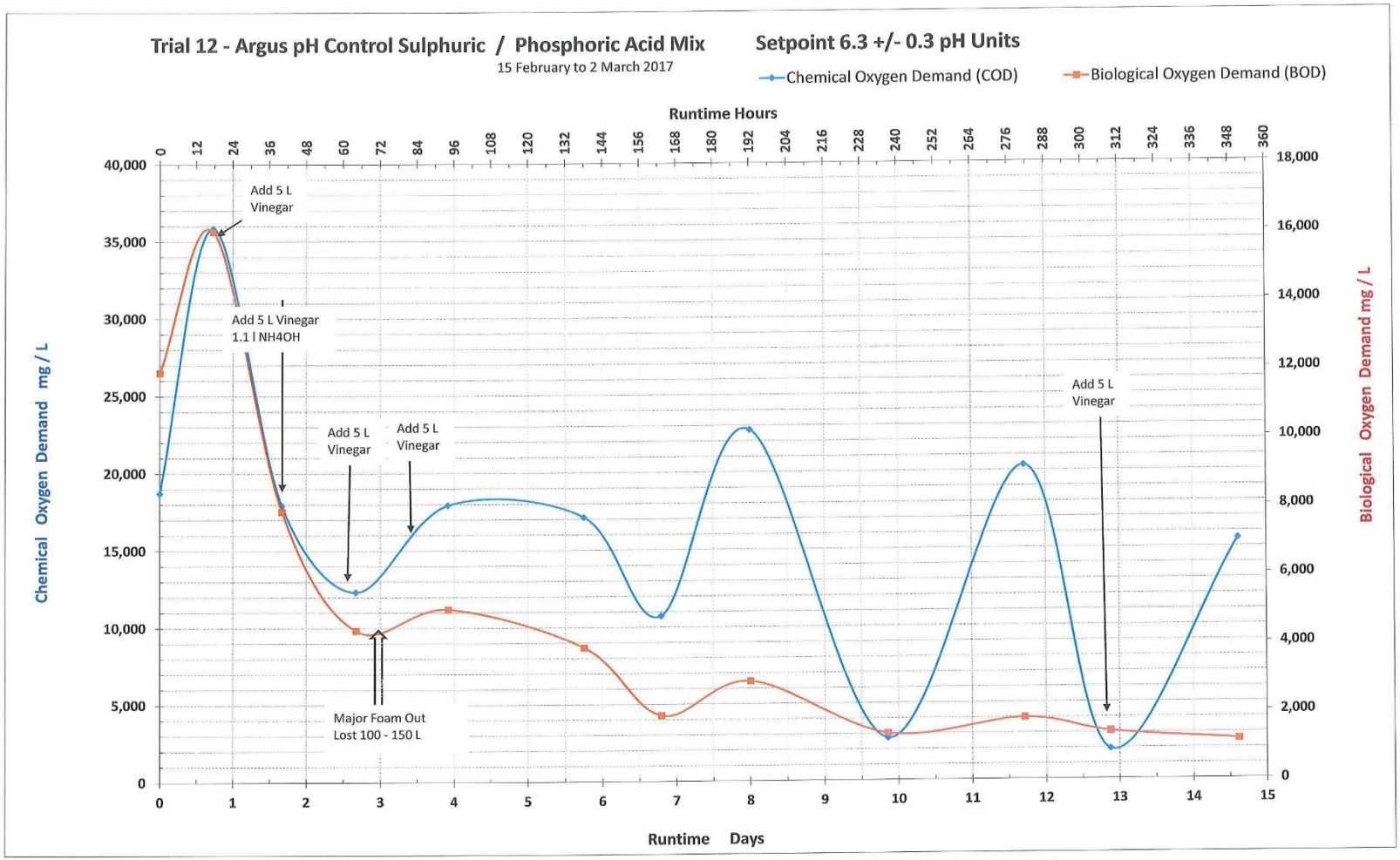

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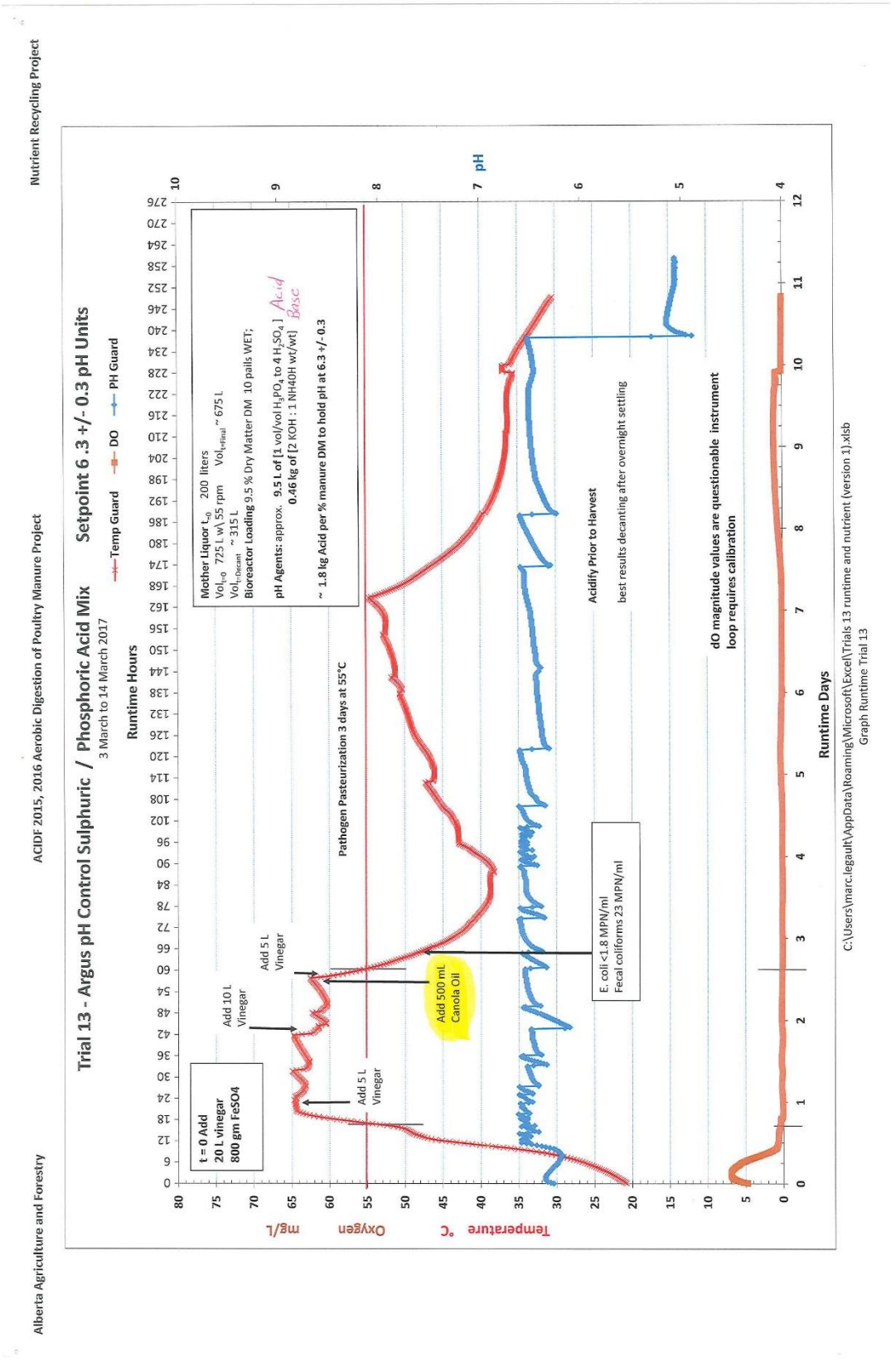
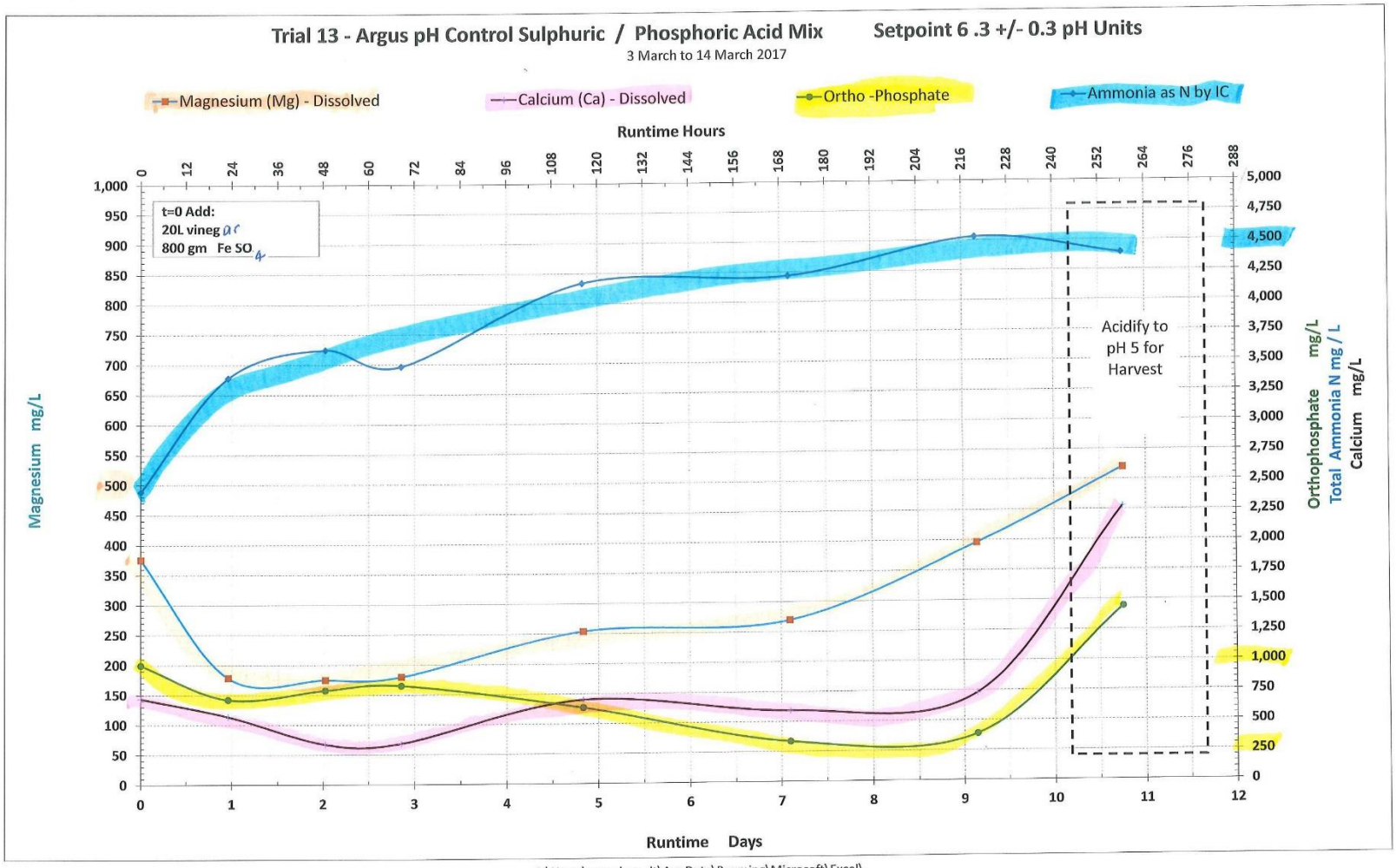
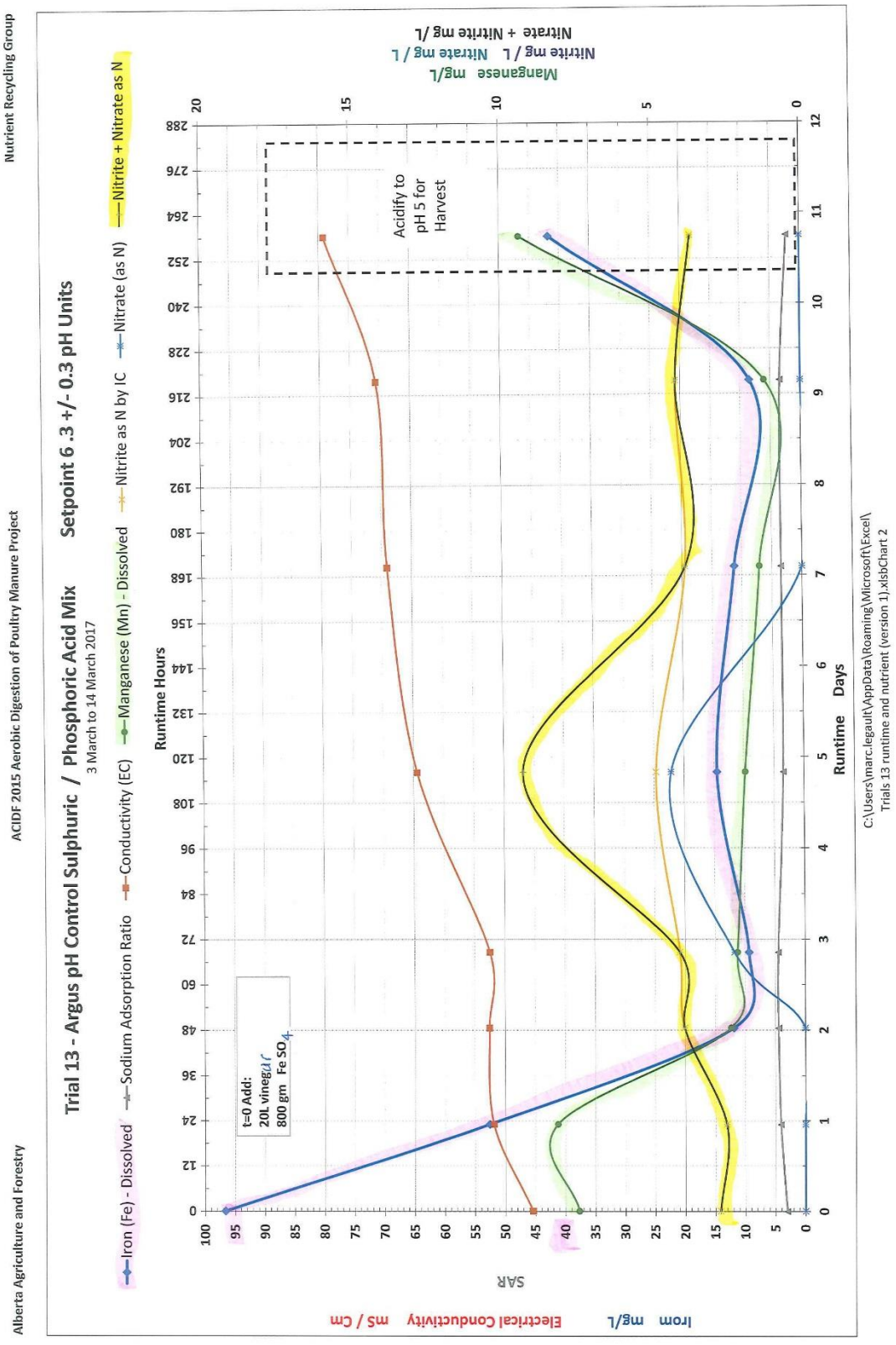
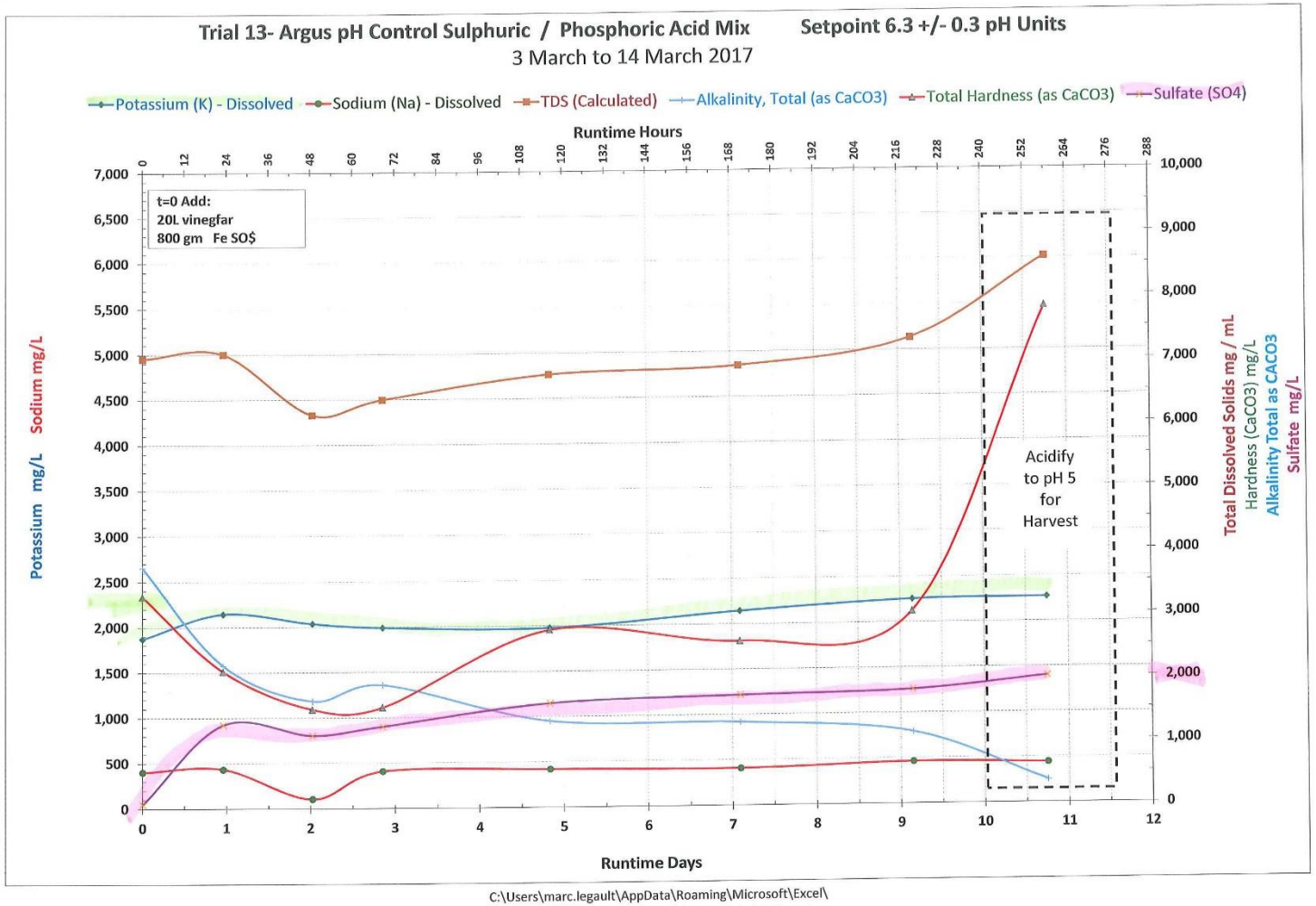
Trials 12 runtime and nutrient.xlsxChart 4

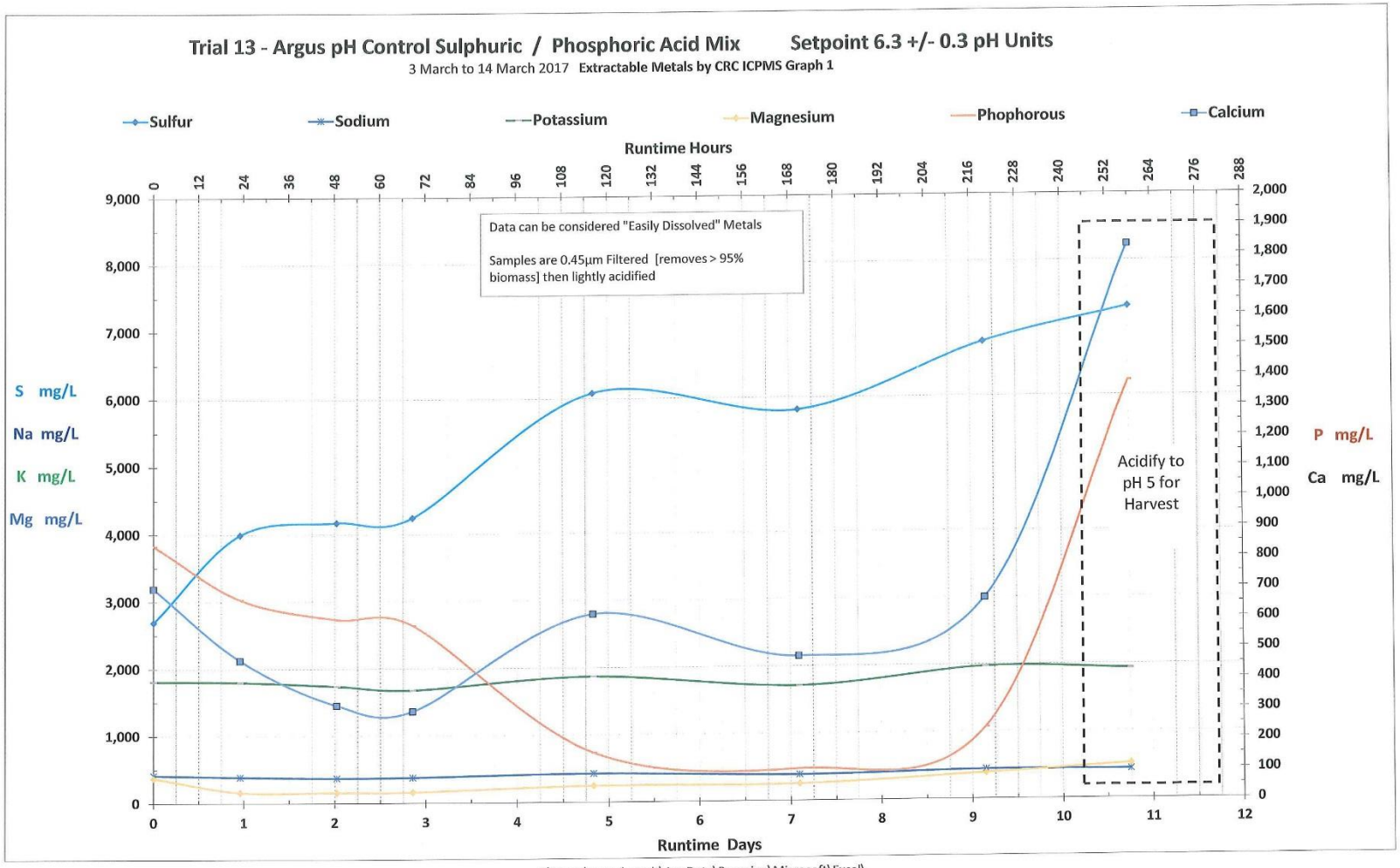

Alberta Agriculture and Forestry
ACIDF 2015, 2016 Aerobic Digestion of Poultry Manure Project
Nutrient Recycling Project
Trial 13 - Argus pH Control Sulphuric / Phosphoric Acid Mix
Setpoint 6 .3 +/- 0.3 pH Units
3 March to 14 March 2017
Temp Guard
DO
PH Guard
Runtime Hours
Runtime Days
Temperature °C
Oxygen mg/L
pH
t = 0 Add
20 L vinegar
800 gm FeSO4
Add 10 L Vinegar
Add 5 L Vinegar
Add 5 L Vinegar
Add 500 mL Canola Oil
Pathogen Pasteurization 3 days at 55°C
E. coli <1.8 MPN/ml
Fecal coliforms 23 MPN/ml
Mother Liquor t=0 200 liters
Vol t=0 725 L w\ 55 rpm Vol t=final ~ 675 L
Vol t=Decant ~ 315 L
Bioreactor Loading 9.5 % Dry Matter DM 10 pails WET;
pH Agents: approx. 9.5 L of [1 vol/vol H3PO4 to 4 H2SO4]
0.46 kg of [2 KOH : 1 NH4OH wt/wt]
~ 1.8 kg Acid per % manure DM to hold pH at 6.3 +/- 0.3
Acidify Prior to Harvest
best results decanting after overnight settling
dO magnitude values are questionable instrument
loop requires calibration
C:\Users\marc.legault\AppData\Roaming\Microsoft\Excel\Trials 13 runtime and nutrient (version 1).xlsb
Graph Runtime Trial 13


Trial 13 - Argus pH Control Sulphuric / Phosphoric Acid Mix
Setpoint 6 .3 +/- 0.3 pH Units
3 March to 14 March 2017
Magnesium (Mg) - Dissolved
Calcium (Ca) - Dissolved
Ortho -Phosphate
Ammonia as N by IC
Runtime Hours
t=0 Add:
20L vineg ar
800 gm Fe SO4
Acidify to pH 5 for Harvest
Magnesium mg/L
Orthophosphate mg/L
Total Ammonia N mg / L
Calcium mg/L
Runtime Days
C:\Users\marc.legault\AppData\Roaming\Microsoft\Excel\
Trials 13 runtime and nutrient (version 1).xlsbChart 3

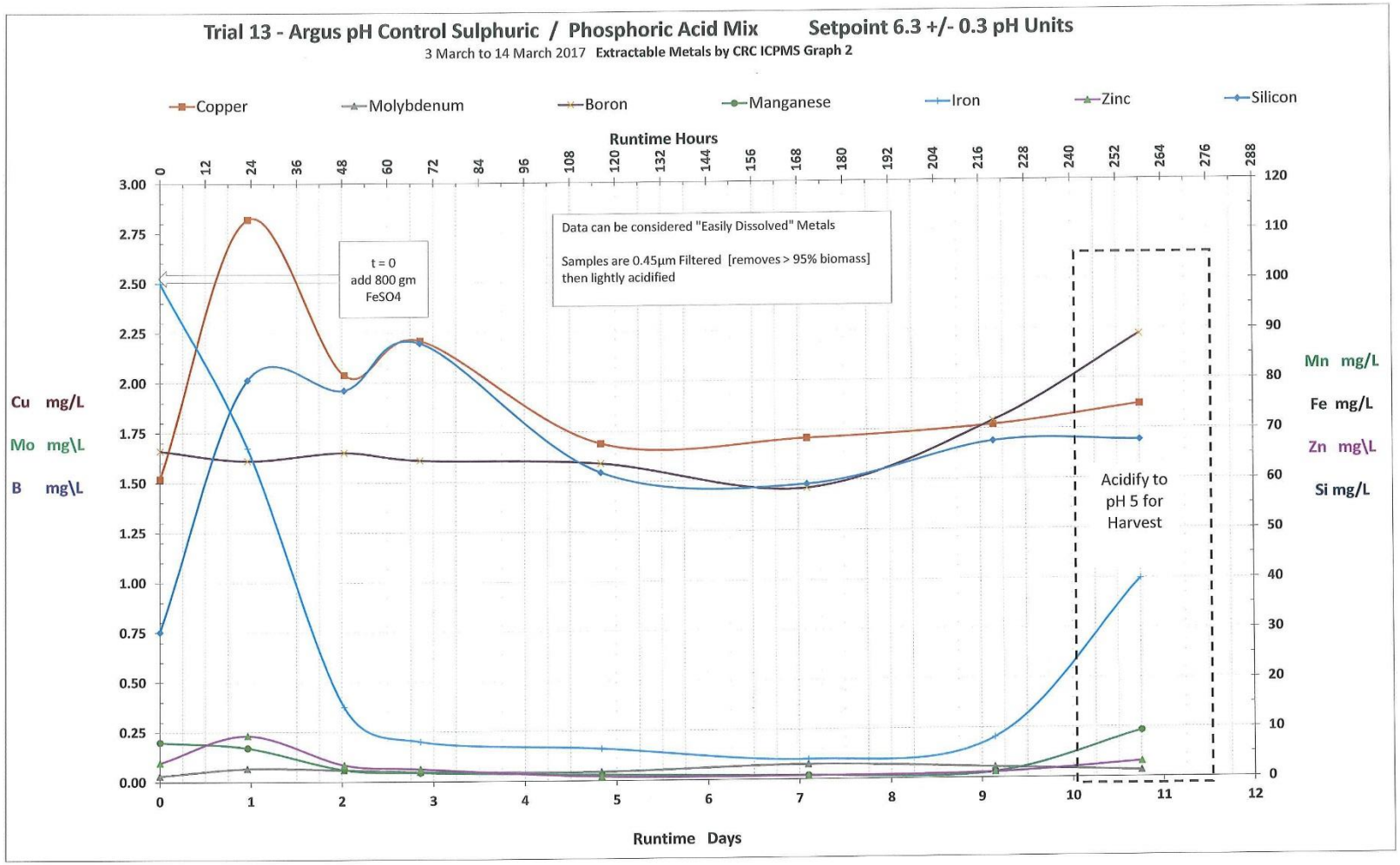

Alberta Agriculture and Forestry
ACIDF 2015 Aerobic Digestion of Poultry Manure Project
Nutrient Recycling Group
Trial 13 - Argus pH Control Sulphuric / Phosphoric Acid Mix
Setpoint 6 .3 +/- 0.3 pH Units
3 March to 14 March 2017
Iron (Fe) - Dissolved
Sodium Adsorption Ratio
Conductivity (EC)
Manganese (Mn) - Dissolved
Nitrite as N by IC
Nitrate (as N)
Nitrite + Nitrate as N
Runtime Hours
Runtime Days
t=0 Add:
20L vinegar
800 gm Fe SO4
Acidify to
pH 5 for
Harvest
Iron mg/L
Electrical Conductivity mS / Cm
SAR
Manganese mg/L
Nitrite mg / L Nitrate mg / L
Nitrate + Nitrite mg /L
C:\Users\marc.legault\AppData\Roaming\Microsoft\Excel\
Trials 13 runtime and nutrient (version 1).xlsbChart 2



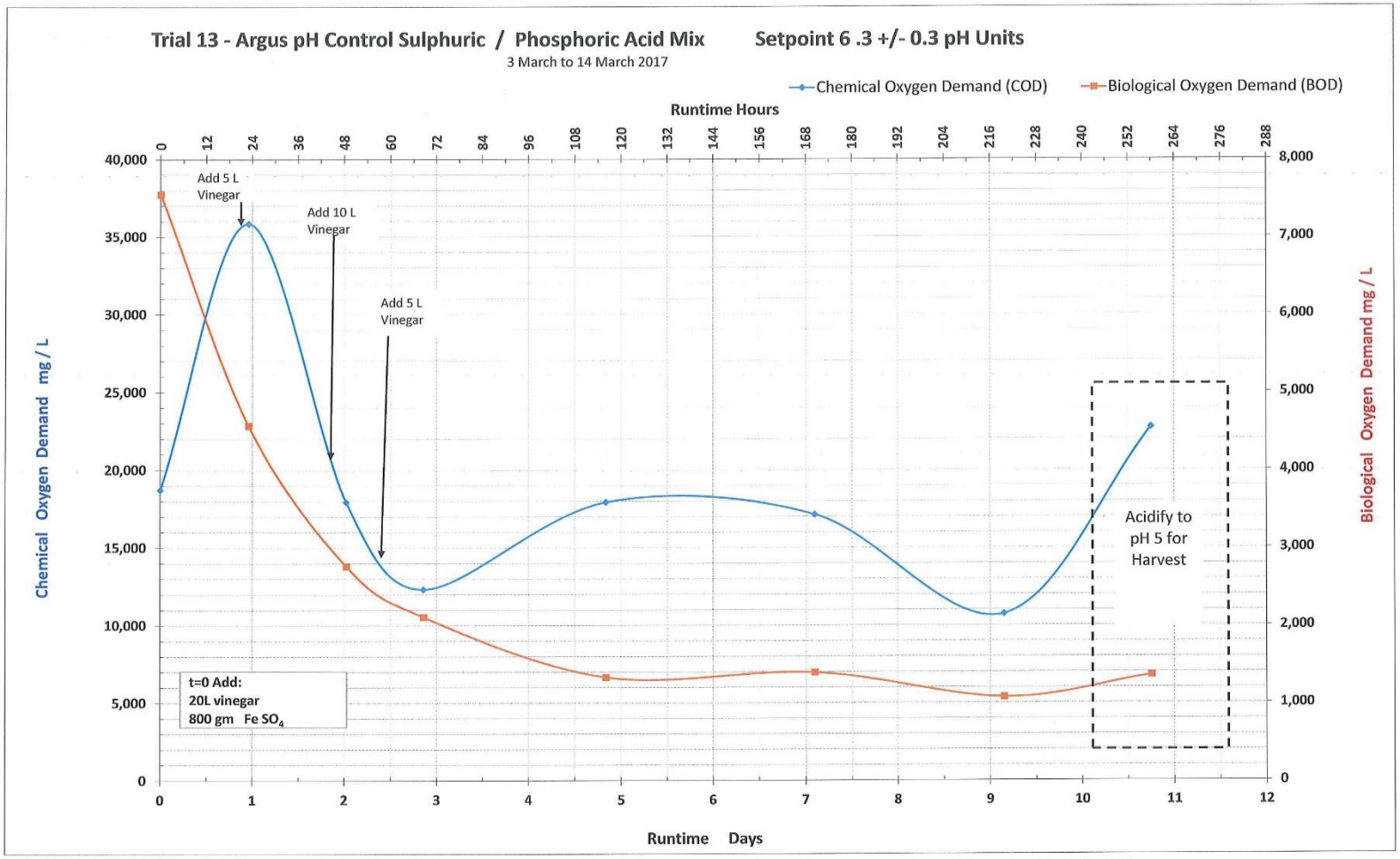
C:\Users\marc.legault\AppData\Roaming\Microsoft\Excel\
Trials 13 runtime and nutrient (version 1).xlsbChart 1



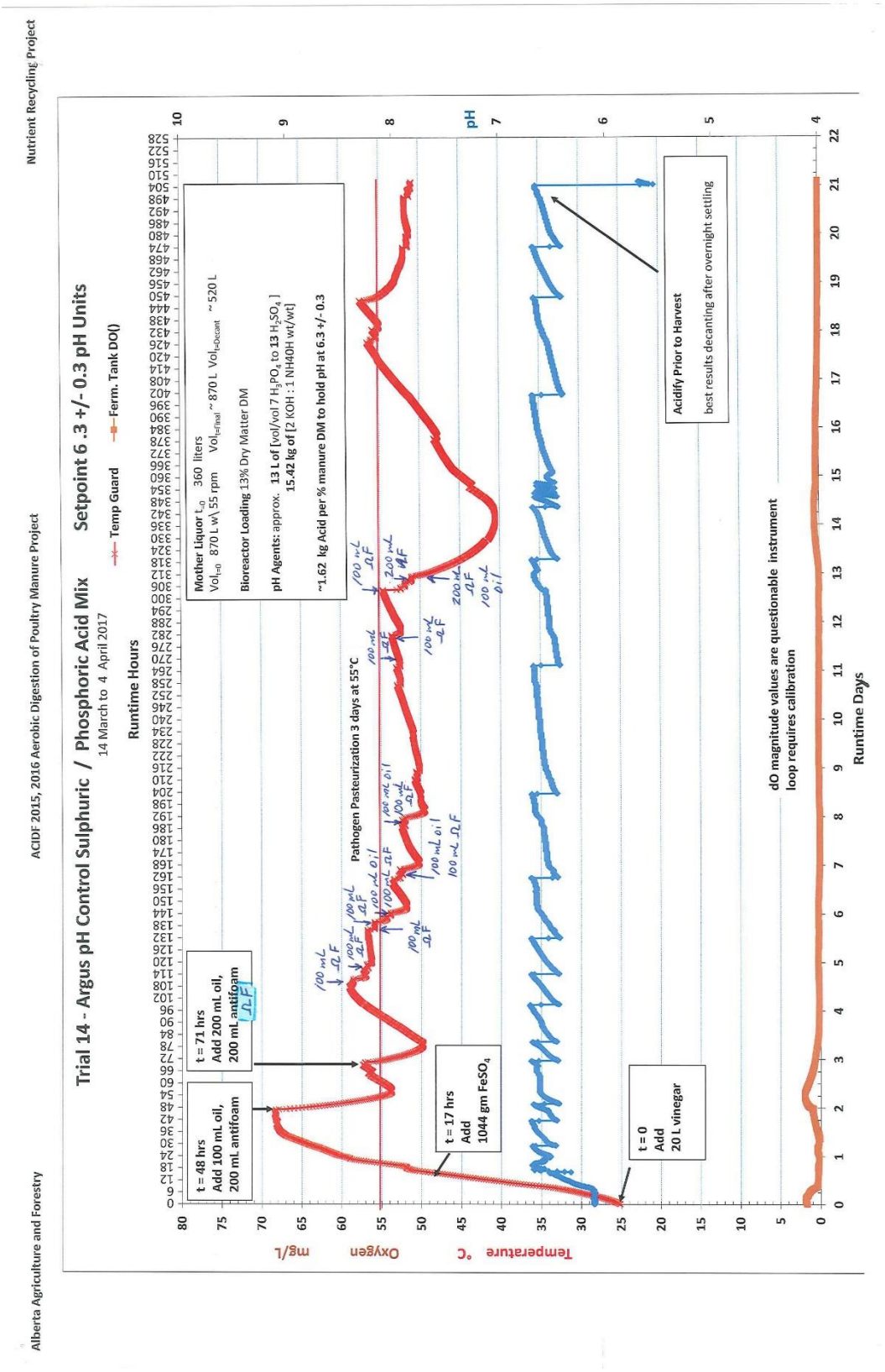
C:\Users\marc.legault\AppData\Roaming\Microsoft\Excel\
Trials 13 runtime and nutrient (version 1).xlsb Easily Dissolved Metals Graph 1

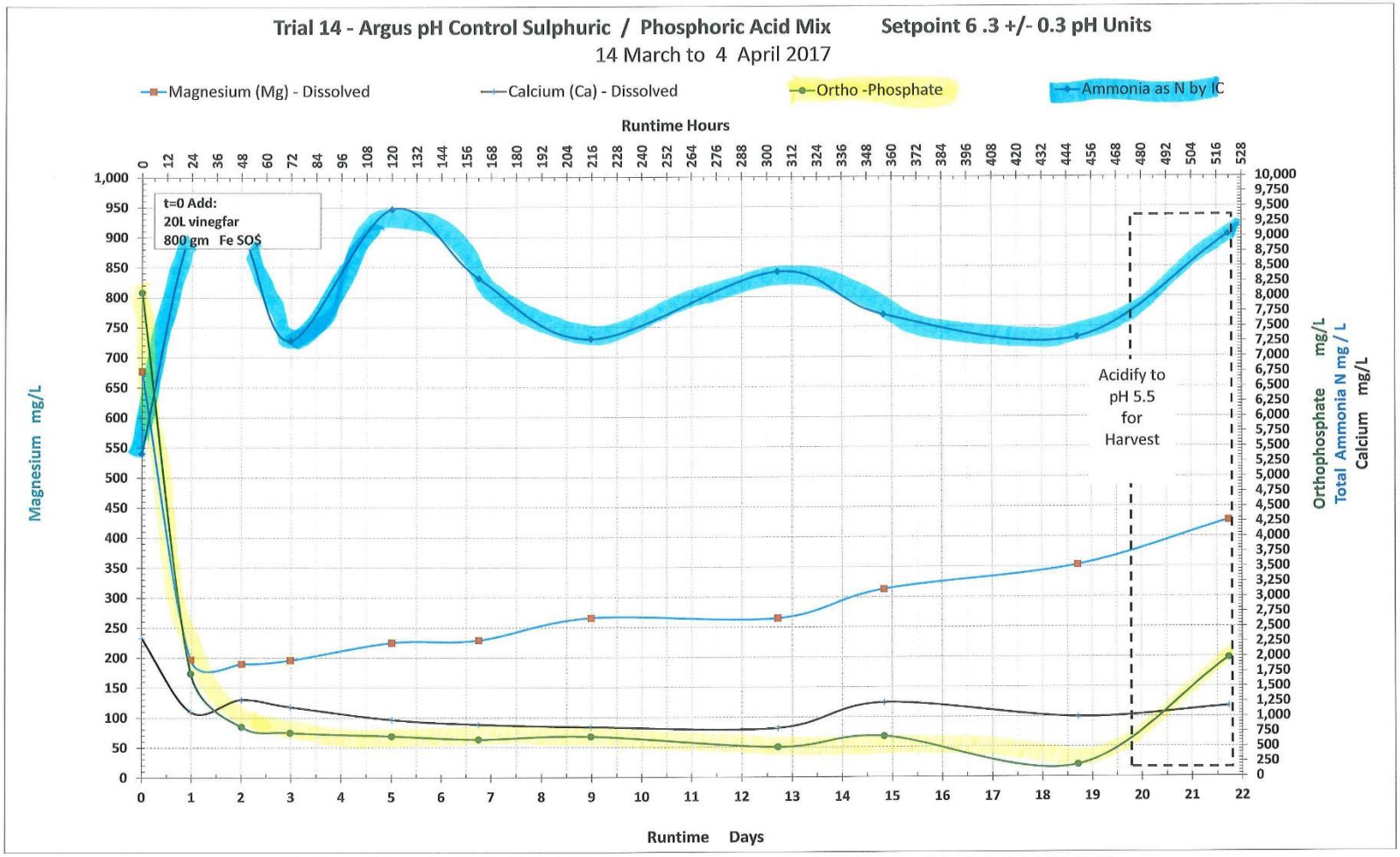


C:\Users\marc.legault\AppData\Roaming\Microsoft\Excel\Trials 13 runtime and nutrient (version 1).xlsb Easily Dissolved Metals Graph 2

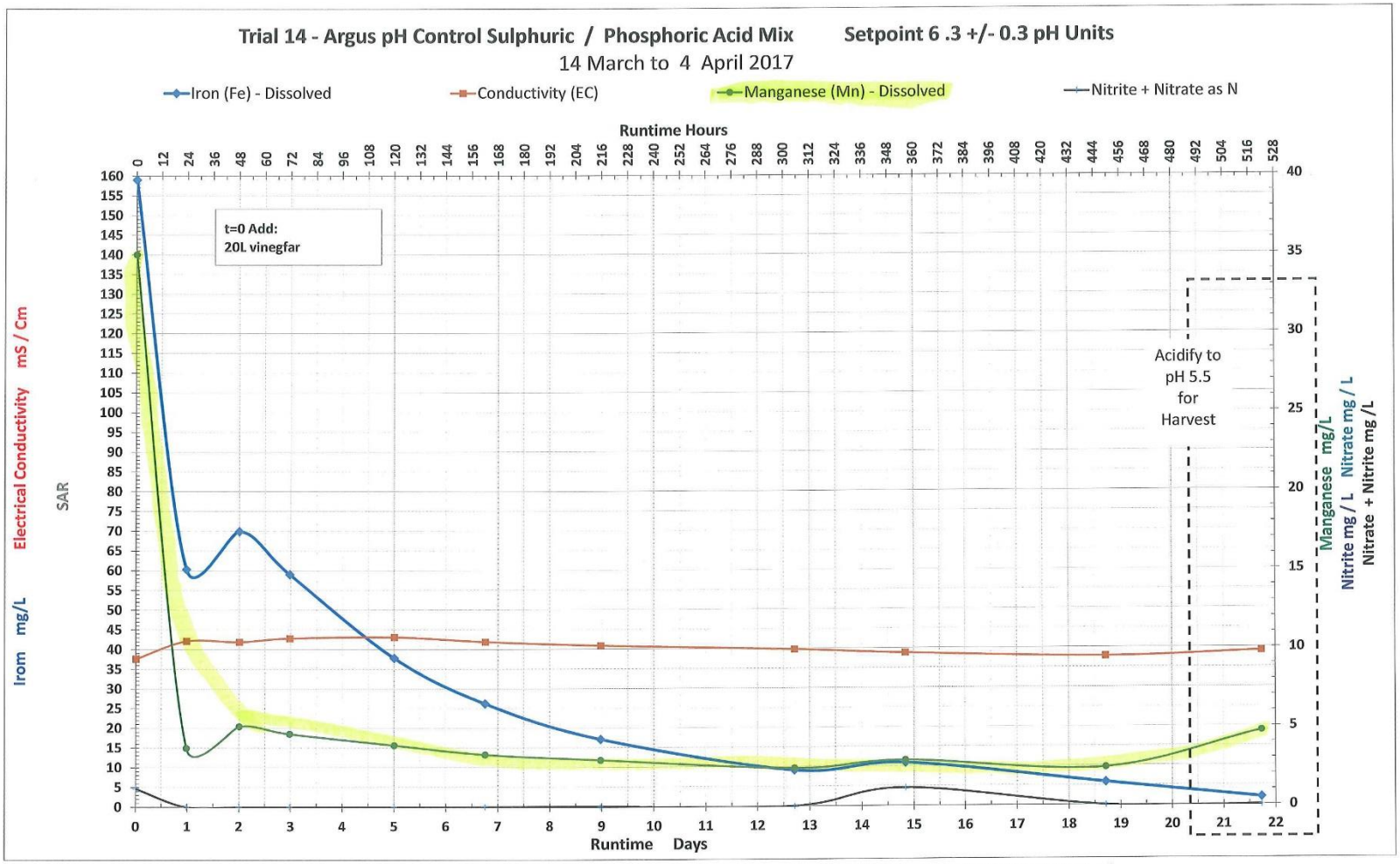


\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015, 2016, 2017\Graph's and Data\Sulpheric and phosphoric Trials 12, 13, 14, 15\Trial 13\
Trials 13 runtime and nutrient.xlsxChart 4

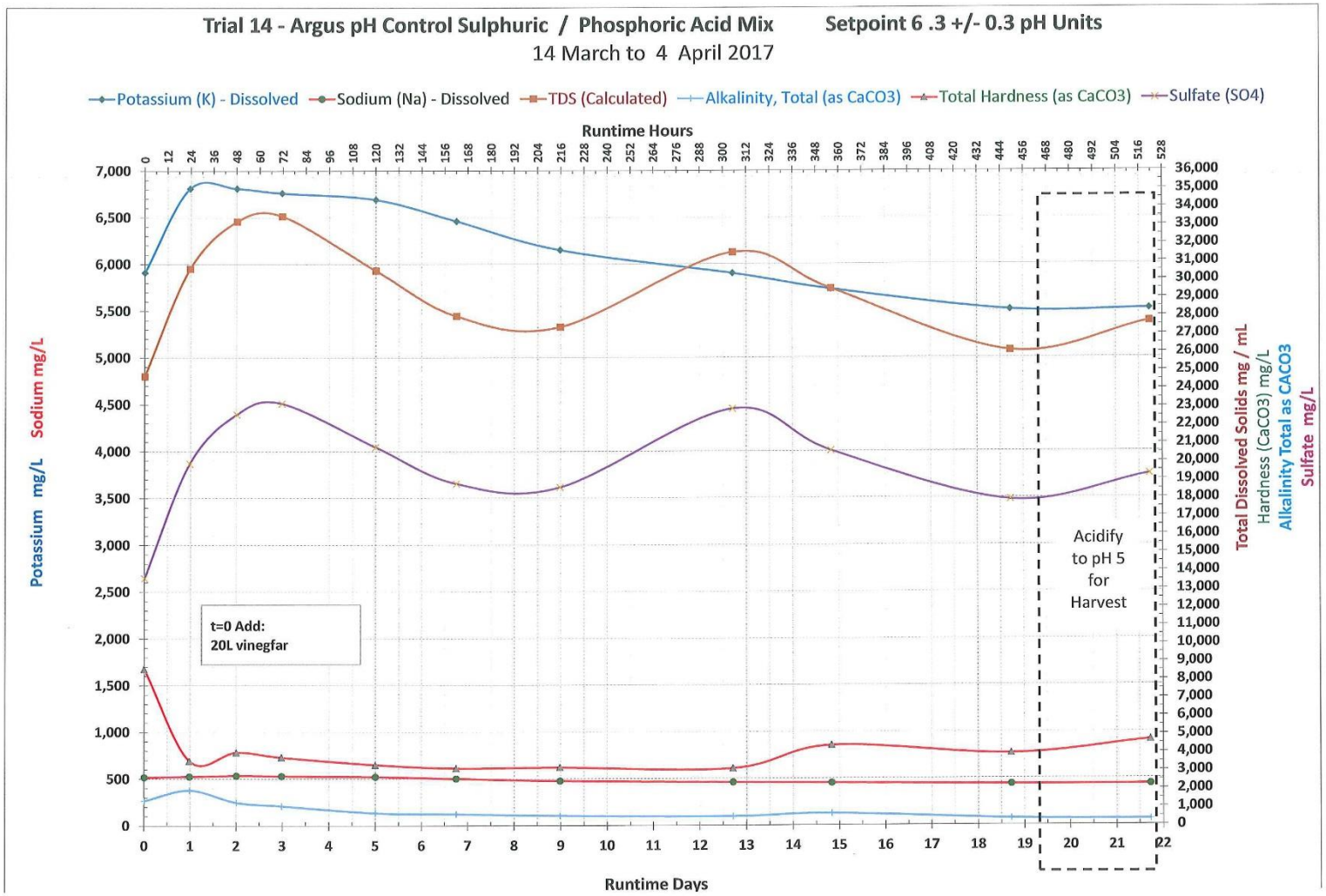

Alberta Agriculture and Forestry
ACIDF 2015, 2016 Aerobic Digestion of Poultry Manure Project
Nutrient Recycling Project
Trial 14 - Argus pH Control Sulphuric / Phosphoric Acid Mix
Setpoint 6 .3 +/- 0.3 pH Units
14 March to 4 April 2017
Temp Guard
Ferm. Tank DO()
Runtime Hours
Runtime Days
Oxygen mg/L
Temperature °C
pH
Mother Liquor t_{-0} 360 liters
$Vol_{t=0}$ 870 L w\ 55 rpm $Vol_{t=final}$ ~ 870 L $Vol_{t=Decant}$ ~ 520 L
Bioreactor Loading 13% Dry Matter DM
pH Agents: approx. 13 L of [vol/vol 7 H_3PO_4 to 13 H_2SO_4]
15.42 kg of [2 KOH : 1 NH4OH wt/wt]
~1.62 kg Acid per % manure DM to hold pH at 6.3 +/- 0.3
t = 48 hrs
Add 100 mL oil,
200 mL antifoam
t = 71 hrs
Add 200 mL oil,
200 mL antifoam
t = 17 hrs
Add
1044 gm $FeSO_4$
t = 0
Add
20 L vinegar
Pathogen Pasteurization 3 days at 55°C
Acidify Prior to Harvest
best results decanting after overnight settling
dO magnitude values are questionable instrument
loop requires calibration

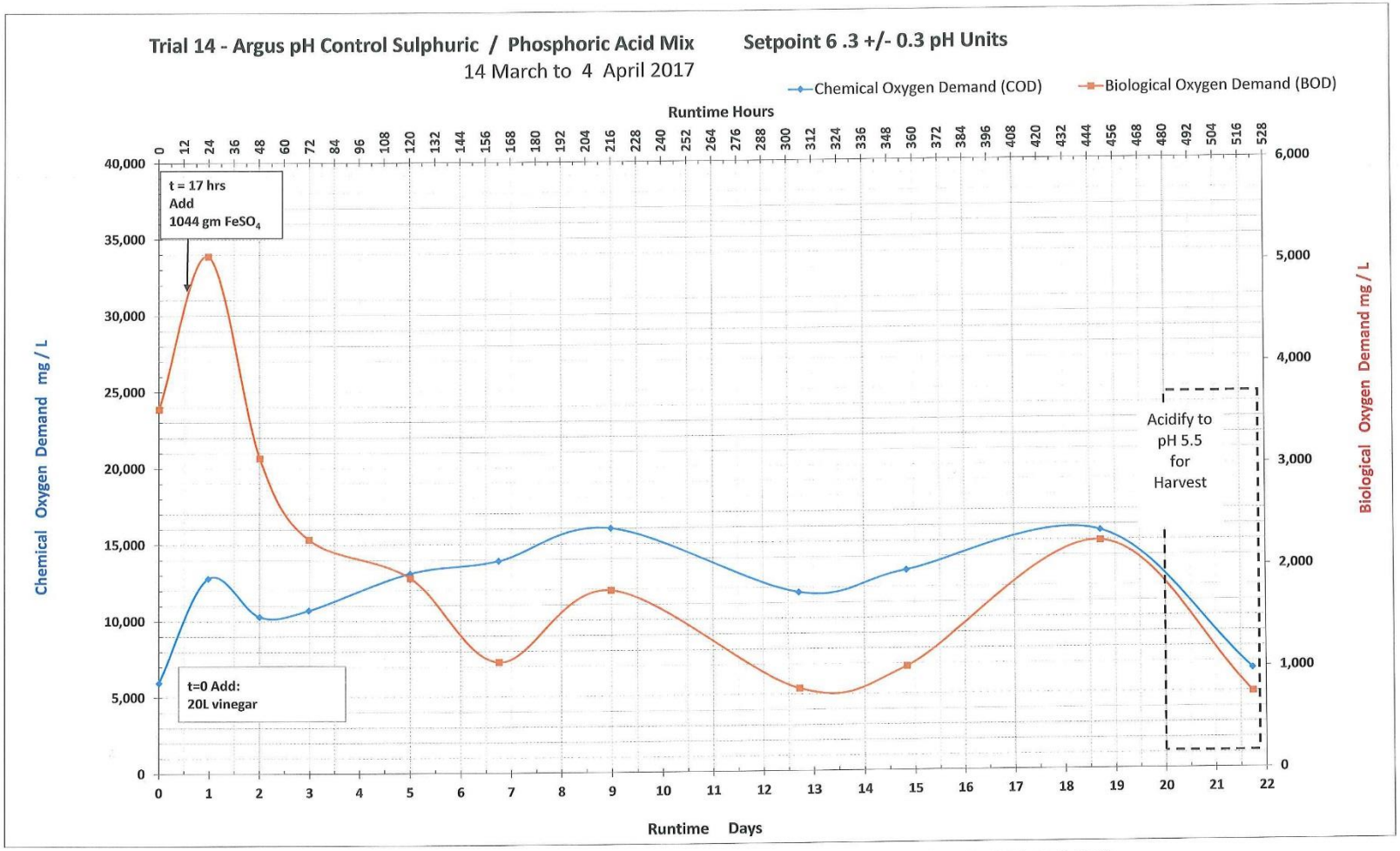


\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015, 2016, 2017\Graph's and Data\Sulpheric and phosphoric Trials 12, 13, 14, 15\
Trial 14 Data.xlsxChart 3



\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015, 2016, 2017\Graph's and Data\Sulpheric and phosphoric Trials 12, 13, 14, 15\
Trial 14 Data.xlsxChart 2


Trial 14 - Argus pH Control Sulphuric / Phosphoric Acid Mix
Setpoint 6 .3 +/- 0.3 pH Units
14 March to 4 April 2017
Potassium (K) - Dissolved
Sodium (Na) - Dissolved
TDS (Calculated)
Alkalinity, Total (as CaCO3)
Total Hardness (as CaCO3)
Sulfate (SO4)
Runtime Hours
Runtime Days
Potassium mg/L
Sodium mg/L
Total Dissolved Solids mg / mL
Hardness (CaCO3) mg/L
Alkalinity Total as CACO3
Sulfate mg/L
t=0 Add:
20L vinegfar
Acidify
to pH 5
for
Harvest



\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015, 2016, 2017\Graph's and Data\Sulpheric and phosphoric Trials 12, 13, 14, 15\
Trial 14 Data.xlsxChart 4

Trial 13

AAAF 170323-01 (17-WY-08)

pH Control Agents 1 vol/vol $H_3PO_4 : 4 H_2SO_4$ 2 KOH : 1 NH_4OH

PBIR Laboratories Inc.
Call Toll Free: 1-866-450-3957

Certificate of Analysis

Sample ID and Parameters Measured	Result	Detection Limit	Units	Quantity Analyzed	Test Method	*DF	Date Analyzed	Note
PBR ID: 17-WY-08								
Client ID: Trial 13 Sample 8 2017/03/14 8:30								
Matrix: Poultry Manure Digestate								
Chloride by IC								
Chloride (Cl)	831.20	0.50	mg/L	10ml	Prot#1412	10^{-2}	170316	
Nitrate as N by IC								
Nitrate (as N)	<0.1	0.1	mg/L	10ml	Prot#1412	10^{-2}	170316	
Sulfate by IC								
Sulfate (SO4)	1997.80	0.50	mg/L	10ml	Prot#1412	10^{-3}	170317	
Nitrite as N by IC								
Nitrite (as N)	3.65	0.10	mg/L	10ml	Prot#1412	10^{-2}	170316	
Nitrite + Nitrate as N	3.65	0.10	mg/L	NA	Calculation			
Ammonia as N by IC	4391.64							
NH4-N		0.70	mg/L	10ml	Prot#1410	10^{-2}	170321	
Dissolved Metals by ICP-OES								
Calcium (Ca) - Dissolved	2276.40	0.50	mg/L	20ml	Prot#1420	10^{-2}	170316	
Iron (Fe) - Dissolved	41.81	0.05	mg/L	20ml	Prot#1420	10^{-1}	170316	
Magnesium (Mg) - Dissolved	519.76	0.30	mg/L	20ml	Prot#1420	10^{-1}	170316	
Manganese (Mn) - Dissolved	9.35	0.01	mg/L	20ml	Prot#1420	10^{-1}	170316	
Potassium (K) - Dissolved	2265.62	0.30	mg/L	20ml	Prot#1420	10^{-2}	170316	
Sodium (Na) - Dissolved	441.41	0.10	mg/L	20ml	Prot#1420	10^{-1}	170316	
Total Hardness (as CaCO3)	7822.01	5	mg/L	NA	calculation	NA		
Extractable Regulated Metals (Al, Ag, As,B, Ba, Be, Ca, Cd, Co, Cr, Cu, Fe, K, Li, Mg, Mn, Mo, Na, Ni, P, Pb, S, Sb, Se, Si, Sn, Sr, Ti, Tl, U, V, Zn)								Please see sublet report attached.
pH, Conductivity and Total Alkalinity								
pH	5.22	0.10	pH	50ml	Prot#1401	NA	170314	
Conductivity (EC)	79.2	0.20	mS/cm	50ml	Prot#1403	10^{-3}	170321	
Bicarbonate (HCO3)	435.0	5.0	mg/L	NA	calculation	NA		
Carbonate (CO3)	<5	5.0	mg/L	NA	calculation	NA		
Hydroxide (OH)	<5	5.0	mg/L	NA	calculation	NA		
Alkalinity, Total (as $CaCO_3$)	356.6	5.0	mg/L	50ml	Prot#1405	10^{-2}	170321	
p-Alkalinity	<5	5.0	mg/L	NA	Prot#1405	NA		
TDS (Calculated)	8597.66	5.0	mg/L	NA	calculation	NA		
Ion Balance	53		%	NA	calculation	NA		
Sodium Adsorption Ratio	2.20			NA	calculation	NA		
Ortho -Phosphate	1443.5	0.1	mg/L	50ml	Prot#1421	10^{-3}	170320	
Biological Oxygen Demand (BOD)	1356.0	2.0	mg/L	0.5ml	Prot#1422	NA	170316	
Chemical Oxygen Demand (COD)	5600.00	3	mgO2/L	2.5ml	APHA 5220C	10^{-2}	170321	

*DF = Dilution Factor used for analysis

Vaishalli

Vaishalli Girdhar (Analyst)
Date: March 23 ,2017

Approved by: RSingh

Dr. Rajeshwar Singh, Director Laboratory Operations
Date: March 23 ,2017

9960 — 67th Avenue, Edmonton, Alberta, Canada T6E 0P5 Phone: (780) 450-3957 Fax: (780) 450-3960 Website : www.pbr.ca E-mail: pbr@pbr.ca

Manure loading 9.56 Dry Matter

Trial 10 ↑[SO4] H_2SO_4 pH Control Agent

PBR Laboratories Inc.
Call Toll Free: 1-866-450-3957

NH_4OH Base Agent

Certificate of Analysis

Sample ID and Parameters Measured	Result	Detection Limit	Units	Quantity Analyzed	Test Method	*DF	Date Analyzed	Note
PBR ID: 16-BMG-03								
Client ID: Quarantine Tank sample 6 2016/08/30 9:05								
Matrix: Greenhouse water								
Chloride by IC								
Chloride (Cl)	1121.00	0.50	mg/L	10ml	Prot#1412	10^{-2}	160831	
Nitrate as N by IC								
Nitrate (as N)	40.5	0.1	mg/L	10ml	Prot#1412	10^{-2}	160831	
Sulfate by IC								
* Sulfate (SO4)	28746.00	0.50	mg/L	10ml	Prot#1412	10^{-3}	160831	
Nitrite as N by IC								
Nitrite (as N)	24.20	0.10	mg/L	10ml	Prot#1412	10^{-2}	160831	
Nitrite + Nitrate as N	64.70	0.10	mg/L	NA	Calculation			
Ammonia as N by IC								
NH4-N	7109.00	0.70	mg/L	10ml	Prot#1410	10^{-1}	160902	
Dissolved Metals by ICP-OES								
Calcium (Ca) - Dissolved	2171.63	0.50	mg/L	20ml	Prot#1420	10^{-2}	160831	
Iron (Fe) - Dissolved	34.51	0.05	mg/L	20ml	Prot#1420	10^{-1}	160831	
Magnesium (Mg) - Dissolved	633.31	0.30	mg/L	20ml	Prot#1420	10^{-1}	160831	
Manganese (Mn) - Dissolved	42.15	0.01	mg/L	20ml	Prot#1420	10^{-1}	160831	
Potassium (K) - Dissolved	2449.78	0.30	mg/L	20ml	Prot#1420	10^{-2}	160831	
Sodium (Na) - Dissolved	560.71	0.10	mg/L	20ml	Prot#1420	10^{-1}	160831	
Total Hardness (as CaCO3)	8025.65	5	mg/L	NA	calculation	NA		In House
pH, Conductivity and Total Alkalinity								30 Aug 12 Sept
pH	6.13	0.10	pH	50ml	Prot#1401	NA	160830	5.8 5.4
Conductivity (EC)	95.47	0.20	mS/cm	50ml	Prot#1403	NA	160831	51.5 56
Bicarbonate (HCO3)	754.0	5.0	mg/L	NA	calculation	10^{-1}	160906	
Carbonate (CO3)	<5	5.0	mg/L	NA	calculation	NA	160906	
Hydroxide (OH)	<5	5.0	mg/L	NA	calculation	NA	160906	
Alkalinity, Total (as $CaCO_3$)	618.0	5.0	mg/L	50ml	Prot#1405	10^{-1}	160906	
p-Alkalinity	<5	5.0	mg/L	NA	Prot#1405	NA	160906	
TDS (Calculated)	36321.67	5.0	mg/L	NA	calculation	NA		
Ion Balance	45		%	NA	calculation	NA		
Sodium Adsorption Ratio	2.7			NA	calculation	NA		
Ortho -Phosphate	1199.1	0.1	mg/L	50ml	Prot#1421	10^{-4}	160902	

*DF = Dilution Factor used for analysis

Manure Loading 13.4%

Vaishalli Girdhar (Analyst)
Date: Sept 7, 2016

Dry Matter

Approved by: Dr. Rajeshwar Singh, Director Laboratory Operations
Date: Sept 7, 2016

9960 – 67th Avenue, Edmonton, Alberta, Canada T6E 0P5 Phone: (780) 450-3957 Fax: (780) 450-3960 Website : www.pbr.ca E-mail: pbr@pbr.ca

Alberta Agriculture and Forestry | **The ACIDF Project** | **Nutrient Recycling Group**

Trial 6

ROUTINE WATER - FILTERED (WATER)

	UNITS	TRIAL 6DECANT
Runtime Days		**33.9**
Runtime Hours		**813.0**
Calculated Parameters		
Anion Sum meq/L	**meq/L**	**770**
Cation Sum meq/L	**meq/L**	**520**
Hardness (CaCO3)	**mg/L**	**<0.50**
Ion Balance	N/A	0.67
Dissolved Nitrate (NO3)	**mg/L**	**<4.4**
Nitrate plus Nitrite (N)	**mg/L**	**<0.020**
Dissolved Nitrite (NO2)	**mg/L**	**<3.3**
Total Dissolved Solids	**mg/L**	**44000**
Misc. Inorganics		
Conductivity	**mS/cm**	
Anions		
Alkalinity (PP as CaCO3)	mg/L	<10 (9)
Alkalinity (Total as CaCO3	**mg/L**	**7000 (9)**
Bicarbonate (HCO3)	mg/L	8500 (9)
Carbonate (CO3)	mg/L	<10 (9)
Hydroxide (OH)	mg/L	<10 (9)
Dissolved Sulphate (SO4)	**mg/L**	**1300 (9)**
Dissolved Chloride (Cl)	mg/L	1100 (9)
Nutrients		
Dissolved Nitrite (N)	**mg/L**	**<1.0 (9)**
Dissolved Nitrate (N)	**mg/L**	**<1.0 (9)**
Lab Filtered Elements		
Dissolved Calcium (Ca)	**mg/L**	**<30**
Dissolved Iron (Fe)	**mg/L**	**0.83**
Dissolved Magnesium (Mg)	**mg/L**	**<20**
Dissolved Manganese (Mn)	**mg/L**	**<0.40**
Dissolved Potassium (K)	**mg/L**	**1500**
Dissolved Sodium (Na)	**mg/L**	**300**
RESULTS OF CHEMICAL ANALYSES OF WATER		
Maxxam ID		OF0794
Sampling Date		2016/03/01 10:30
Demand Parameters		
Biochemical Oxygen Deman	mg/L	380 (30)
Total Chemical Oxygen Dem	mg/L	9500 (14)
Misc. Inorganics		
Conductivity MS/cm	mS/cm	32.0
pH	pH	6.47
Anions		
Dissolved Fluoride (F)	mg/L	0.32
Total Ammonia (N) mg/L	mg/L	6500 (29)
Orthophosphate (P) mg/L	mg/L	8800 (29)

Maxxam Job Number: B615638
Report Date: 2016/03/09

Maxxam ID		OF0794
Sampling Date		2016/03/01 10:30

H_3PO_4 pH Control Agents

NH_4OH

Bioreactor Loading
6.46 Dry Matter Manure
Total Broth (manure + Mother Liquor)
11.2% dry matter

\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015, 2016\Graph's and Data\Nutrient Profile using H3PO4 as pH Control Agent.xlsxAF data Result (1)

AAAF 160907-01 (16-BMG-05) Stock Tank HNO_3 pH

PBIR Laboratories Inc.

Call Toll Free: 1-866-450-3957

Control Agent

Certificate of Analysis

Sample ID and Parameters Measured	Result	Detection Limit	Units	Quantity Analyzed	Test Method	*DF	Date Analyzed	Note
PBR ID: 16-BMG-05								
Client ID: Nutrient Stock Tank sample 9 2016/08/30 9:10								
Matrix: Greenhouse water								
Chloride by IC								
Chloride (Cl)	1233.80	0.50	mg/L	10ml	Prot#1412	10^{-2}	160831	
Nitrate as N by IC								
* Nitrate (as N)	7523.65	0.1	mg/L	10ml	Prot#1412	10^{-3}	160831	
Sulfate by IC								
Sulfate (SO4)	1735.10	0.50	mg/L	10ml	Prot#1412	10^{-2}	160831	
Nitrite as N by IC								
Nitrite (as N)	3.83	0.10	mg/L	10ml	Prot#1412	10^{-2}	160831	
Nitrite + Nitrate as N	7527.48	0.10	mg/L	NA	Calculation			
Ammonia as N by IC								
NH4-N	5882.33	0.70	mg/L	10ml	Prot#1410	10^{-1}	160902	
Dissolved Metals by ICP-OES								
Calcium (Ca) - Dissolved	912.97	0.50	mg/L	20ml	Prot#1420	10^{-2}	160831	
Iron (Fe) - Dissolved	13.25	0.05	mg/L	20ml	Prot#1420	10^{-2}	160831	
Magnesium (Mg) - Dissolved	395.50	0.30	mg/L	20ml	Prot#1420	10^{-2}	160831	
Manganese (Mn) - Dissolved	20.44	0.01	mg/L	20ml	Prot#1420	10^{-2}	160831	
Potassium (K) - Dissolved	2295.90	0.30	mg/L	20ml	Prot#1420	10^{-2}	160831	
Sodium (Na) - Dissolved	668.94	0.10	mg/L	20ml	Prot#1420	10^{-2}	160831	
Total Hardness (as CaCO3)	3903.98	5	mg/L	NA	calculation	NA		In House
pH, Conductivity and Total Alkalinity								30 Aug 12 Sep
pH	4.86	0.10	pH	50ml	Prot#1401	NA	160830	4.6 4.57
Conductivity (EC)	82.94	0.20	mS/cm	50ml	Prot#1403	NA	160831	57 70
Bicarbonate (HCO3)	141.5	5.0	mg/L	NA	calculation	10^{-1}	160906	
Carbonate (CO3)	<5	5.0	mg/L	NA	calculation	NA	160906	
Hydroxide (OH)	<5	5.0	mg/L	NA	calculation	NA	160906	
Alkalinity, Total (as $CaCO_3$)	116.0	5.0	mg/L	50ml	Prot#1405	10^{-1}	160906	
p-Alkalinity	<5	5.0	mg/L	NA	Prot#1405	NA	160906	
TDS (Calculated)	40631.97	5.0	mg/L	NA	calculation	NA		
Ion Balance	57		%	NA	calculation	NA		
Sodium Adsorption Ratio	4.7			NA	calculation	NA		
Ortho -Phosphate	1280.5	0.1	mg/L	50ml	Prot#1421	10^{-4}	160902	

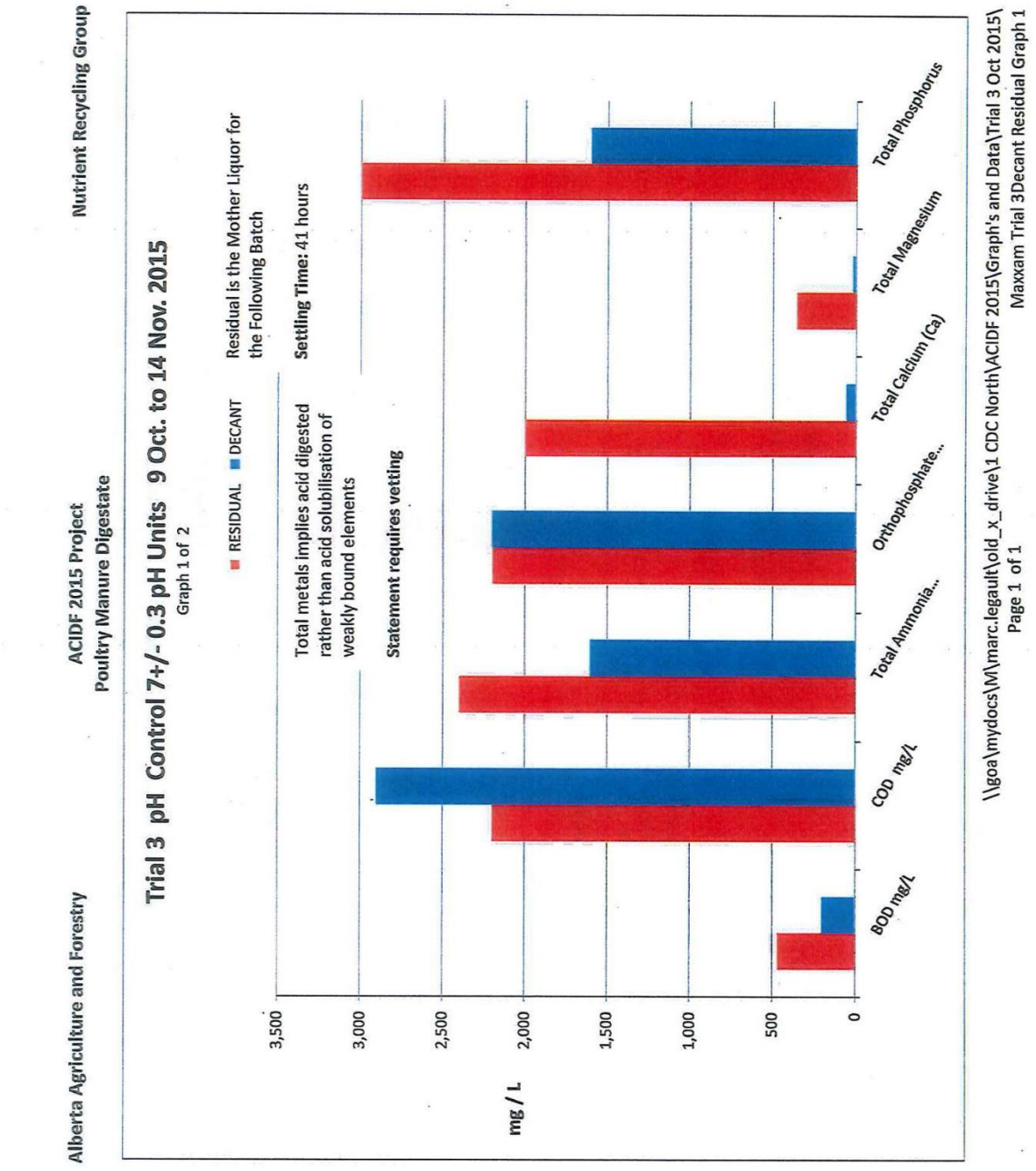
*DF = Dilution Factor used for analysis

Vaishalli Girdhar (Analyst)
Date: Sept 7, 2016

Approved by:
Dr. Rajeshwar Singh, Director Laboratory Operations
Date: Sept 7, 2016

9960 — 67th Avenue, Edmonton, Alberta, Canada T6E 0P5 Phone: (780) 450-3957 Fax: (780) 450-3960 Website : www.pbr.ca E-mail: pbr@pbr.ca

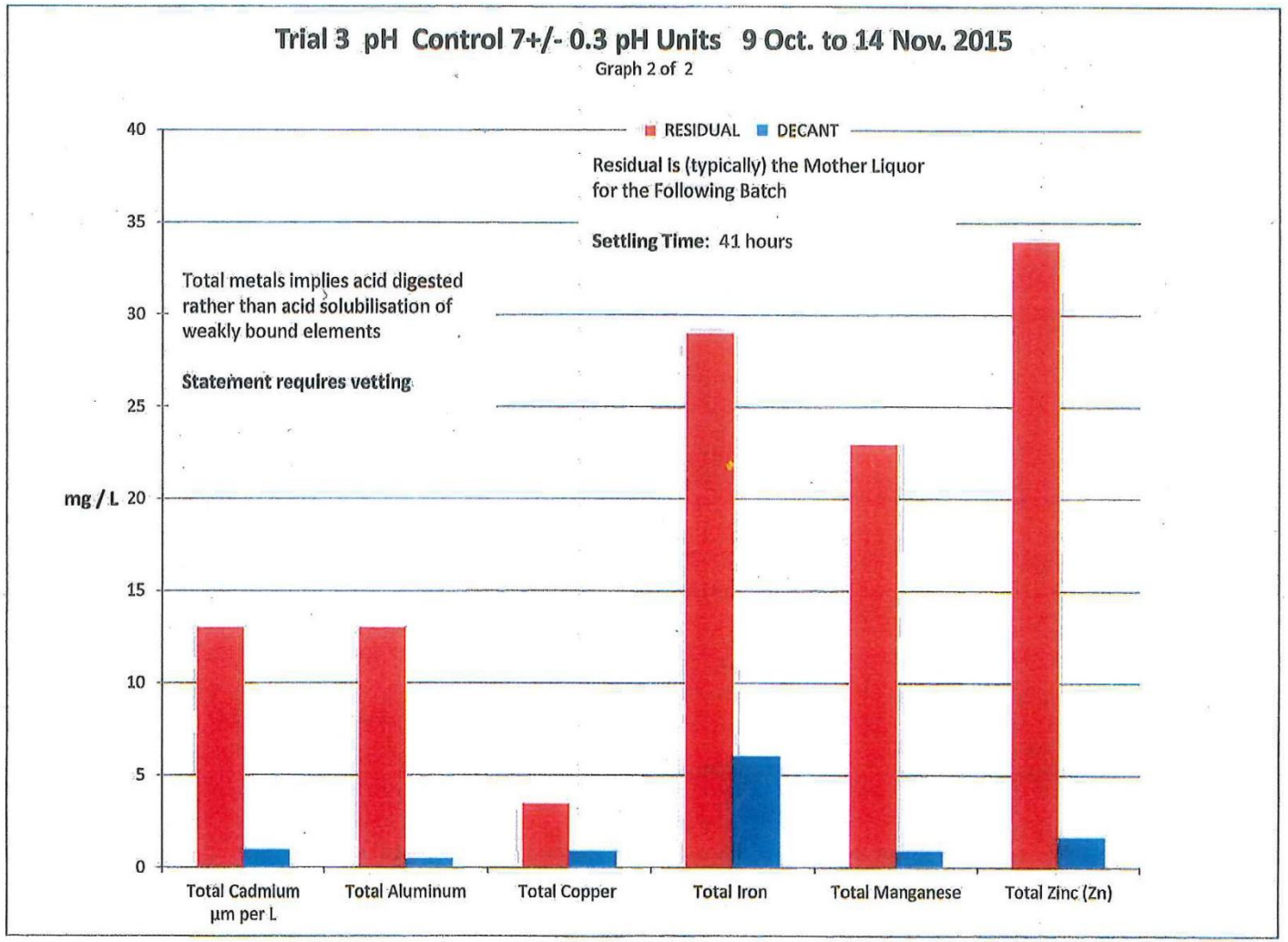
Attachment 6 – Residual vs Decant Nutrient Bar Graphs



Alberta Agriculture and Forestry

ACIDF 2015 Project
Poultry Manure Project

Nutrient Recycling Group



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Maxxam Trial 3Decant Residual Graph 2

REGULATED METALS (CCME/AT1) - TOTAL

Maxxam ID	NQ6759	NQ6760							
Sampling Date	2015/11/16 09:00	2015/11/16 09:00		Alberta Tier 1 soil remediation			Category A Manure Max. Conc. Within Product from Guidelines for Compost Quality PN 1340 Can. Council of Ministers of the Environment, 2005		
	TRIAL 3 DECANT	**TRIAL 3 RESIDUAL**	**UNITS**	**Value**	**Units**	**Notes**	**Value**	**Units**	**Notes**
Low Level Elements									
Total Cadmium	0.92	13	ug/L	1.4	mg/kg		3.00	mg/kg	
Elements									
Total Aluminum	0.46	13	mg/L						
Total Antimony (Sb)	<0.0024	<0.0024	mg/L	20	mg/kg				
Total Arsenic (As)	0.011	0.014	mg/L	17	mg/kg	inorganic	13	mg/kg	
Total Barium (Ba)	<1.0	1.7	mg/L	750	mg/kg	non-barite			
Total Beryllium (Be)	<0.0040	<0.0040	mg/L	5	mg/kg				
Total Boron (B)	**<2.0**	**<2.0**	**mg/L**	**2**		**hot water soluble**			
Total Calcium (Ca)	58	2000	mg/L						
Total Chromium (Cr)	0.013	0.20	mg/L	64	mg/kg	total [hexavalent 0.4 mg/kg]	210	mg/kg	
Total Cobalt (Co)	0.014	0.031	mg/L	20	mg/kg				
Total Copper	0.85	3.5	mg/L	63	mg/kg		400	mg/kg	
Total Iron	<6.0	29	mg/L						
Total Lead (Pb)	0.0016	0.039	mg/L	70	mg/kg		150	mg/kg	
Total Lithium (Li)	<2.0	<2.0	mg/L						
Total Magnesium	<20	360	mg/L						
Total Manganese	0.85	23	mg/L						
Total Molybdenum	0.10	0.074	mg/L	4	mg/kg		5	mg/kg	
Total Nickel (Ni)	0.076	0.27	mg/L	50	mg/kg		62	mg/kg	
Total Phosphorus	1600	3000	mg/L						
Total Potassium (K)	530	540	mg/L						
Total Selenium (Se)	0.019	0.035	mg/L	1	mg/kg		2	mg/kg	
Total Silicon (Si)	17	22	mg/L						
Total Silver (Ag)	0.00054	0.00078	mg/L	20	mg/kg				
Total Sodium (Na)	130	130	mg/L						
Total Strontium (Sr)	<2.0	2.6	mg/L						
Total Sulphur (S)	170	160	mg/L						
Total Thallium (Tl)	0.0014	0.0052	mg/L	1	mg/kg				
Total Tin (Sn)	<0.0040	<0.0040	mg/L	5	mg/kg				
Total Titanium (Ti)	0.045	0.33	mg/L						
Total Uranium (U)	0.0044	0.092	mg/L	23	mg/kg				
Total Vanadium (V)	0.019	0.11	mg/L	130	mg/kg				
Total Zinc (Zn)	**1.6**	**34 (1)**	**mg/L**	**200**	**mg/kg**		**700**	mg/kg	
mercury inorganic				6.6			0.80	mg/kg	

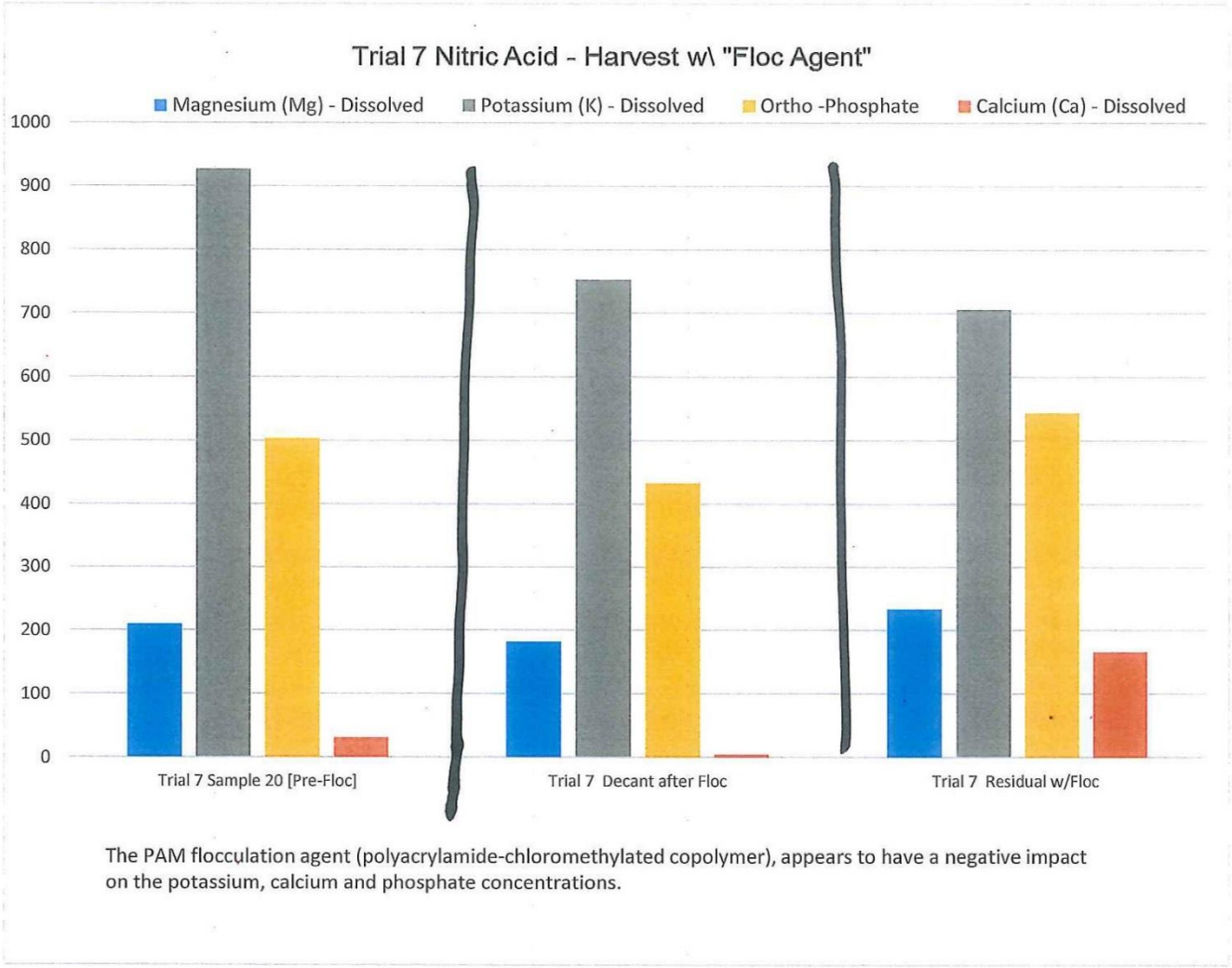
Alberta Agriculture and Forestry

ACIDF 2015 2016 Project Poultry Manure Digestate

Nutrient Recycling Group

The PAM flocculation agent (polyacrylamide-chloromethylated copolymer), appears to have a negative impact on the dissolved nitrate and ammonium concentrations.

\\GOA\MyDocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015, 2016\Graph's and Data\Nitric Trial 7 to\Trial 7 Mar 2016\
Trial 7 Decant vs Residual w Floc Agent .xlsx Graphs



The PAM flocculation agent (polyacrylamide-chloromethylated copolymer), appears to have a negative impact on the potassium, calcium and phosphate concentrations.

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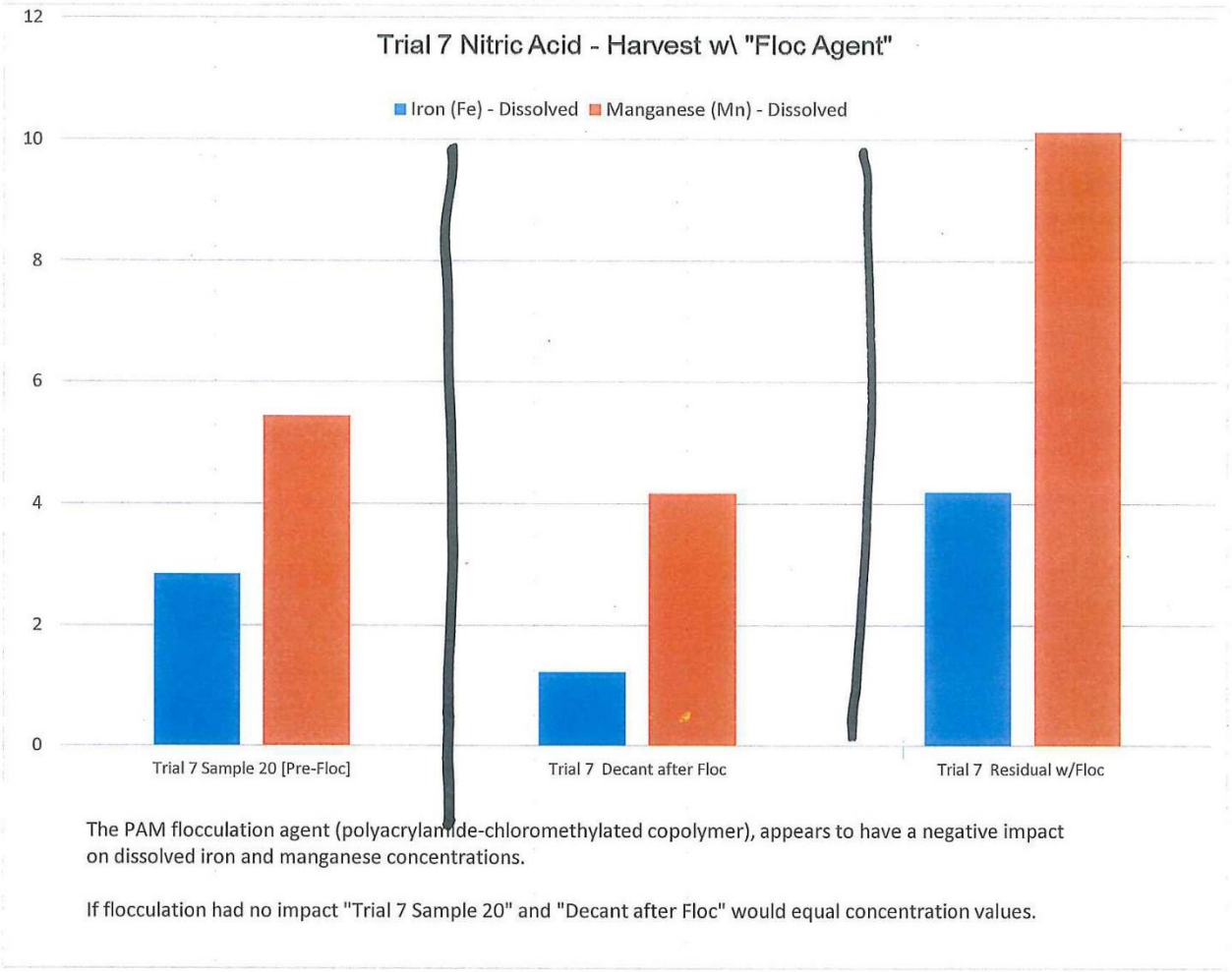
Trial 7 Decant vs Residual w Floc Agent .xlsx Graphs



The PAM flocculation agent (polyacrylamide-chloromethylated copolymer), appears to have a negative impact on dissolved iron and manganese concentrations.

If flocculation had no impact "Trial 7 Sample 20" and "Decant after Floc" would equal concentration values.

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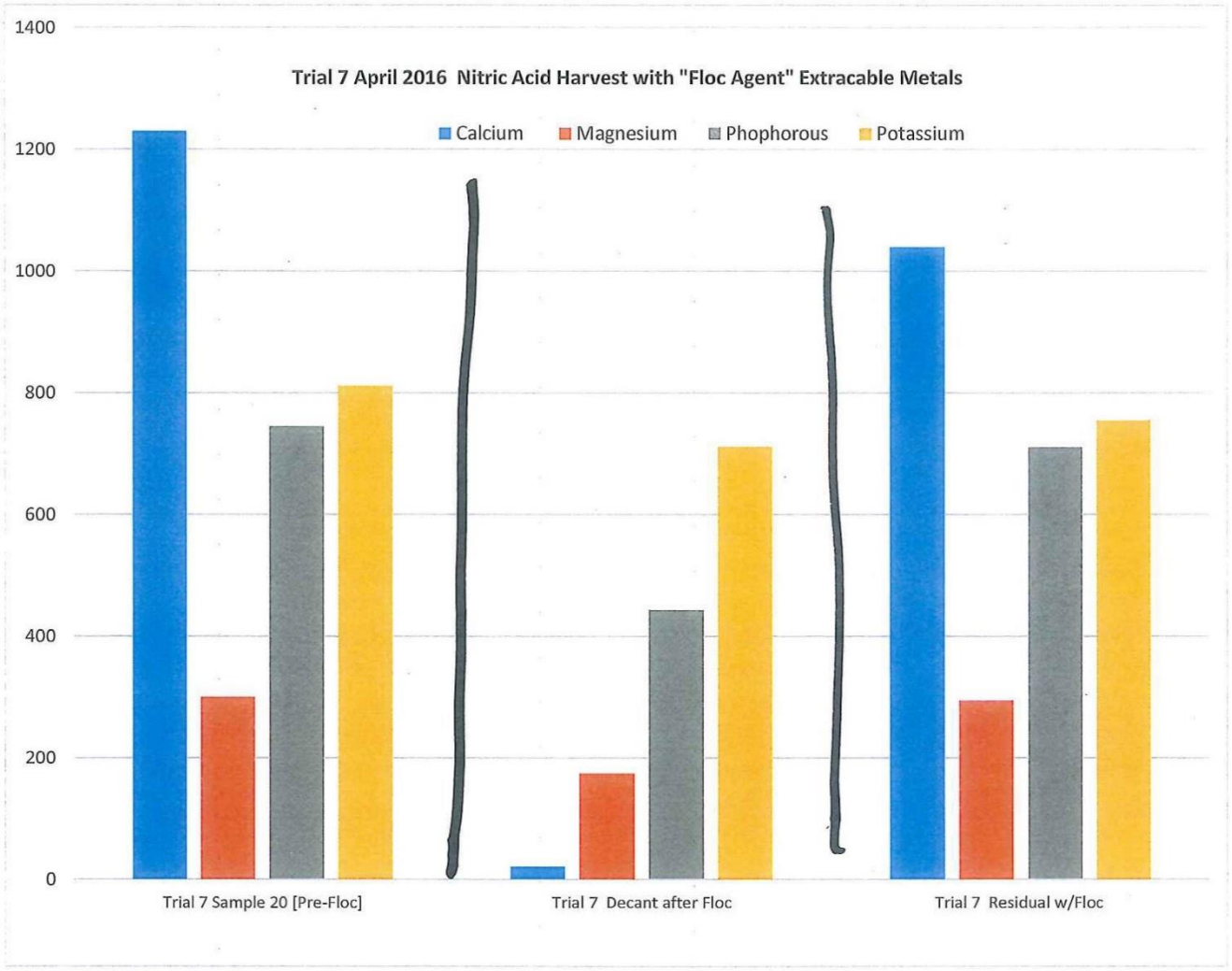
Trial 7 Decant vs Residual w Floc Agent .xlsx Graphs



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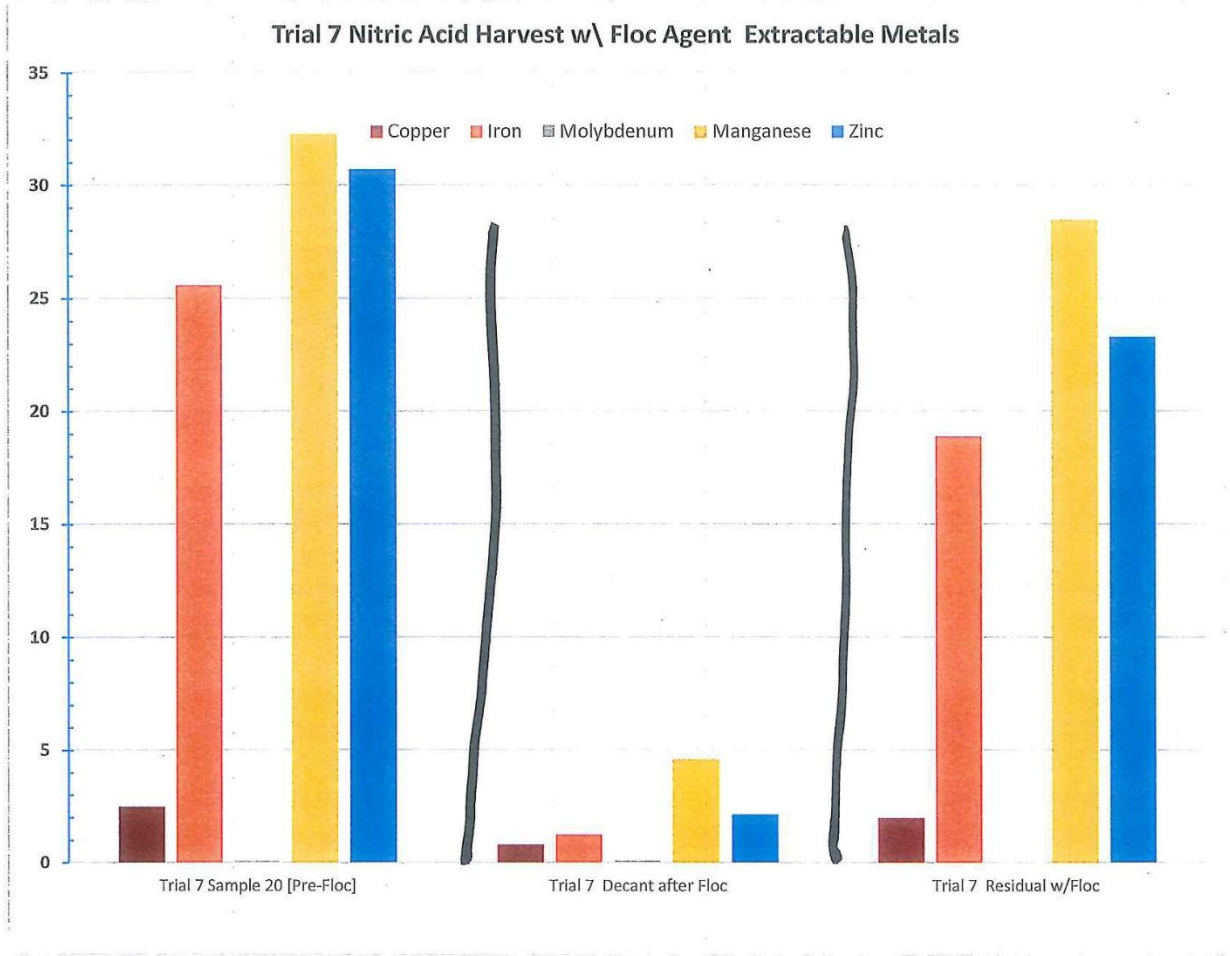
Trial 7 Decant vs Residual w Floc Agent .xlsx Graphs

Alberta Agriculture and Forestry | ACIDF 2015 2016 Project Poultry Manure Digestate | Nutrient Recycling Group



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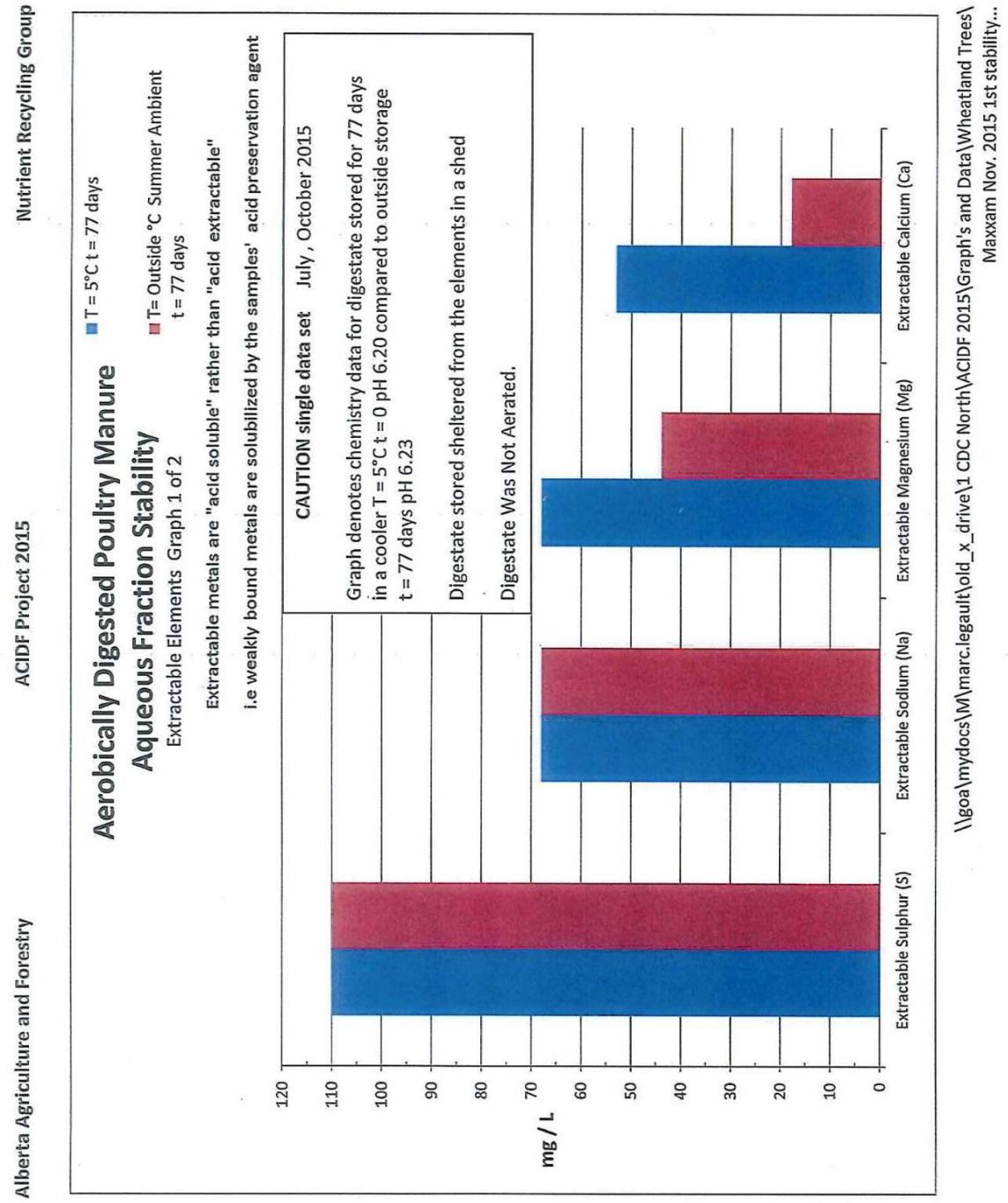
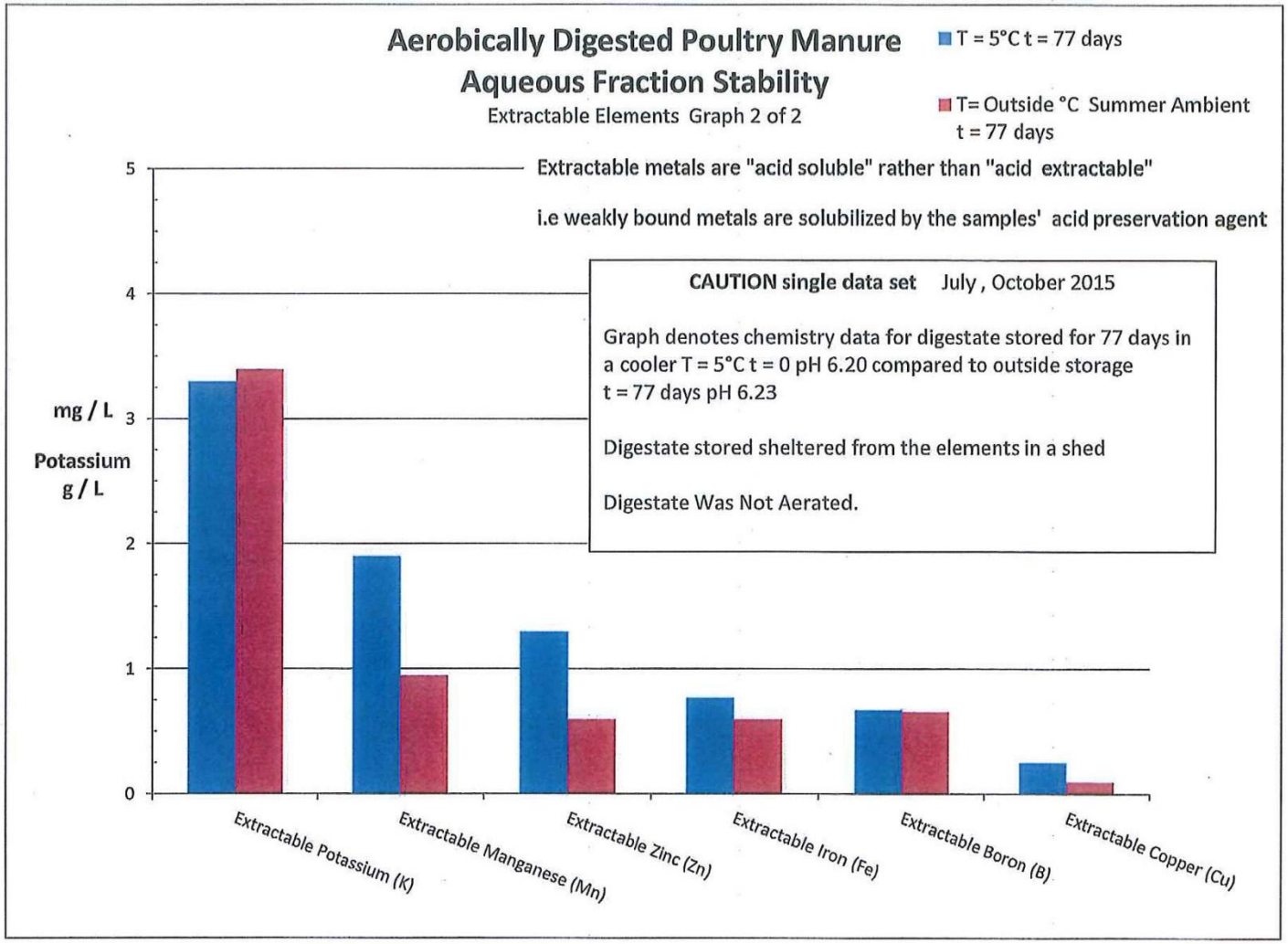
Trial 7 Decant vs Residual w Floc Agent .xlsx Graphs


Alberta Agriculture and Forestry
ACIDF Project 2015
Nutrient Recycling Group
Aerobically Digested Poultry Manure
Aqueous Fraction Stability
Extractable Elements Graph 1 of 2
Extractable metals are "acid soluble" rather than "acid extractable"
i.e weakly bound metals are solubilized by the samples' acid preservation agent
T = 5°C t = 77 days
T= Outside °C Summer Ambient
t = 77 days
CAUTION single data set July , October 2015
Graph denotes chemistry data for digestate stored for 77 days in a cooler T = 5°C t = 0 pH 6.20 compared to outside storage t = 77 days pH 6.23
Digestate stored sheltered from the elements in a shed
Digestate Was Not Aerated.
mg / L
120
110
100
90
80
70
60
50
40
30
20
10
0
Extractable Sulphur (S)
Extractable Sodium (Na)
Extractable Magnesium (Mg)
Extractable Calcium (Ca)
\\goa\mydocs\M\marc.legault\old_x_drive\1 CDC North\ACIDF 2015\Graph's and Data\Wheatland Trees\
Maxxam Nov. 2015 1st stability...



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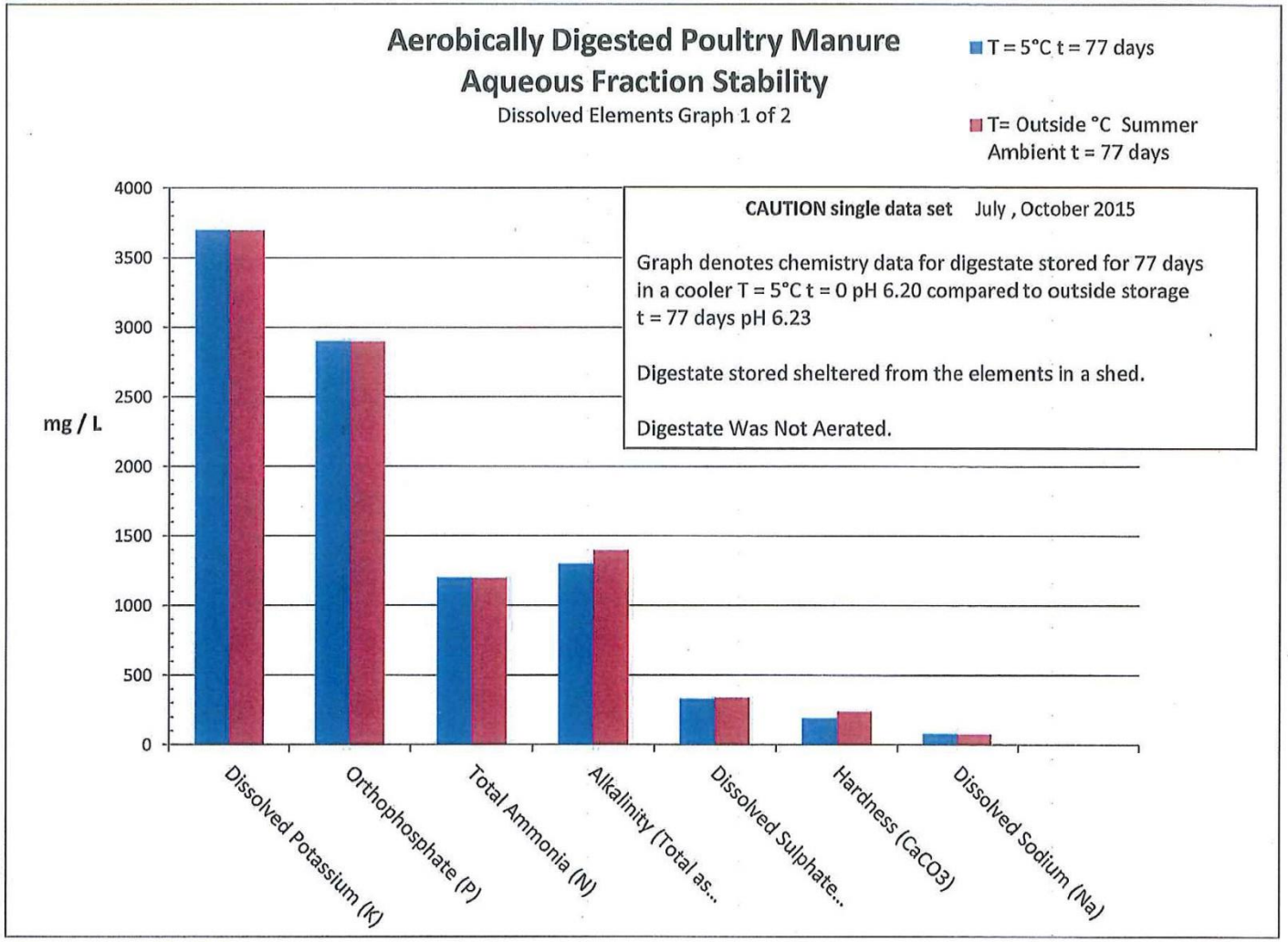

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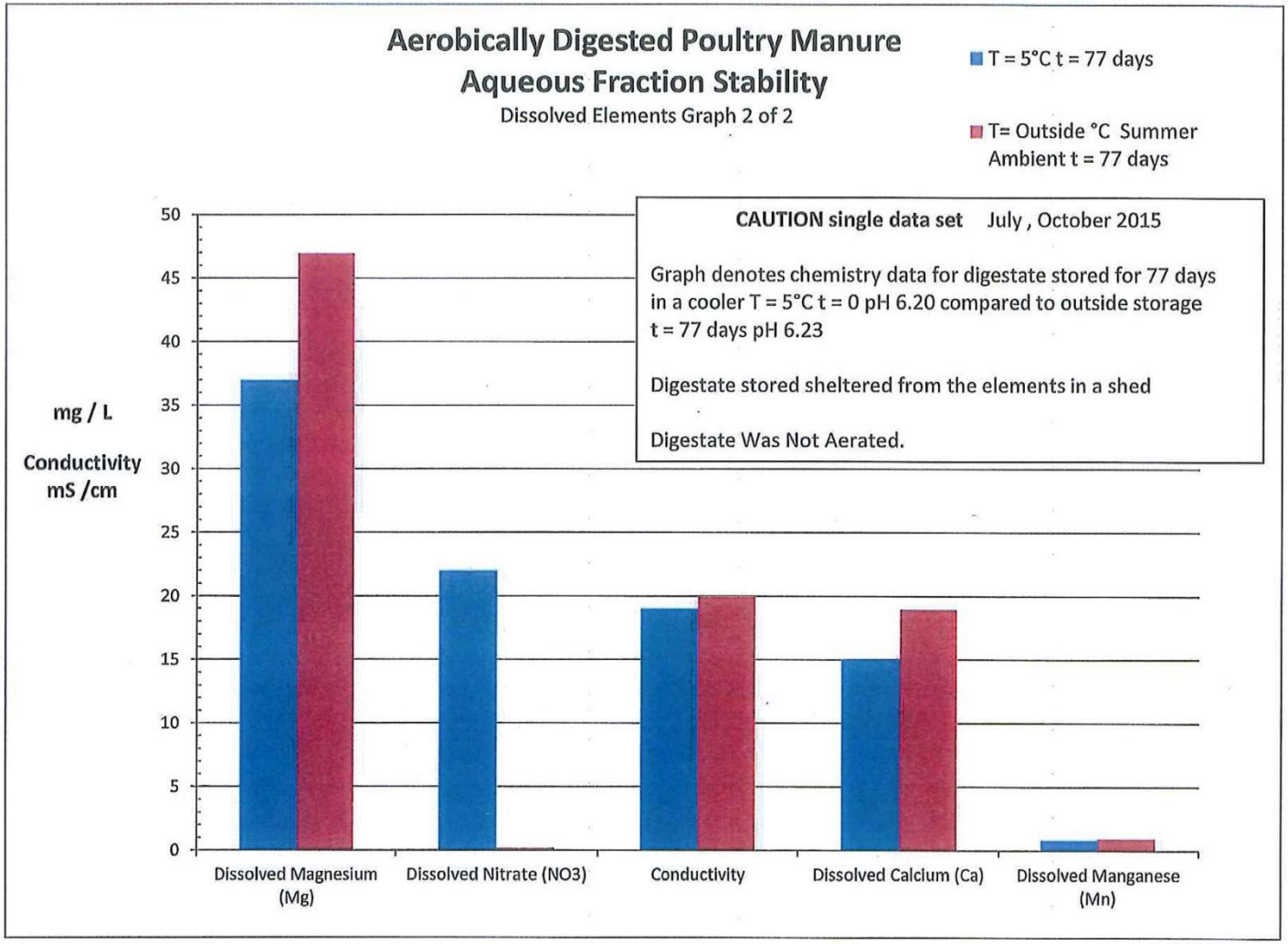
Maxxam Nov. 2015 1st stability...


Aerobically Digested Poultry Manure
Aqueous Fraction Stability
Dissolved Elements Graph 2 of 2
T = 5°C t = 77 days
T= Outside °C Summer Ambient t = 77 days
CAUTION single data set July , October 2015
Graph denotes chemistry data for digestate stored for 77 days in a cooler T = 5°C t = 0 pH 6.20 compared to outside storage t = 77 days pH 6.23
Digestate stored sheltered from the elements in a shed
Digestate Was Not Aerated.
mg / L
Conductivity mS /cm
50
45
40
35
30
25
20
15
10
5
0
Dissolved Magnesium (Mg)
Dissolved Nitrate (NO3)
Conductivity
Dissolved Calcium (Ca)
Dissolved Manganese (Mn)