# THE POTENTIAL OF GOAT MANURE AS A HYDROPONIC NUTRIENT ALTERNATIVE FOR TOMATO PRODUCTION IN NAMIBIA

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## ABSTRACT

Hydroponics is a cultivation method of growing plants in water. It is a technology suitable for farming that is under artificially controlled environments but can be adapted to situations where land is a constraint. In Namibia this is a potential method for vegetable cultivation. Hydroponics vegetable production has been demonstrated successfully using a balanced conventional nutrient solution, globally, and therefore, an evaluation of its suitability in Namibia using goat manure solution as a source of organic hydroponic nutrients. An alternative hydroponics nutrient solution has been sought after but often failed due to nitrogen in organic sources being predominantly organic, a form plants cannot access. The study had two stages, the first being a laboratory experiment aimed at formulating a Goat Manure Derived Hydroponic Nutrient Solution (GMDHNS). This involved the determination of source of microorganisms, concentration of microorganisms and amount of goat manure for optimal generation of nitrates from goat manure. Variables measured included nitrate levels generated and levels of other nutrients. The second stage was a field experiment which tested the GMDHNS effect on tomato performance and quality. Variables measured included, plant height, stem diameter, nutrient use efficiency, plant biomass, number of flowers, number of fruits, yield, total soluble solids, and lycopene content. Commercial Hydroponic Nutrient Solution and tap water were used as controls. Locally sourced microorganisms significantly ($P \leq 0.05$) transformed organic nitrogen in goat manure into plant available nitrates reaching levels of 198mg/L hence was the basis of the organic hydroponic nutrient solution. Tomato vegetative growth (height, stem diameter and leaves) were significantly ($P \leq 0.05$) improved by the formulated GMDHNS compared to growing tomatoes in Tap Water (TW) where no fertiliser was added and that GMDHNS was comparable to using Commercial Hydroponic Nutrient Solution (CHNS) in terms of plant growth. Plant reproduction (flower and fruits) were also significantly ($P \leq 0.05$) influenced by the formulated GMDHNS more than by the TW treatment even though yield from GMDHNS was less than that from the CHNS. The quality (lycopene, Total Soluble Solids, fruit size and weight) of fruits produced from plants in the GMDHNS was significantly ($P \leq 0.05$) more than that in fruits from the CHNS. Thus, based on the results from this study, it can be concluded that a hydroponic nutrient solution can be formulated from goat manure. Furthermore, it can

be concluded that the locally formulated hydroponic nutrient solution enhances quality, growth and reproductive performance of tomato, even though it may still need further optimization. Follow-up studies to the present study should be done to further optimise GMDHNS and that artefactual should also follow for practical uptake of these findings.

## PUBLICATIONS

1. Mowa, E., Akundabweni, L., Chimwamurombe, P., Oku, E., & Mupambwa, H. A. (2017). The influence of organic manure formulated from goat manure on growth and yield of tomato (Lycopersicum esculentum). *African Journal of Agricultural Research*, *12* (41), 3061-3067.
2. Mowa, E., Akundabweni, L., Chimwamurombe, P., & Liswaniso, G. (2018). Analysis of nitrifying microbial community for organic hydroponics. African Journal of Agricultural Research, *12* (1), 1-8.

*3.* Mowa, E., Akundabweni, L., Chimwamurombe, P., Oku, E. IN PRESS. Formulation of an organic hydroponic nutrient solution using nitrifying microorganisms. *International Science & Technology Journal of Namibia.*

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**These are definitions or meanings of terminologies frequently used in this study and are as follows:**

**Hydroponics –** a soilless cultivation method for producing horticultural crops in water without soil (dos Santos, 2013).

**Organic farming –** Is a farming method which depends on fertilizers of organic origin like animal manure, avoiding the use of synthetic substances whilst promoting utilisation of naturally occurring substances (Lampkin, 1990).

**Conventional farming -** The intensive use of fertilizers, pesticides and other synthetic for intensive crop production (Pimentel *et al.*, 2005).

**AOB - A**mmonia-Oxidizing Bacteria, microorganisms oxidising ammonia into nitrite (Saijai *et al.*, 2016).

**NOB -** Nitrite-Oxidizing Bacteria, microorganisms oxidising nitrite into nitrate (Saijai *et al.*, 2016).

**TSS –** Total Soluble Solids

**GMDHNS –**Goat Manure Derived Hydroponic Nutrient Solution

**CHNS –** Commercial Hydroponic Nutrient Solution.

**TW –** Tap Water

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## DEDICATIONS

I would like to dedicate this work to my late parents for their foundation in my life has moved strides in my education.

## DECLARATIONS

I, Edgar Mowa Mowa, hereby declare that this study is my own work and is a true reflection of my research, and that this work, or any part thereof has not been submitted for a degree at any other institution.



…….. …… …….

**Name of Student** **Signature** **Date**

# CHAPTER 1: INTRODUCTION

## 1.1 Rationale for organic hydroponic nutrient solution formulation and efficacy on tomato crop

The use of mineral fertilisers for agriculture is relatively expensive worldwide (Dopler, 1980; Osman *et al.*, 2009) and particularly in Africa (Sanchez, 2002). Yet nutrition required for food production (quality and quantity) remains a priority for food security in general, and for vegetables value chains in particular. Farmers therefore, use little or no commercial fertilisers for fear of high cost (Mowa, 2015). Therefore, the current trend depending on expensive fertilisers has failed to achieve the benefit of increased production from use of available critical macro and micro nutrients as a means of increasing the value addition of specialised production for the horticultural market. The goal in retrospect is to search for alternative means for specialised horticultural production.

The group of farmers who have been successful in Namibia have successfully farmed with adequate fertilisers input to increase their commercial production. That was particularly true during the colonial era. At present, the country has a mix of groups of farmers who can afford fertilisers, specialised production (e.g. horticultural) and those groups who would wish to use fertilisers and produce, but do not use any because of sustainability concerns (Mendelsohn, 2006). Thus, the gap of farmers who are able and those who are unable to afford fertilisers. It is in the context of the aforementioned gap that the problem of the commercial horticultural production is contributing to the law of the minimum(also known as Liebig's law of the minimum), which purports that when one resource among others required is scarce, it will be the most limiting to

otherwise desired level of production (Van der Ploeg *et al.*, 1999).Therefore, the group that is unable to afford fertilisers will be the victims of the law of the minimum and in turn horticultural production will not reach the level where Namibia becomes self-sustained in terms of vegetable production, limited by those unable to afford fertilisers. It is only when groups of horticultural producers in Namibia are all enabled to afford fertilisers and water, that full production to sufficiently meet and sustain vegetable supply demand will be realised. Therefore, currently there is a weak-link at the production stage in the value addition chain, that is brought about by fertiliser as a limiting resource for some other group of farmers who would have been otherwise equally productive and contributing to required production that would see Namibia produce enough for own consumption and even export surplus.

Such production could include important vegetables in Namibia such as tomatoes which are important both economically and nutritionally. Currently tomato production in Namibia depends on rain-fed systems and expensive fertilisers to many. Yet there exist alternatives for tomato cultivation such as hydroponics (Shinohara *et al.*, 2011). The challenge in the use of hydroponics for tomato cultivation in Namibia is that it relies on the unaffordable fertilisers is an untested technology in Namibia).

The questions arising from this situation therefore, are:

1. According to Liebig's law of the minimum, is horticultural production controlled by the total amount of fertiliser resources available?
2. For specialised production, are there cheaper and practical hydroponic production alternatives that can be enhanced for home-grown hydroponic solutions?

3. Can locally formulated hydroponic solutions in horticultural production provide alternatives to scarce fertiliser resources?
4. Can locally formulated hydroponic solutions in horticultural production improve quality and quantity of horticultural produce?

The goal of this study therefore, is to find an alternative hydroponic method of producing tomatoes using local resources. The route to take such an investment tends to go hand in hand with (a) research hypotheses and (b) objectives.

## 1.2 Research Hypotheses

1. Hydroponic nutrient solutions can be formulated from goat manure
2. Formulations made from goat manure have positive effects on the growth and development (and therefore, on performance and quality) of tomato similar to other solution based media of cultivation.
3. Formulations made from goat manure have positive effects on quality of tomato fruits similar to other solution based media of cultivation.

In view of the foregoing, the objectives are:

## 1.3 Objectives

1. To formulate hydroponic nutrient solution from Goat Manure
2. To determine performance of tomato using goat manure-derived hydroponic nutrient solution
3. To determine the quality of tomatoes produced from organic hydroponic nutrients.

## 1.4 Significance

The current study seeks to investigate manure as a hydroponic alternative to conventional hydroponic fertilizers that are too costly for most Namibian farmers. The breakthrough in this regard will be to provide relief to communities on the Namibian coast specifically, and the Namibian population in general, who will in turn benefit economically and in terms of income generation. Therefore, a patent is envisaged for this work considering it is the first of its kind. Moreover, the current study will contribute to environmental safeguarding in that using hydroponics versus using soil based productions will lead to less underground water pollution and soil acidification, hence the green economy. Locals who will benefit the most include farmers who currently have poor soils and less fresh water in Namibia, making the manure from their animals worthless even if they applied it to their soils, it will not add value. Therefore, they will benefit greatly from the application of this cultivation method adding value to manure in their environments. Manure at the moment in Namibia is mostly freely available and those who charge do so at a minimal fee around N$10 per 20 kg (50 cents per kilogram), compared to N$ 800 per 25 kg conventional fertiliser (N$32 per kilogram).

## 1.5 Limitations of the study

Due to the fact that this is a baseline study in Africa, it only focuses on one type of animal manure (goat) and one vegetable (tomato). The study further narrows to focus on the limiting nutrient (nitrogen) so-far in organic hydroponic nutrient production. Hence, there will be need for further investigations into other liquid manures and vegetables for more comprehensive knowledge.

## 1.6 Conceptual framework to this study

Whereas this study is typically a standard field trial within the expected norms (Fig. 1 – top most arrow) the bottom most line is included to represent a projected subsequent spinoff that is suggested as a required artefactual business experiment in order that the findings can be actualized under the real-world conditions (Voors *et al.*, 2016). A conceptual framework is a useful tool in that its conceptual constructs when simply used can give a sense of a roadmap to a linear study such as the current one and also can advance the rationale for conducting a study. According to Fig.1, the orientation of the experimental method is as shown in the top most arrow that progresses through into an expected outcome of which value is described here as the output. The later seeks to develop empirically founded experimental propositions as precedents that are 'talking' to the different levels of tomato product value chain development (HTPVD) as represented by the bottom most arrow (Fig. 1). The HTPVD continuum in this study is included to suggest that the results so far found, may not be immediately applicable to business users unless artefactual (inclusive of framed field) business experiment has to be the second phase to this study.

This is in consideration of factors that will lead to the success of the organic hydroponic solution as outlined by Voors *et al* (2016). First, adoption of hydroponics technology is practical to local farmers in that it uses simplified local resources such as goat manures which are abundantly available in the Erongo region as inputs. In contrast to the already failed adoption of hydroponics based on the non-accessible costly resources that come along with the use of hydroponics based on conventional hydroponic fertilizers (Voors *et al.*, 2016). In the context of Voors et al., (2016), the

review that follows are as discussed below in the interest of a possible adoption potential Hydroponic Tomato Product Value-chain Development (HTPVD).

Therefore, in the second instance, access to finance for local farmers to participate in hydroponic vegetable production is another handicap for most cannot afford to sustain hydroponic operations based on the current costs associated with conventional hydroponics. In the third instances, with the abundance of organic sources of nutrients such as goat, cattle and chicken manures in the Erongo region, local community members i.e. those raring goats can form social network groups where they could encourage those within their circles to upscale the local organic hydroponic solution for vegetable production in contrast to the conventional hydroponic nutrient solution which according to the locals is to be afforded by only certain members of society with financial abilities. This leaves the majority of them inaccessible to this technology even if they wished to part-take.

Fourth, with the awareness by locals that what they regard as waste and is abundantly available with them can be added value to, they would take ownership of the technology knowing they can benefit more than one way of just cultivating, but also selling of manure. This could lead to even some keeping goats specifically for manure production and selling. Finally, migrants who flock into coastal towns such as Henties Bay for seasonal employments such as during the culling of seals season, would upscale the technology for periods that vegetable production using goat manure hydroponically will be active, they can switch to when they wait for the next seasonal offers.

Voors et al (2016) recently expounded on the concept of artefactual field studies which in other words are described as lab-in-the-field experiments. Binswanger (1981) further adds framed field experiments as an additional context to that of Voors et al (2016).

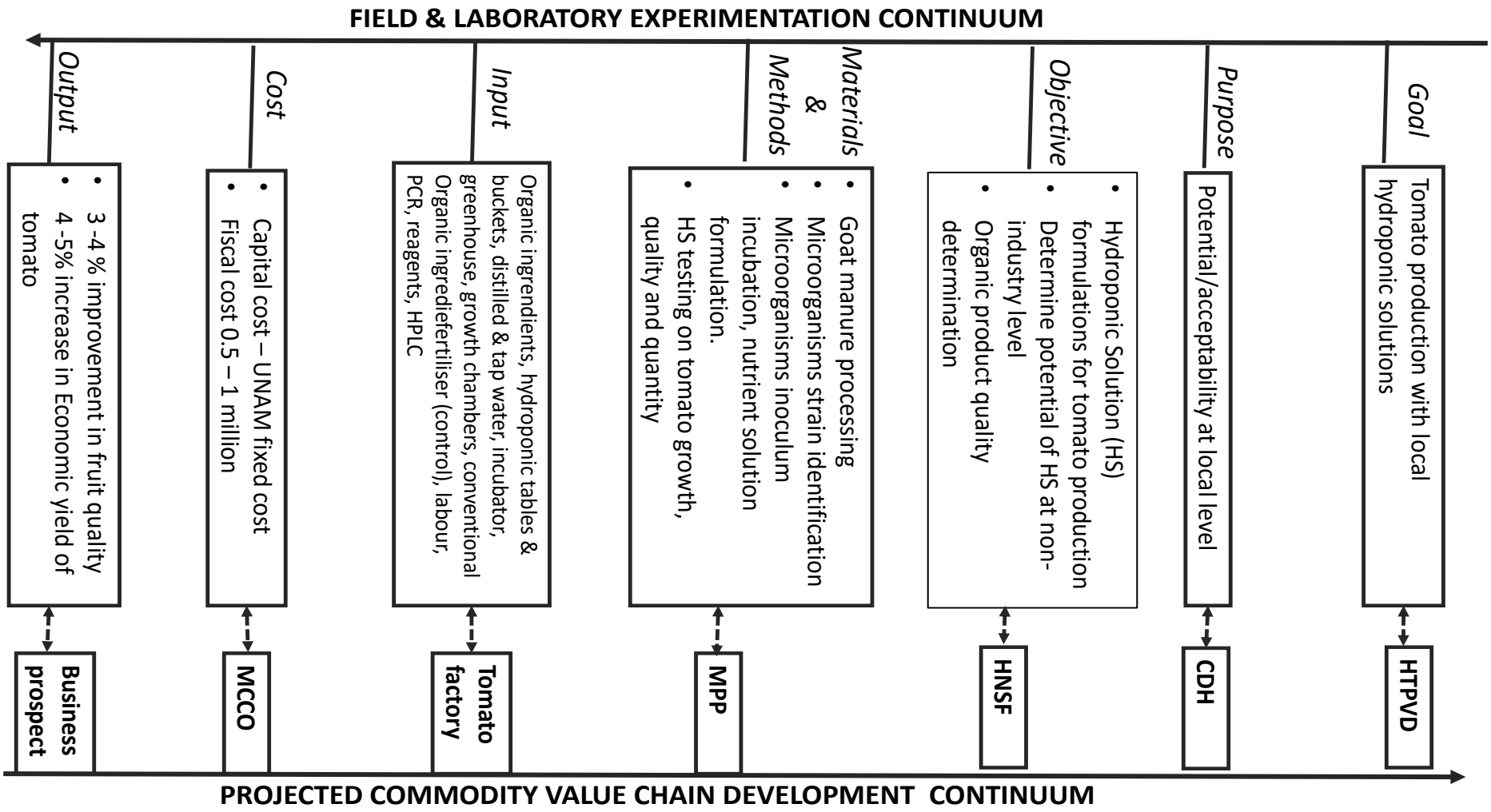



**Figure 1 Log-frame based conceptual framework (Adopted from Akundabweni-Personal Communication 2017). MCCO – Medium Cost Capital Outlay (less than 1 million NAD; MPP – Material Preparation & Production; HNSF – Hydroponic Nutrient Solution Formulation; CDH – Coastal Desert Horticulture; HTPVD – Hydroponic Tomato Product Value chain Development**

## REFERENCES


1. Akundabweni, L. (2017). Personal communication. Ogongo Campus, University of Namibia
2. Binswanger, H.P. (1981). Attitudes toward risk: Theoretical implications of an experiment in rural India. *Economic Journal* 91(364): 867–90.
3. Doppler, W. (1980). *The economics of pasture improvement and beef production in semi-humid West Africa*. German Agency for Technical Cooperation (GTZ).
4. Mendelsohn, J. (2006). Farming systems in Namibia; A publication for the Namibia National Farmers Union (NNFU). *Published by RAISON*, Windhoek, Namibia.
5. Mowa, E. (2015). Organic manure for vegetable production under hydroponic conditions in arid Namibia. International Science & Technology Journal of Namibia, 2015, 5:3-12.
6. Osman, K.A., Al-Rehiayani, S. M., Al-Deghairi, M.A., & Salama, A.K. (2009). Bioremediation of oxamyl in sandy soil using animal manures. *International Biodeterioration & Biodegradation*, *63*(3), 341-346.
7. Sanchez, P.A. (2002). Soil fertility and hunger in Africa. *Science (Washington)*, *295*(5562), 2019-2020.
8. Shinohara, M., Aoyama, C., Fujiwara, K., Watanabe, A., Ohmori, H., Uehara, Y., & Takano, M. (2011). Microbial mineralization of organic nitrogen into nitrate to allow the use of organic fertilizer in hydroponics. *Soil science and plant nutrition*, *57*(2), 190-203.

9. Van der Ploeg, R.R., Bohm, W., Kirkham, M.B. (1999). On the origin of the theory of mineral nutrition of plants and the law of the minimum. *Soil Sci. Soc. Am*. J. 63, 1055–1062.
10. Voors, M., Demont, M., & Bulte, E. (2016). New experiments in agriculture. *African Journal of Agricultural and Resource Economics Volume, 11*(1), 1-7.

# CHAPTER 2: LITERATURE REVIEW

## 2.1 Review of farming systems

There has been continuous reduction in arable land worldwide over the years and is expected to continue further shrinking to 0.15 ha per capita in 2050. Similarly, the per capita water availability has dropped from 5600 cu meters to 1200 cu meters worldwide, which could slip further to the water-deficient level i.e. below 1000 cu meters per year in years to come (Sheikh, 2006). Therefore, to meet growing demands for food there is a growing need for farming systems adaptable to increasing food production while maintaining the ecosystem stability and rehabilitation of the environment (Sheikh, 2006). This is because the current intensive agricultural practices are negatively affecting sustainability of soils, our support systems (Pimentel *et al*., 2005; Montgomery, 2007).

Current farming systems producing food for the world depend on commercial synthetic fertilizers and have kept food demands of the world population in check, consequently without commercial fertilizers the food produced now could not be produced Borlaug and Doswell (1994). Therefore, the use of commercial fertilizers has increased steadily in recent years to meet food demands for the increasing population, rising almost 20-fold to the rate of 100 million tons of nitrogen per year (Glass, 2003). However, the use of these fertilizers is actually not balanced in that, Africa is the least in terms of using fertilizers for food production compared to other continents (Heffer & Prud'homme., 2010) which results in less food production to meet Africa's demands. Partly because of climate change impacting the rain-fed farming systems of sub-Saharan Africa (SSA), leading to chronic food shortages and poverty which may hamper Sustainable Development Goals set for

Africa. Fertilizer costs that are unaffordable by most in Africa lead to reluctance in making such investments when the outcomes seem so uncertain due to climate change especially on the livelihoods of resource poor farmers who rely mostly on rain-fed agriculture (Cooper *et al.,* 2008). Furthermore, current fertilisers are designed to be used by all, when in reality poor farmers would need a different approach. Moreover, fertilisers are generally expensive in Africa on one hand, meanwhile prices for produce is mostly less, hence less interest to sustain it by most poor though agriculture is the backbone for most rural communities in Africa. Historically, Africa used relatively more fertilisers in the 1970s and 1960s but stalled in the 1980s, whereas other continents continued to intensify fertiliser use (Morris, 2007). However, there remain concerns of fertilizers' negative impacts on soil systems even where they have been intensively used. Therefore, alternatives continue being sought, which will be friendly to the environment (soil) yet productive in terms of food.

In Namibia, horticultural production is only made in smaller parts of the country with good soils and sufficient rains/water, considering only part of the Central Namib and the Northern regions of the country receive sufficient rains/water and have fertile soils that could be brought under cropping. Therefore, inhabitants of the rest of Namibian settlements are only beneficiaries of surpluses produced by other regions of the country and imports from South Africa (Mendelsohn, 2006). This typifies Liebig's law of the minimum, whereby the limiting factor is the total involvement of all potential farmers in Namibia to contribute towards total horticultural production for the country. Therefore, there is a compelling need for alternative means of horticultural production in order to engage all potential farmers into full production even in the face of climate change effects.

Such alternatives include the use of water saving techniques such as drip irrigation (Sneh, 1995) and hydroponics (dos Santos, 2013), which are systems designed for arid countries like Namibia.

## 2.2 Review of hydroponics and plant performance

### 2.2.1 Definition and uses

Hydroponics is a method of growing plants using mineral nutrient solutions in water without soil (Resh, 2012). It is a technology designed for arid countries like Namibia, where it is advantageous over soil based vegetable production in that it conserves water (Resh, 2012), avoids soil-borne diseases, makes vegetable production possible even in areas with poor soil fertility (Savvas, 2003) and generally enhances vegetable production (Sgherri *et al.*, 2010) and quality (Machado *et al.*, 2006).

### 2.2.2 Applications and effects of hydroponics in vegetable cultivation

The standard method of growing vegetables throughout the world is on soil. However, soil based vegetable production has varied challenges such as; soil-borne diseases, constant water supply for watering plants, particularly in the presence of climate change, etc. (Jensen, 1997). These challenges have been addressed by the advent of hydroponics, a soilless cultivation method for producing horticultural crops in water (dos Santos *et al*, 2013).

According to Jones (2016) hydroponics have the following advantages over soil based productions:

- Cultivation of crops is possible even in the absence of fertile soils or uncultivable soils.
- The labour for working out the soil, and other customary ways associated with soil based cultivations are gotten away with.
- Great and high yields are practical, which makes hydroponics more advantageous and economically beneficial even in urban areas.
- It is possible to conserve water and nutrients considering the system contains these until the next change, which results in less pollution of water systems.
- There are no soil associated plant diseases because hydroponics has the advantage in that they are a closed to soil contacts.
- The grower gets full control of the growing conditions when using hydroponics more than when using soil based techniques.
- Nutrient solution with mineral nutrients gets to be used for a long time before change considering they are contained in a controlled media.
- It is possible to produce horticultural vegetables even in towns and cities.

Over the years horticultural production has been realized via hydroponics for a varied range of crops (Jones, 2016). Knott (1966) found that when tomato was grown hydroponically in desert greenhouses, it yielded five times more than when grown in good open fields. Furthermore, tomatoes have been successfully produced hydroponically in Germany (Auerswald, 1999), the United States of America (Rippy *et al.,* 2004; Kubota *et*

*al.*, 2012), Argentina (Premuzic *et al.*, 1998), Japan (Sakamoto *et al.*, 1999), South Africa (Maboko *et al.*, 2011) and in Namibia (Mwazi *et al.*, 2012; Mowa, 2015).

Regionally, Linsley-Noakes *et al* (2004) successfully produced strawberries using hydroponics in South Africa. At the national level in Namibia, commercial production of vegetables using hydroponics has been limited to a handful including Agrigro Namibia that uses hydroponics for tomato production in Okahandja (Anton, personal communication, 2016), and community projects such as the Lironga Eparu HIV-AIDS project at Rundu that have been involved in hydroponic gardening since 2005 (Nampa, 2005). Furthermore, the German Government made available money for the construction of hydroponic nurseries at three centres in Windhoek with the following organizations benefiting from the contributions: the PEACE Centre, the AIDS Care Trust and the Ombili Community Centre in Okuryangava (Philander, 2007). Elsewhere in Namibia, hydroponic vegetable production was introduced in Keetmanshoop where new agricultural technologies suitable to the environment of Keetmanshoop were being sought after to produce vegetables, culinary herbs fruits and animal fodder (UNDP, 2015).

At the regional level in Namibia, in the Erongo region, commercial passive hydroponics have been successfully used for *Solanum lycopersicum* (tomato) production (Mowa, 2015), *Brassica oleracea var. capitate* (cabbage), *Brassica oleracea var. italica* (broccoli), *Lactuca sativa* (lettuce) and *Beta vulgaris* subsp. *cicla var. flavescens* (spinach) (Mwazi *et al.*, 2010; Mwazi, 2012). Vegetable production and promotion hydroponically

has been done extensively at the Erongo region's coastal town of Henties Bay, Erongo region, to community members but has seen less upscaling due to fertilizer affordability (Omoregie, 2013). Moreover, still in the Erongo region, hydroponics gardening was introduced in areas such as Usakos and Spitzkoppe by AREVA mining Namibia in order to help stimulate economic growth where training and materials including crates, plastic, seeds and fertilizer were given to the communities to set up the gardening schemes (CMN, 2012); however, sustainability concerns arose in terms of fertiliser costs, among others.

Despite practical evident advantages of hydroponics, the major disadvantage of this system in its typical form is capital intensiveness, mainly due to fertilisers and sophisticated instrumentation such as air pumps, reservoirs, water flow tubes, nutrient pumps, growing trays and growing mediums (Schnitzler *et al.*, 2004; Sheikh, 2006). However, there exists versions of hydroponic systems which are based on all cheaper materials in exception of fertilisers which remain without cheaper alternatives. For example, hydroponics were simplified at the Universidad Nacional Agraria La Molina where low-income families were assisted to produce fresh vegetables using basic soilless and raft culture systems (Resh, 2012). In Florida, low cost passive outdoor hydroponic gardens have been demonstrated successfully for over 6 years, where vegetables such as tomato had high yields and were even recommended for small scale farmers for upscale (Tyson, 2002). Passive hydroponics are non-circulating and do not depend on aeration, nutrient pumps and other sophisticated instrumentation unlike active hydroponics. Furthermore, another simple non-circulating hydroponic system was successfully employed to produce winter and spring crops such as *Lactuca sativa* (lettuce) in Florida

in 1997 (Fedunak & Tyson, 1997). In Thailand, using low-cost local resources to build hydroponics led to high yields of *Solanum lycopersicum* (tomato) (Thippayarugs, 2001). Moreover, in Pakistan, a non-circulating hydroponics system was used to successfully produce *Solanum lycopersicum* (tomatoes) (Shah *et al.*, 2011). In all these cases local, cheap materials replaced those used at high costs in typical hydroponic systems, yet fertiliser input remained expensive considering it was the only input not replaced in comparison to the typical conventional hydroponic systems.

The cost of fertilisers has been particularly pronounced in hydroponic systems which rely on using inorganic fertilisers. To this day the reliance on mineral fertilizers and pesticides still continue worldwide because of increased yields to meet food demands, even though serious impacts on public health and the environment are evident (Pimentel et al., 2005). Therefore, even though hydroponics can be an important crop production solution in water limited countries like Namibia, not all communities can afford to practice horticulture using conventional hydroponics. In fact, simplified hydroponics have been introduced in Namibia since 2005 (Nampa, 2005), where growing beds were constructed using timber planks and gravel or plastic sheeting and Styrofoam, about N$200 per table. This simplified hydroponic system has been in use in few areas in Namibia, including Rundu, Okahandja, Windhoek, Keetmanshoop, Usakos and Henties Bay in the Erongo region, where this technology has been demonstrated for years to the local community by Sam Nujoma Marine & Coastal Resources Research Centre (SANUMARC). Yet, upscaling by local communities has always been a challenge due to affordability of the fertilisers. This

situation can partly be addressed by development of cheaper alternative fertilizers such as using manure for hydroponics.

Traditionally, organic nutrient solution for hydroponics has not been feasible, despite the similarities in plant growth when either conventional or organic fertilizer is applied on soil (Liang *et al.,* 2014). It was not until the early 1990s when liquid organic nutrient solutions for hydroponics were introduced. Challenges with these liquid nutrient solutions emerged, such as organic fertilizer being unsuitable to plant growth (Garland *et al*., 1997; Liedl, *et al.,* 2004) because nitrogen in organic sources is predominantly organic, hence unusable by plants. The forms of nitrogen absorbed by plants are nitrate and ammonium (Strayer *et al*., 1997). Therefore, the nitrogen in manure requires to be mineralised prior to use by plants hydroponically. Several studies including Garland *et al* (1997) and Shinohara *et al* (2011) have since demonstrated that using microorganisms to degrade organic nitrogen in organic sources such as manure results in nitrates and ammonium production which in turn are used for plant production.

Recently, there have been successful hydroponic production of tomato and other vegetables using organic nutrient solutions processed by microorganisms. Chinta *et al* (2015) found that using organic nutrient solution made from corn steep liquor not only made successful *Lactuca sativa* (lettuce) production, but also reduced root rotting. Fujiwara *et al* (2012) found the same effect of reducing root rotting was also observed in tomato plants when organic nutrient solution was used. Furthermore, plant wilting was also reduced in this case. Chinta *et al* (2015) found that using organic nutrient solution made from corn steep liquor provided resistance to air-borne disease in vegetables.

Shinohara *et al* (2011) found that using organic nutrient solutions made from fish-based fertiliser or corn steep liquor hydroponically, produced tomato yield similar to those produced from conventional nutrient solutions. From the same organic nutrient solutions, Shinohara *et al* (2011) further established that when *Lactuca sativa* (lettuce) was grown, the organic system produced significantly greater and fresh *Lactuca sativa* (lettuce) head weight than in the conventional system.

Preliminary studies in Namibia, the only African country to begin research into organic prospects in passive hydroponics, found that when goat manure was used hydroponically after processing in compost piles, *Beta vulgaris* subspecies *cicla var*. *flavescens* (spinach) production was made possible through production was less than conventional hydroponics (Mowa, 2015). Passive hydroponics have been used in Namibia since the early 2000s. The organic nutrient solution used in this regard was not monitored in terms of processing of the manure. Furthermore, the nutrient levels in the organic nutrient solution were not measured and quantified to determine levels that would enable optimal plant growth and yield. Goat manure has not been used hydroponically, anywhere in the world until the preliminary studies done by Mowa (2015).

### 2.2.3 Nutrient use efficiency and plant biomass

In order to achieve desired crop yields, fertilizer input must be optimized with maximum potential use available to maximize yields, yet in most cases, with so much fertilizer input the yield returns are low due to nutrient use efficiency. Nutrient Use Efficiency (NUE) is defined as the ability of a cultivar to acquire nutrients from growth medium and to or

utilize them in the production of plant biomass or plant reproduction material i.e. seed and fruits (Blair, 1993). Nutrient use efficiency can be described as partial factor productivity, which entails kilogram (kg) crop yield per kilogram (kg) nutrient applied. NUE can also be described as agronomic efficiency which is kilogram (kg) crop yield increase per kilogram (kg) nutrient applied. NUE can further be described as apparent recovery efficiency, which is kilogram (kg) nutrient taken up per kilogram (kg) nutrient applied. Moreover, NUE can be described as crop removal efficiency which is the removal of nutrients in harvested crop as percentage of nutrients applied.

NUE is important to establish the ability of cultivars to absorb and use nutrients for maximum performance, which is important for developing countries mainly because there is need for enhancement of crop yields at a reduced cost (Baligar & Bennett., 1986).It is important to increase NUE in plants in order to enhance the crop yield and quality, thereby reducing nutrient input cost whilst receiving maximum yields possible, because the economic benefit to cost ratio has a large influence on farmer adoption of farming systems available. Therefore, application of fertilizer has to target both high yields and nutrient efficiency to benefit farmers, society, and the environment alike (Roberts, 2008).

Generally, when nutrients in the growth media are efficiently used by plants, vegetative and reproductive plant performances are enhanced. In fact, when nutrient availability increases in a growth medium, most plant species increase productivity and yield therefore, nutrient up-taken will equal nutrient lost in such a medium (Aerts, 1990). Therefore, slow and controlled release fertilizers have added advantages in increasing

plant nutrient use in that the nutrients are availed to plants for prolonged periods allowing plants sufficient time to uptake the available nutrients. In the case of nitrogen fertilizers applied on soils, it is estimated that only less than 50% is used by plants efficiently whereas the remaining 50% or more is lost through leaching, fixation and run-off resulting in environmental degradation. This plant nutrient use efficiency is influenced by a varied range of factors such as light, temperature, water, growth media and fertilizer material (Baligar *et al.,* 2001). Indeed, it has been established that improving NUE of plant production systems requires that nutrients used by plants from the total available nutrient pool be considered because that would help to determine the economic impact on yield in relation to applied nitrogen inputs and crop accumulation of nitrogen. This same principle applies to available nitrogen derived from organic nitrogen sources such as legume green manures, cover crops and animal manures. Indeed, challenges with nitrate from animal manures can be equal or greater than from inorganic nitrogen fertilizer when the available nitrogen supply from either source exceeds crop demand by similar amounts for comparable time periods (Cassman *et al.,* 2002).

Moreover, NUE in plants has been found to be also related to shoot demand and dry matter production of nutrients absorbed because levels of nutrients available influence the total dry matter accumulation which in turn influences nutrient demand and subsequently, the yield (Baligar & Duncan., 1990). In fact, it has been established by Zotarelli *et al* (2009) that above-ground plant dry weight at harvest can indicate nutrient use efficiency of such plants from the available pool of nutrients. Moreover, there is need for total plant biomass as opposed to only root or above-ground biomass when relating to NUE (Aerts, 1990).

Total plant dry biomass in tomato has been established when using fertilizers such as pig manure (Atiyeh *et al.,* 2000) and synthetic fertilisers (Zotarelli *et al.,* 2009). Furthermore, it has been proposed that a nutrient-specific analysis, considering the biology of each mineral nutrient rather than grouping plant resources as a whole, is more appropriate than general models in understanding plant responses to nutrient availability (Rubio *et al.,* 2001). Nutrient use efficiency has, however, not been established with goat manure using plants like tomato.

The use of hydroponic nutrient culture in Namibia is not new. It is particularly an interesting practical technology that is appropriate for the Coastal Desert Horticulture within the Erongo region that lies in the Namib Desert ecology. In the latter, the Benguela cold current along the coast delivers cold dry air lacking moisture (Goudie & Viles, 2015). Combined with infertile sandy soils that are low in nitrogen, the coastal area is essentially unsuitable for rainfed crop production. Thus, hydroponic crop production could significantly increase protected crop production. Furthermore, hydroponic crop production is said to increase crop quality and productivity (Trejo-Tellez & Gomez-Merino, 2012). Moreover, the advantages of hydroponics have been ably demonstrated in the above review. Evidence is provided that hydroponics offers advantages over soil-based vegetable productions. Research is, however, warranted to formulate an affordable simplified hydroponics innovation based on local resources such as goat manure for processing to provide organic nitrogen nutrient that efficaciously promotes high value tomato production. Nitrogen is the most limiting in organic hydroponics (Shinohara *et al.,* 2011). Therefore, the current study investigated the efficacy of an organic hydroponic

nutrient solution derived from goat manure, on plant performance. The organic nutrient solution was processed from goat manure using a specialized culture of microbes.

## 2.3 Review of hydroponic nutrient solution formulations

### 2.3.1 Nutrient solution formulation

There are 16 elements required by plants for growth, and they include: carbon (C), hydrogen (H), oxygen (O), phosphorus (P), potassium (K), nitrogen (N), sulfur (S), calcium (Ca), iron (Fe), magnesium (Mg), boron (B), manganese (Mn), copper (Cu), zinc (Zn), molybdenum (Mo), and chlorine (Cl) and Nikel (Hochmuth & Hochmuth, 2001; Salisbury & Ross, 1992). Six of these (N, K, Ca, Mg, P, and S) are mineral nutrients required in more quantities than the remaining nutrients which are micronutrients or trace elements (B, Cl, Cu, Fe, Mn, Mo, and Zn) required in small quantities (Mayland & Wilkinson, 1996). Beneficial elements in addition include sodium, silicon, vanadium, selenium, cobalt, aluminium and iodine among others, stimulate plant growth. They also can mitigate or reduce the toxic effects of other elements, or may also play an almost same role of the essential nutrients but in a less specific mode (Trejo-Téllez & Gómez-Merino, 2012). For most part of the hydroponic nutrient solution, the most important composition considers only nitrogen, phosphorus, potassium, calcium, magnesium and sulphur; and they are supplemented with micronutrients.

The nutrients in the solution require adjustments to satisfy Liebig's law of the minimum (Douglas, 1985) and can come from different sources such as by-products from fish waste, animal manure, or normal nutrients. Nevertheless, generally acceptable concentrations for

nutrient solutions exist, with minimum and maximum concentration ranges for most plants being somewhat similar (Jones, 2016). Inadequate or excessive amounts of these nutrients result in poor crop performance (Hochmuth & Hochmuth, 2001). Therefore, in order to manage these nutrients, an understanding of the nutrient solution concentrations for the various nutrients required by plants is needed. Hydroponically, there are two methods to supply nutrients to the crop: 1) premixed products, or 2) grower formulated solutions (Hochmuth & Hochmuth, 2001). Premixed products exist in various commercial fertilisers, which are already mixed and balanced to levels required by plants. Grower formulated solutions on the other hand uses different sources of elemental nutrients individually brought together to make up a final solution as per the requirement of the plant to be applied on (Hochmuth & Hochmuth, 2001).

Other parameters for consideration in nutrient solution formulation include pH of the nutrient solution, the osmotic potential, the oxygenation in the nutrient solution, temperature, formulation and preparation considering the composition of both micro and macro elements (Trejo-Tellez & Gomez-Merino, 2012). Nutrient solution pH influences the availability and plant uptake of all essential plant nutrients. All essential micronutrients, except molybdenum, become less available as the pH increases (Warncke & Krauskopf, 1983). Conductance on the other hand indicates total soluble salts within the solution. - All soluble ions or nutrients such as nitrate, ammonium, potassium, calcium, magnesium, chloride and sulfate contribute to the soluble salt content of a nutrient solution (Warncke & Krauskopf, 1983). The above-mentioned factors have been found to be optimal when maintained as: pH at 7.7 – 8.4, 35ºC temperature, 6.5 mg/L Dissolved

Oxygen or vary some parameters like temperature at 25ºC and aeration to culture the same nitrifying microorganisms (Shinohara *et al.,* 2011).

Formulations of hydroponic nutrient solutions based on organic sources was not possible since the inception of hydroponics due to the fact that nitrogen in organic sources such as manure, was not available in plant accessible forms such as nitrate and ammonium (Garland *et al*., 1997). Production of nitrates from organic sources for plant use was made possible by the introduction of microbial inoculations in different bioreactor tanks through a succession of steps (Garland *et al.,* 1993; Mackowiak, 1996). Further strides have been made since then, with Shinohara *et al* (2011) recently managing to use microbes from different sources to mineralise organic nitrogen into nitrates in the same media.

The resulting nutrient solutions for crop production have already shown advantages such as efficacy for both soil-borne and air-borne disease control in vegetables such as *Lactuca sativa* (lettuce) and cucumber (Chinta *et al.,* 2015). This development comes as a relief to communities with less financial abilities to access hydroponics via organic sources for plant nutritional requirements. This is considering that the use of conventional fertilisers for agriculture is relatively expensive worldwide (Dopler, 1980; Osman *et al.,* 2009) and in particular in Africa (Sanchez, 2002). The formulation and preparation may be commercial and even local. For the latter, its quality may not be as guaranteed as that of a commercial formulation purchasable from agro-vet outlets but it is said to be good enough for practical purposes in hydroponics (Shinohara *et al.,* 2011). There are various protocols for locally formulating hydroponic solutions among which include the method of Shinohara *et al* (2011), which involves using microorganisms to degrade organic

nitrogen into a nutrient sauce. Thus, a further review of the ingredients required for the formulation is as presented below (sections 2.3.2 & 2.3.3).

### 2.3.2 Nitrifying microorganisms

After the success of Shinohara *et al* (2011) in using nitrifying microorganisms to degrade organic nitrogen into nitrates, follow-up studies such as those of Hu & Qi (2013) confirmed the successful use of microbes in nitrification of other organic sources. Nitrification consists of two consecutive oxidation steps: nitritation and nitratation done by two main groups of bacteria — ammonia-oxidizing bacteria (AOB) and nitrite-oxidizing bacteria (NOB). Most AOBs belong to the beta-subclass of *Proteobacteria* and four genera belonging to this lineage are *Nitrosomonas* (with *Nitrosococcus mobilis*), *Nitrosospira*, *Nitrosolobus*, and *Nitrosovibrio* (Woese *et al.*, 1984; Pommering-Röser *et al.*, 1996). NOBs on the other hand consist of the *Proteobacteria* genera *Nitrobacter*, *Nitrococcus* and *Nitrospina.* The genus *Nitrobacter* has four species *N. winogradskyi*, *N. hamburgensis*, *N. vulgaris* and *N. alkalicus* (Daims, 2001). The genera *Nitrococcus* contains *Nitrococcus mobilis* whereas the genera *Nitrospina* contains *Nitrospina gracilis.* The genus *Nitrospira* consists of *N. marina* from ocean water and *N. moscoviensis* (Daims, 2001). These microorganisms can be cultured and stored for use in water and soil media to degrade organic nitrogen (Willuweit *et al.*, 2008). All they require is an ecosystem with favourable conditions for them to function in their specific diversity (Saijai *et al.*, 2016). But culturing has a major disadvantage in that many nitrifying bacteria are not cultivable. It is estimated that in seawater samples at best only 0.1% (Kogure *et al.*, 1980), in freshwater only 0.25% (Jones, 1977), in soil samples 0.5% (Torsvik *et al.*, 1990), and in

activated sludge 15% (Kämpfer *et al.*, 1996) of the indigenous bacteria could be cultivated. Traditionally, *Nitrosomonas* and *Nitrobacter* were thought to be the only microbes responsible for nitrification in wastewater treatment plants based on the experience that *Nitrosomonas* and *Nitrobacter* species can be isolated from every activated sludge. It was only when *Nitrobacter* was not detected in an aquarium, yet there was nitrification, that it was considered that there are other unculturable yet important microbes doing the same job. Therefore, microbial ecology needs cultivation-independent tools to quantify bacteria directly in environmental samples (Daims, 2001).

It has been established that under 40% of soil water filled pore space decreases abundance of AOB than over 40% (Barrena *et al.,* 2017). This abundance is influenced by pH of soil, with acidic soils (4.0/5.4) negatively affecting abundance of AOBs, whereas neutral pH positively influences AOBs (Duncan *et al.,* 2017). Furthermore, AOA may affect N cycling more in soils receiving animal manures, whereas AOB are functionally more important in chemically fertilized soils (Zhou *et al.*, 2015). Moreover, abundances of archaeal 16S rRNA and amoA genes have been found to be positively correlated with soil nitrate, N and C contents (Rughöft *et al.,* 2016), whereby changes to physical properties of soil determine nitrifying and ammonifying capacity and, therefore, mineral nitrogen content. Maximum content of nitrate and mineral nitrogen and the biggest nitrifying capacity are at soil temperatures of +15°C, whereas temperature at -4°C have recorded the lowest abundances of NOBs (Wertz *et al.,* 2013). Moistening conditions optimal for developing nitrifying and ammonifying bacteria are formed at soil moisture of 20-25% (60-75% WFC) (Evdokimova *et al.,* 2016). Analysing and studying these nitrifying bacteria can be done using functional genes. Functional genes have been found to be good

molecular markers for studying the diversity within functional groups (Poly *et al*., 2008; Calvo *et al.,* 2005). For nitrification, the *amo*A gene encoding ammonia monooxygenase is being used for microbial diversity and phylogenetic characteristics of soil and water AOB communities (Rotthauwe *et al*., 1997; Purkhold *et al.*, 2000). In Nitrobacter, the oxidation of nitrite to nitrate is performed by the nitrite oxidoreductase (NXR), encoded by the nxr operon (Starkenburg *et al*., 2006) previously called nor (Kirstein & Bock, 1993).

Due to ability to reveal the previously hidden diversity of microscopic life, metagenomics offers a powerful lens for viewing the microbial world that has the potential to revolutionize understanding of the entire living world (Marco, 2011). Metagenomics refers to the idea, that a collection of genes sequenced from an environment could be analysed in a way analogous to the study of a single genome (Handelsman, 2004). In other words, in sequencing nitrifying microorganisms, for example, one would expect that several genomes from a given environment are involved. Thus, such data may be looked at in a similar manner as for a single genome sequencing.

The need to incorporate metagenomics has been driven by the inability to culture the majority of microbes from an ecosystem and the logistics of using a myriad of media and culturing conditions to capture those that can be grown *in vitro* (Marchesi, 2012). Moreover, the advent of Next Generation Sequencing (NGS) technologies allow us to sequence DNA and RNA much more quickly and cheaply than the previously used Sanger sequencing due to the inexpensive production of large volumes of sequence data (Metzker, 2010). The results can thereafter be put through binning where methods such as Basic

Local Alignment Search Tool (BLAST) are used to search for phylogenetic markers or similar sequences in existing public databases (Huson *et al.*, 2007). Indeed, this study in view of the above advances used the NGS technology to enable the sequencing of the DNA in the raw materials used in the current study.

### 2.3.3 Goat manure as source of plant required nutrients

Goat manure, like other animal manures, has essential elements required for plant growth including nitrogen, though such composition may vary geographically depending on diet that goats feed on (Chadwick et al., 2000; Moreno-Caselles *et al.*, 2002. In fact, when compared to other animal manures, goat manure has been shown to release high concentrations of plant required nutrients more than cattle manure, horse manure and others for except poultry manure (Azeez & Van Averbeke, 2010). Table 1 & 2 illustrate the superiority of goat manure (Bandara & Tennakoon, 2003).

**Table 1: The comparison of goat manure macro nutrient composition with other sources**

| | N | P | K | Mg | Ca |
|---|---|---|---|---|---|
| Cattle manure | 1.2-1.9 | 0.2-0.5 | 0.5-1.1 | 0.5-1.8 | 1.3-1.8 |
| Goat manure | 2.2-3.4 | 0.3-0.7 | 1.5-2.5 | 0.4-0.8 | 1.5-2.4 |
| Pig manure | 1.0-2.0 | 0.6-0.9 | 0.4-0.9 | 0.4-0.6 | 1.0-1.5 |
| *Chicken manure | 3.7 | 1.4 | 1.7 | 0.7 | 2.6 |

Source: (Bandara & Tennakoon, 2003; *Azeez & Van Averbeke, 2010)

Even though animal manure varies in Nitrogen content due to the type of feed given to animals, generally total nitrogen content in goat manure for example, is about 2,3% of dry weight manure. The total nitrogen requires to be processed in order to get plant available

nitrogen (nitrate, ammonium), of which mineralizable nitrogen is about 20% of total nitrogen (Munoz *et al.,* 2004; Pratt & Castellanos, 1981).

**Table 2: The comparison of goat manure micro nutrient composition with other sources**

| Organic material | Fe | Mn | Cu | Zn | B | Cd |
|---|---|---|---|---|---|---|
| Cattle manure | 690-1518 | 167-389 | 24-40 | 128-183 | 13-30 | 2-3 |
| Pig manure | 1020-1990 | 180-207 | 45-48 | 186-575 | 4-13 | 2-3 |
| Goat | 1449-2174 | 246-505 | 20-38 | 112-184 | 29-66 | 2-3 |
| *Chicken | 1880 | 759 | 99 | 545 | - | - |

Source: (Bandara & Tennakoon, 2003; * Azeez & Van Averbeke, 2010)

On soil based cultivation, goat manure can be processed before addition to the soil or it can be applied directly on soil for microorganisms in the soil to naturally process manure in soil. This is because most of the nutrients, particularly nitrogen, become available through mineralization, the conversion of nutrients from organic to inorganic (plant-available) forms. In contrast to nitrogen, phosphorus availability from manure is often high (more than 70 percent) because most of the P is inorganic and readily available. Nitrogen mineralization is associated with the ease with which microorganisms decompose the carbon in the manures. Nitrogen available from manures depends on the nitrogen content, and the stability of the nitrogen or the ease with which it is mineralized. Aged manures lose nitrogen by volatilization of ammonia; the remaining nitrogen is more stable or resistant to mineralization (Pratt & Castellanos., 1981). Therefore, it is advisable to work with manure that is not too aged or composted since the composting process produces lower nitrogen from goat manure, thus the ideal manure management for optimizing nitrogen availability is to apply the manure and mix it with the soil as soon as possible. The longer the time between production and incorporation, the lower the available nitrogen (Pratt & Castellanos., 1981).

Goat manure world-wide has been applied directly to the soil where it was processed to avail plant required nutrients. For example, In Sri Lanka, direct addition of goat manure to soil for coconut plantation resulted in the supply of sufficient plant required nutrients for the coconut plantation such that there was a significant increase in nut yield (Tennakoon, 1990). In an organically managed field trial, in the United States of America, it was established that over a three-year period, yield of corn was always higher in a raw dairy manure treatment compared with a compost of dairy manure and leaves (Reider *et al*., 2000).

Furthermore, in Nigeria, goat manure collected from local villages was applied directly on top soil without composting first at the onset of the rainy season. Micro-organisms and macro-organisms such as termites, beetles or arthropods decomposed the manure and released sufficient rates of nitrogen (N), phosphorus (P) and potassium (K) from goat manure and that decomposition was faster on biologically crusted soils than on sandy soils. The quantities of N, P and K released from goat manure during the rainy season were up to 10-fold larger than the annual nutrient uptake of pearl millet (*Pennisetum glaucum* L.) that was grown in the area (Esse *et al.*, 2001). Additionally, still in Nigeria, direct incorporation of goat manure on soil for nutrient improvement resulted in sufficient release and supply of plant required nutrients that improved growth and yield of *Piper nigrum* (pepper) (Awodun *et al.*, 2007).

Elsewhere in Kenya, goat manure applied directly to the soil was helpful in not only providing nutrients (Kihanda *et al.*, 2004), but also helped with reducing soil acidity **(**Gitari *et al.*, 2015).

Regionally, in South Africa, directly adding manure on soil was made where the mixture of soil and goat manure was inoculated with soil occurring microorganisms for the goat manure to be degraded. The results indicated that organic matter, total nitrogen (N), total phosphorus (P) and total carbon (C) were significantly higher in goat manure compared to cattle manure. The release phases of nitrogen from goat manure was in phases: initial rapid N release at 0–30 days; phase of constant release; 40–55 days; decline phase in N release 70–90 days and sharp increase in N release at 120 days (Azeez & Van Averbeke, 2010).

Though composting goat manure results in less nitrogen, composting will still provide useful end products for plant growth, in fact contents of calcium, magnesium, available phosphorus and available potassium were increased during composting compared to their values in goat dung before composting (Shah *et al.,* 2015). Goat manure can be composted using different materials such as *Triticum aestivum* straw (Kulcu & Yaldiz, 2007; Herbert *et al.,* 1991), *Vitellaria paradoxa* cake (Tchegueni, 2013), fresh *Calotropis gigantea* leaves (Shah *et al.,* 2015), *Pinus* bark (Mupondi *et al.,* 2006) and many other materials. The composting period varies from 30 days to 180 days (Kulcu & Yaldiz, 2007). Abu-Bakar & Ibrahim (2013) further suggest that indigenous microorganisms can be added to the compost pile to hasten goat manure degradation during composting. Mupondi *et al* (2006) add that microorganisms can be introduced to the composting process as they established that the introduction of Effective Microorganisms (EM) to goat manure composting resulted in composting efficiency (Mupondi *et al.,* 2006). Though composts result in less nitrogen, it has been shown that incorporation of N rich, low (15 or less)

carbon to nitrogen (C: N) ratio residues of fresh manures leads to rapid mineralization and a large rise in soil mineral N (Qian & Schoenau, 2002). Therefore, mineralization is high from fresh material than it is from composted materials (Eghball *et al.,* 2002). Whether processed or not before application to the soil, the value of manure is so well-recognized that some farmers keep livestock primarily for the benefits such as plant nutrients, important organic matter to the soil, maintaining its structure, water retention and drainage capacity (Sansoucy, 1995).

## 2.4 Review of organic vegetable quality

Vegetable quality involves both subjective evaluation (taste, flavour, demand, as given by consumers) and quantifiable evaluation based on product orientation (Gruda, 2005). Subjective evaluation has led to a state where consumers are now prepared to pay more for organic fruits and vegetables based on their perception that organic foodstuffs have better quality than conventionally grown ones (Lester, 2006). Some of the factors considered in vegetable quality are health-enhancing nutrients like antioxidants and polyphenolic compounds (such as flavonoids; phenolic acids), vitamins (such as ascorbic acid, vitamin E, and carotene (Scalbert *et al.,* 2005, Zaller, 2007). The concentration of these in fruits is known to be influenced by fertilizers (Toor *et al.,* 2006). Furthermore, fertilizers also influence yield and fruit size (Amiri, & Fallahi, 2009).

The rise of healthy lifestyle awareness spurred by healthy foods in modern times has led to food reforms which purports that people who eat foods produced the natural way will have healthy lifestyles compared to those eating foods produced using industrialized

production methods, depending on technologically led systems, for example genetically modified foods (Lockeretz, 2007). Even though industrialized production methods meet food security demands, such methods have clearly shown severe limitations such as a worldwide contamination of the food chain and water by persistent pesticide residues, and reduced nutrient and flavour quality contents through low-cost intensive food production and/or processing (Lairon, 2011). As a result, there has been a rising demand calling for vegetarian diets due to health benefits, more so for vegetables produced organically due to their perceived quality over conventionally produced vegetables (Lockeretz, 2007). In the wake of food loosing quality due to mineral fertilizers and synthetic pesticides organic vegetable farming, which involves working with the environment and not working on the environment, organic vegetable farming has been established to be environmentally friendly in comparison to conventional farming systems.

Recently there has been studies evaluating the nutritional and sanitary qualities of organic foods where it was established that organic plant products contain more dry matter and minerals (Fe, Mg), contain more anti-oxidant micronutrients such as phenols and salicylic acid, 94–100% of organic food does not contain any pesticide residues, organic vegetables contain far less nitrates (Lairon, 2011). Such findings point to the quality associated with organically produced vegetables in comparison to conventionally produced vegetables. Consumer awareness of the relationship between foods and health, together with environmental concerns, has led to an increased demand for organically produced foods. Organic food sales have increased by about 20% per year since 1990 and were estimated at $10.4 billion in 2003 on the U.S. market alone. Consumer studies have shown multiple reasons for organic preferences, including environmental and socioeconomic concerns,

opinions of taste, and the belief that organic foods are healthier (Chassy *et al.*, 2006). This superior quality in organic foods has been linked to the pace of crop physiological development and size which is relatively slower than that from conventional production systems (Benbrook, 2005). Therefore, with global value chains becoming increasingly buyer-driven, quality awareness about what they consume is leading to certain accepted standards and subsequently organic produce will receive preference (Ponte & Gibbon, 2005).

## 2.5 Review of the tomato plant

### 2.5.1 Its basic biology

Tomato is an edible vegetable belonging to the genus Lycopersicon, under the Solanaceae family. Tomato plants are either determinate or indeterminate. Determinate varieties of tomatoes grow bushy to a certain height and stop growing when they start fruit setting and will usually ripen at approximately the same time. Indeterminate varieties on the other hand, continue to grow like vines and produce fruit throughout the season until killed by frost (Maršić et al., 2005). Some of the varieties under Lycopersicon are *L. esculentum* (common tomato), *L. pimpinellifolium* (currant tomato), *Lycopersicum esculentum Mill*., and *L. esculentum var. cerasiforme* (cherry tomato) (Jones, 2007).

### 2.5.2 Tomato importance and uses

Tomato fruits are used for various purposes including canning, pulp, salad, sandwiches and sauces (Maršić et al., 2005). Currently 37% of tomatoes consumed in Namibia are imported from South Africa, despite the contribution from local soil-based farmers (NAB, 2017). This is due to poor soils covering much of Namibia for crop production leading to a situation where crop production and horticultural production are limited only to some parts of central Namibia and some Northern parts of the country (Mendelsohn, 2006).

Tomato is one of the vegetables whose quality has received much attention in recent history owing to the health benefits derived from quality tomatoes. Heeb *et al* (2005) argue that there are higher scores of quality in terms of sweetness, acidity, flavour and acceptance for the tomatoes grown with the organic fertiliser compared with the tomatoes grown using the conventional fertiliser. Hallmann & Rembiałkowska (2007) maintain that there are significant differences in the quality of tomatoes of the two cultivation systems compared, with the organic fruits characterised by a significantly higher content of total sugars, organic acids, vitamin C and phenolic compounds such as quercetin-3-*O*rutinoside, myricetin and quercetin, compared with conventional systems.

Mitchell *et al* (1991) add that vegetables levels of quercetin and kaempferol in organic tomatoes were 79 and 97% higher than those in conventional tomatoes (64.6 and 32.06 mg g-1 of DM), respectively. The levels of flavonoids increased over time in samples from organic treatments compared to samples from conventional treatments. Brandt *et al* (2011) further add that life expectancy can be increased to 17 days for women and 25 days for men by switching from conventional to organic produce without changing the amount consumed per day. In some cases, even size quality of tomato fruits produced with organic

systems were comparable with those obtained using mineral N fertilizers, indicating that organic systems can be a suitable replacement for inorganic fertilizer in tomato production (Olaniyi & Ajibola, 2008). However, other researchers such as Heeb *et al* (2006) argue that yields of red tomatoes from the organically fertilised plants are significantly lower than yields from plants that received mineral fertilisers because organic fertilisers release nutrients more slowly than mineral fertilisers which limits growth and yield in organic treatments.

Oliveira *et al* (2013) and Xu *et al* (2001) ascertain that growth is reduced in fruits from organic farming but they add that even though fruit growth and yield may be less in organic systems, such fruits from organic systems will score high in quality (titratable acidity, the soluble solids content and the concentrations in vitamin C). This superiority with organic tomatoes is due to the fact that tomato fruits from organic farming experience stressing conditions more than conventional tomatoes and that this oxidative stress leads to the accumulation of higher concentrations of soluble solids as sugars and other compounds contributing to fruit nutritional quality such as vitamin C and phenolic compounds. Mitchell *et al* (1991) maintain that tomato plant stress leads to increased phenolic compounds in tomato fruits. Pieper & Barrett (2009) on the other hand, claim that the higher moisture content in conventional tomato fruits may be responsible for lower soluble and total solids contents due to dilution, and must be considered for all other parameters measured where dilution may play a role. They reason that conventional tomato fruits may contain more water, which dilutes the concentration per unit.

Furthermore, it has been found that on average, organic vegetables and fruits contain more secondary metabolites/compounds than conventionally grown fruits and vegetables (Brandt & Mølgaard, 2001). Moreover, Carbonaro et al (2002) established that polyphenol and antioxidant levels are higher in the organic treatments compared to the conventionally grown crops, because improvement in the antioxidant defense system of the plant occurs as a consequence of the organic cultivation practice where there is less protection against damage of fruit when grown in the absence of pesticides.

Lycopene is a red pigment and major carotenoid in tomato fruit, a precursor to production of a-carotene and b-carotene. Fresh tomatoes are major sources of lycopene, leading to reduced prostate, mouth and colon cancers (Giovannucci, 1999). Shi & Maguer (2000) adds that an increased supply of lycopene from the consumption of tomatoes leads to diminished risks of cancer. The lycopene content in tomato typically ranges from 70 to 130 mg/kg and depends on the variety, geographic location, technique of cultivation, climatic conditions and degree of ripeness of tomato fruits (Shi & Maguer, 2000). Methods for determining lycopene in tomato exist, including colorimetric measurements, spectrophotometric which are less time consuming and less destructive. However, they are less accurate than the High-Perfomance Liquid Chromatography (HPLC) which allows for accurate quantification of individual pigments and separation of isomers, is also a very laborious process and requires a high level of skill to produce consistent results. Though chemicals used in extraction of carotenoids and their elution on chromatographic columns pose hazards to the experimenter and on the environment (Hyman *et al.,* 2004).

### 2.5.3 Tomato cultivation and growth requirements

Tomato cultivation has been established in both greenhouse and open-field settings. It has been found that when grown in greenhouse, tomato fruits ripen on the vines making it possible to deliver to the market freshly ripened tomatoes compared to open-field grown tomatoes that would mostly get harvested before they are ripened on the vines and would only get ripened during shipment to the customer (Abeles et al., 1992). Tomato has been found to perform poorly when grown in temperatures below 10°C because seed germination gets delayed, vegetative development is inhibited, fruit set gets reduced, and fruit ripening gets impaired. Therefore, the optimal temperature for tomato is between 18.5°C and 26°C (Jones, 2007). Light quality has been found to influence tomato plant growth more than yield. When there was excess blue light over red light, plant growth was found to be shortened, and darkened in colour. When there was excess red light over blue light, growth became soft with long internodes, yet yield was comparable between the two light qualities (Jones, 2007).

Tomato requires plenty of water but have been found to perform poorly when there was excess water due to inability of roots to function under anaerobic conditions. This leads to poor growth, lower fruit set and increased fruit disorders (Jones, 2007). Tomato requires the following daily accumulations (milligrams) in order to grow vegetatively: 20 (N), 2 (P), 25 (K), 120 (Ca), 10 (Mg). Whereas tomato requires the following daily accumulations (milligrams) in order for reproductive growth: 150 (N), 21 (P), 231 (K), 6 (Ca), 10 (Mg). Moreover, tomato prefers a pH range of 5.5 to 6.8, whereas the preferred electrical conductivity is 2.5 dS $m^{-1}$.

## REFERENCES


1. Abeles, F.B., P.W. Morgan, and M.E. Saltveit. (1992). Ethylene in Plant Biology. Academic Press, San Diego, CA.
2. Abu-Bakar, N.A., & Ibrahim, N. (2013). Indigenous microorganisms production and the effect on composting process. In AIP Conference Proceedings (Vol. 1571, No. 1, pp. 283-286). AIP.
3. Aerts, R. (1990). Nutrient use efficiency in evergreen and deciduous species from heathlands. *Oecologia*, *84* (3) 391-397.
4. Amiri, M., and Fallahi, E. (2009). Impact of animal manure on soil chemistry, mineral nutrients, yield, and fruit quality in 'Golden Delicious' apple. *Journal of plant nutrition*, *32*(4), 610-617.
5. Anton. (2017). Personal communication. Agrigro retailers. Windhoek
6. Atiyeh, R.M., Arancon, N., Edwards, C.A., and Metzger, J.D. (2000). Influence of earthworm-processed pig manure on the growth and yield of greenhouse tomatoes. *Bioresource Technology*, *75*(3), 175-180.
7. Auerswald, H., Schwarz, D., Kornelson, C., Krumbein, A., & Brückner, B. (1999). Sensory analysis, sugar and acid content of tomato at different EC values of the nutrient solution. *Scientia horticulturae*, *82*(3), 227-242.
8. Awodun, M.A., Omonijo, L.I., & Ojeniyi, S.O. (2007). Effect of goat dung and NPK fertilizer on soil and leaf nutrient content, growth and yield of pepper. International Journal of Soil Science, 2(2), 142-147.
9. Azeez, J.O., & Van Averbeke, W. (2010). Nitrogen mineralization potential of three animal manures applied on a sandy clay loam soil. Bioresource technology, 101(14), 5645-5651.

10. Baligar, V. C., R. R. Duncan, and N. K. Fageria. 1990. Soil-plant interaction on nutrient use efficiency in plants: An overview. pp 351–373. In: V C Baligar and R Duncan (eds.), Crops as Enhancers of Nutrient Use. Academic Press, San Diego, CA.
11. Baligar, V.C., and Bennett, O.L. (1986). Outlook on fertilizer use efficiency in the tropics. *Fertilizer Research, 10*(1), 83-96.
12. Baligar, V.C., Fageria, N.K., and He, Z.L. (2001). Nutrient use efficiency in plants. *Communications in Soil Science and Plant Analysis, 32*(7-8), 921-950.
13. Bandara, S.D.H., & Tennakoon, N.A. (2003). Nutrient content of some locally available organic materials and their potential as alternative sources of nutrients for coconut. COCOS, 15, 23-30.
14. Barrena I., Menéndez S., Correa-Galeote, D., Vega-Mas, I., Bedmar, E.J., González-Murua, C., Estavillo, J.M. (2017). Soil water content modulates the effect of the nitrification inhibitor 3,4-dimethylpyrazole phosphate (DMPP) on nitrifying and denitrifying bacteria. *Geoderma, 303*: 1-8.
15. Benbrook, C.M. (2005). Elevating antioxidant levels in food through organic farming and food processing. *Organic Center, State of Science Review, January*.
16. Blair, G. (1993). Nutrient efficiency—what do we really mean? In *Genetic aspects of plant mineral nutrition* (pp. 205-213). Springer Netherlands.
17. Borlaug, N.E., and Doswell, C.R. (1994). Feeding a human population that increasingly crowds a fragile planet 15th World Congress of Soil Sci. Supplement to Trans. 10p. Acapulco, Mexico.
18. Brandt, K., and Mølgaard, J.P. (2001). Organic agriculture: does it enhance or reduce the nutritional value of plant foods? *Journal of the Science of Food and Agriculture, 81*(9), 924-931.

19. Calvo, L., Cortey, M., Garcia-Marin, J.L., Garcia-Gil, L.J. (2005). Polygenic analysis of ammonia-oxidizing bacteria using 16S rDNA, amoA, and amoB genes. *Int Microbiol* **8**: 103–110.
20. Carbonaro, M., Mattera, M., Nicoli, S., Bergamo, P., & Cappelloni, M. (2002). Modulation of antioxidant compounds in organic vs conventional fruit (peach, Prunus persica L., and pear, Pyrus communis L.). *Journal of agricultural and food chemistry*, *50*(19), 5458-5462.
21. Cassman, K.G., Dobermann, A., and Walters, D.T. (2002). Agroecosystems, nitrogen-use efficiency, and nitrogen management. *AMBIO: A Journal of the Human Environment*, *31*(2), 132-140.
22. Chadwick, D.R., Pain, B.F., Brookman, S.K.E. (2000). Nitrous oxide and methane emmissiona following application of animal manures to grassland. Journal of Environmental Quality 29 (1): 277 – 287.
23. Chamber of Mines of Namibia (CMN). (2012). Recognizing our Responsibility. Annual review report: 2011/12.
24. Chassy, A.W., Bui, L., Renaud, E.N., Van Horn, M., and Mitchell, A.E. (2006). Three-year comparison of the content of antioxidant microconstituents and several quality characteristics in organic and conventionally managed tomatoes and bell peppers. *Journal of agricultural and food chemistry*, *54*(21), 8244-8252.
25. Chinta, Y.D., Eguchi, Y., Widiastuti, A., Shinohara, M., & Sato, T. (2015). Organic hydroponics induces systemic resistance against the air-borne pathogen, Botrytis cinerea (gray mould). *Journal of Plant Interactions*, *10*(1), 243-251.
26. Cooper, P.J.M., Dimes, J., Rao, K.P.C., Shapiro, B., Shiferaw, B., & Twomlow, S. (2008). Coping better with current climatic variability in the rain-fed farming

systems of sub-Saharan Africa: An essential first step in adapting to future climate change? *Agriculture, Ecosystems & Environment, 126*(1), 24-35.

27. Daims, H. (2001). Population Structure and Functional Analyses, by In Situ Techniques, of Nitrifying Bacteria in Wastewater Treatment Plants (Doctoral dissertation, Dissertation Technische Universität München).
28. dos Santos, J.D., da Silva, A.L.L., da Luz Costa, J., Scheidt, G.N., Novak, A.C., Sydney, E.B., and Soccol, C.R. (2013). Development of vinasse nutritive solution for hydroponics. *Journal of environmental management, 114*, 8-12.
29. Douglas, J.S. (1985). Advanced guide to hydroponics: (soiless cultivation). London: Pelham Books. pp. 169–187, 289–320, & 345–351. ISBN 9780720715712.
30. Duncan, EG., O'Sullivan, C.A, Simonsen, A.K, Roper, M.M, Peoples, M.B., Treble, K., Whisson, K. (2017). The nitrification inhibitor 3,4,-dimethylpyrazole phosphate strongly inhibits nitrification in coarse-grained soils containing a low abundance of nitrifying microbiota. *Soil Research, 55* (1), pp. 28-37.
31. Eghball, B., Wienhold, B.J., Gilley, J.E., & Eigenberg, R.A. (2002). Mineralization of manure nutrients. Journal of Soil and Water Conservation 57 (6): 470 - 473.
32. Esse, P.C., Buerkert, A., Hiernaux, P., & Assa, A. (2001). Decomposition of and nutrient release from ruminant manure on acid sandy soils in the Sahelian zone of Niger, West Africa. Agriculture, ecosystems & environment, 83(1), 55-63.
33. Evdokimova, M.A., Novoselov, S.I., Novoselova, E.S., Mantsurovna, A., Kuzminykh, A.N., Pashkova, G.I., Marina-Chermnykh, O.G. (2016). The influence of physical properties of sod-podzol soil on its ammonifying and nitrifying capacity. *Biology and Medicine* 8 (7) 351.

34. Fedunak, C.A., & Tyson, R.V. (1997). Lettuce cultivars for low-tech non-circulating hydroponics. In PROCEEDINGS-FLORIDA STATE HORTICULTURAL SOCIETY (Vol. 110, pp. 384-384). FLORIDA STATE HORTICULTURAL SOCIETY.
35. Fujiwara, K., Aoyama, C., Takano, M., and Shinohara, M. (2012). Suppression of Ralstonia solanacearum bacterial wilt disease by an organic hydroponic system. *Journal of general plant pathology*, *78*(3), 217-220.
36. Garland, J. L., Mackowiak, C. L., Strayer, R. F., & Finger, B. W. (1997). Integration of waste processing and biomass production systems as part of the KSC Breadboard project. *Advances in Space Research*, *20*(10), 1821-1826.
37. Garland, J.L., Mackowiak, C.L., & Sager, J.C. (1993). Hydroponic crop production using recycled nutrients from inedible crop residues (No. 932173). *SAE Technical Paper*.
38. Giovannucci, E. (1999). Tomatoes, tomato-based products, lycopene, and cancer; Review of the epidemiologic literature. *Journal of the cancer institute, 91*(4), 317-331.
39. Gitari, H.I., Mochoge, B.E., & Danga, B.O. (2015). Effect of lime and goat manure on soil acidity and maize (Zea mays) growth parameters at Kavutiri, Embu County-Central Kenya. Journal of Soil Science and Environmental Management, 6(10), 275-283.
40. Glass, A. (2003). "Nitrogen Use Efficiency of Crop Plants: Physiological Constraints upon Nitrogen Absorption". *Critical Reviews in Plant Sciences*. **22** (5): 453–470.
41. Goudie, A., and Viles, H. (2015). Climate. In *Landscapes and Landforms of Namibia* (pp. 37-46). Springer Netherlands.

42. Gruda, N. (2005). Impact of environmental factors on product quality of greenhouse vegetables for fresh consumption. *Critical Reviews in Plant Sciences*, *24*(3), 227-247.

43. Hallmann, E., and Rembiałkowska, E. (2007). Comparison of the nutritive quality of tomato fruits from organic and conventional production in Poland. Presented at the 3rd International Congress of the European Integrated Project Quality Low Input Food (QLIF) Congress: Improving Sustainability in Organic and Low Input Food Production Systems, University of Hohenheim, Germany, 20–23.

44. Handelsman, J. (2004). Metagenomics: application of genomics to uncultured microorganisms. *Microbiology and molecular biology reviews*, *68*(4), 669-685.

45. Heeb, A., Lundegårdh, B., Ericsson, T., and Savage, G. P. (2005). Nitrogen form affects yield and taste of tomatoes. *Journal of the Science of Food and Agriculture*, *85*(8), 1405-1414.

46. Heffer, P., & Prud'homme, M. (2010). Fertilizer outlook 2010–2014. In *78th IFA annual conference, Paris* (Vol. 31).

47. Herbert, M., Karam, A., Parent, L.E. (1991). Mineralization of nitrogen and carbon in soils amended with composted manure. Biol. Agric. Hortic. 7, 349–361.

48. Hochmuth, G.J., & Hochmuth, R.C. (2001). Nutrient solution formulation for hydroponic (perlite, rockwool, NFT) tomatoes in Florida. HS796. Univ. Fla. Coop. Ext. Serv., Gainesville.

49. Hu, C., and Qi, Y. (2013). Long-term effective microorganisms application promote growth and increase yields and nutrition of wheat in China. *European Journal of Agronomy*, *46*, 63-67.

50. Huson, D.H., Auch, A.F., Qi, J., Schuster, S.C. (2007). MEGAN analysis of metagenomic data. Genome research, 17(3), 377-386.
51. Hyman, J.R., Gaus, J., and Foolad, M.R. (2004). A rapid and accurate method for estimating tomato lycopene content by measuring chromaticity values of fruit puree. *Journal of the American Society for Horticultural Science, 129*(5), 717-723.
52. Ingham, E.R. (2000). The compost tea brewing manual. Corvallis, Oregon, USA: Unisun Communications.
53. Jensen, M.H. (1997). Hydroponics worldwide. In International Symposium on Growing Media and Hydroponics 481 (pp. 719-730).
54. Jones, J. B. (2007). Tomato plant culture: in the field, greenhouse, and home garden. CRC press
55. Jones, J.B. (2016). Hydroponics: A practical guide for the soilless grower. CRC press.
56. Jones, J.G. (1977). The effect of environmental factors on estimated viable and total populations of planktonic bacteria in lakes and experimental inclosures. *Freshwater Biol. 7*: 67-91.
57. Kämpfer, P., Erhart, R., Beimfohr, C., Böhringer, J., Wagner, M., Amann, R. (1996). Characterization of bacterial communities from activated sludge: culture-dependent numerical identification versus in situ identification using group- and genus-specific rRNA-targeted oligonucleotide probes. *Microb. Ecol. 32*: 101-121.
58. Kihanda, F.M., Warren, G.P., & Atwal, S.S. (2004). The influence of goat manure application on crop yield and soil nitrate variations in semi-arid eastern Kenya. Managing nutrient cycles to sustain soil fertility in Sub-Saharan Africa.

Academy Science Publishers in association with the Tropical Soil Biology and Fertility Institute of CIAT, Nairobi, 173-186.

59. Kirstein, K., and Bock, E. (1993). Close genetic relationship between Nitrobacter hamburgensis nitrite oxidoreductase and Escherichia coli nitrate reductases. *Archives of microbiology*, *160*(6), 447-453.
60. Knott, J.E. (1966) Handbook for vegetable growers. John Wiley, New York, 238 p.
61. Kogure, K., Simidu, U., Taga, N. (1980). Distribution of viable marine bacteria in neritic seawater around Japan. *Can. J. Microbiol. 26*: 318-323.
62. Kubota, C., Kroggel, M., Torabi, M., Dietrich, K.A., Kim, H.J., Fonseca, J., and Thomson, C.A. (2012). Changes in selected quality attributes of greenhouse tomato fruit as affected by pre-and postharvest environmental conditions in year-round production. *HortScience*, *47*(12), 1698-1704.
63. Kulcu, R., & Yaldiz, O. (2007). Composting of goat manure and wheat straw using pine cones as a bulking agent. Bioresource technology, 98(14), 2700-2704.
64. Lairon, D. (2011). Nutritional quality and safety of organic food. In *Sustainable Agriculture Volume 2* (pp. 99-110). Springer Netherlands.
65. Lee, J.G., Lee, B.Y., & Lee, H.J. (2006). Accumulation of phytotoxic organic acids in reused nutrient solution during hydroponic cultivation of lettuce (Lactuca sativa L.). *Scientia horticulturae, 110* (2), 119-128.
66. Lester, G. (2006). Organic versus conventionally grown produce: Quality differences, and guidelines for comparison studies. *HortScience*, *41*(2), 296-300.
67. Liang, Q., Chen, H., Gong, Y., Yang, H., Fan, M., Kuzyakov, Y. (2014). Effects of 15 years of manure and mineral fertilizers on enzyme activities in particle-size fractions in a North China Plain soil. *European Journal of Soil Biology*, *60*, 112-119.

68. Liedl, B.E., Cummins, M., Young, A., Williams, M.L., Chatfield, J.M. (2004). Liquid effluent from poultry waste bioremediation as a potential nutrient source for hydroponic tomato production. In *VII International Symposium on Protected Cultivation in Mild Winter Climates: Production, Pest Management and Global Competition 659* (pp. 647-652).
69. Linsley-Noakes, G., Wilken, L., & de Villiers, S. (2004). High density, vertical hydroponics growing system for strawberries. In V International Strawberry Symposium 708 (pp. 365-370).
70. Lockeretz, W. (2007). Organic Farming: An International History. CABI, Cambridge, MA.
71. Maboko, M.M., Du Plooy, C.P., & Bertling, I. (2011). Comparative performance of tomato cultivars cultivated in two hydroponic production systems. South African Journal of Plant and Soil, 28(2), 97-102.
72. Machado, D.C., Maia, C.M., Carvalho, I.D., Silva, N.F.D., André, M.C.D.P.B., and Serafini, Á.B. (2006). Microbiological quality of organic vegetables produced in soil treated with different types of manure and mineral fertilizer. *Brazilian Journal of Microbiology*, *37*(4), 538-544.
73. Mackowiak, C.L., Garland, J.L., Strayer, R.F., Finger, B.W., and Wheeler, R.M. (1996). Comparison of aerobically-treated and untreated crop residue as a source of recycled nutrients in a recirculating hydroponic system. *Advances in Space Research*, *18*(1), 281-287.
74. Marchesi, J.R (2012). Metagenomics: current innovations and future trends. Future Microbiology, 7(7), 813-814.

75. Marco, D. (2011). Metagenomics: current innovations and future trends. Horizon Scientific Press.
76. Maršić, N. K., Osvald, J., & Jakše, M. (2005). Evaluation of ten cultivars of determinate tomato (Lycopersicum esculentum Mill.), grown under different climatic conditions. Acta Agriculturae Slovenica, 85(2), 321-328.
77. Mayland, H.F., and Wilkinson, S.R. (1996). Mineral nutrition. p. 165–191. In L.E. Moser et al. (ed.) Cool-season forage grasses. Agron. Monogr. 34. ASA, CSSA, and SSSA, Madison, WI.
78. Mendelsohn, J. (2006). Farming systems in Namibia; A publication for the Namibia National Farmers Union (NNFU). Published by RAISON, Windhoek, Namibia.
79. Metzker, M.L. (2010). Sequencing technologies—the next generation. Nature reviews genetics, 11(1), 31-46.
80. Mitchell, J.P., Shennan, C., Grattan, S.R., & May, D.M. (1991). Tomato fruit yields and quality under water deficit and salinity. *Journal of the American Society for Horticultural Science*, *116*(2), 215-221.
81. Montgomery, D.R. (2007). Soil erosion and agricultural sustainability. Proceedings of the National Academy of Sciences, USA, 104(33), 13268-13272.
82. Moreno-Caselles, J., Moral, R., Perez-Murcia, M., Perez-Espinosa, A., & Rufete, B. (2002). Nutrient value of animal manures in front of environmental hazards. Communications in Soil Science and Plant Analysis, 33(15-18), 3023-3032.
83. Morris, M.L. (2007). *Fertilizer use in African agriculture: Lessons learned and good practice guidelines*. World Bank Publications.

84. Mowa, E. (2015). Organic manure for vegetable production under hydroponic conditions in arid Namibia. International Science & Technology Journal of Namibia, 2015, 5:3-12.
85. Munoz, G.R., Kelling, K.A., Powell, J.M., Speth, P.E. (2004). Comparison of estimates of first-year dairy manure nitrogen availability or recovery using nitrogen – 15 and other techniques. Journal of environmental quality, 33 (2): 719 – 727.
86. Mupondi, L.T., Mnkeni, P.N.S., & Brutsch, M.O. (2006). Evaluation of pine bark or pine bark with goat manure or sewage sludge cocomposts as growing media for vegetable seedlings. Compost science & utilization, 14(4), 238-243.
87. MWAZI, F. (2012). Green house evaluation of the effects of salinity on germination, growth and quality of vegetables grown in a hydroponic system along the coast of Namibia. AGRICOLA, 45.
88. Mwazi, F.N., Amoonga, S., & Mubiana, F.S. (2010). Evaluation of the effects of salinity on spinach (*Beta vulgaris var. cicla*) grown in a hydroponic system along the coast of Namibia. Agricola, 20, 14-17.
89. Namibia Agronomic Board (NAB). (2017). Market Statistics. Namibia Agronomic Board, Windhoek, Namibia.
90. Nampa. (2005, December 20). RUNDU - The Ministry of Agriculture is promoting the introduction of hydroponic gardening in the Kavango Region. The Namibian, p. 15.
91. Olaniyi, J.O., and Ajibola, A.T. (2008). Effects of inorganic and organic fertilizers application on the growth, fruit yield and quality of tomato (Lycopersicon lycopersicum). *Journal of Applied Biosciences*, *8*(1), 236-242.

92. Oliveira, A.B., Moura, C.F., Gomes-Filho, E., Marco, C.A., Urban, L., and Miranda, M.R.A. (2013). The impact of organic farming on quality of tomatoes is associated to increased oxidative stress during fruit development. *PLoS One*, *8*(2), e56354.

93. Omoregie, E. (2013). Annual report. University of Namibia (UNAM).

94. Philander, F. (2007, October 31). Germany Helps Out in Hydroponics. New Era, p. 10

95. Pieper, J.R., and Barrett, D.M. (2009). Effects of organic and conventional production systems on quality and nutritional parameters of processing tomatoes. *Journal of the Science of Food and Agriculture*, *89*(2), 177-194.

96. Pimentel, D., Hepperly, P., Hanson, J., Douds, D., and Seidel, R. (2005). Environmental, energetic, and economic comparisons of organic and conventional farming systems. BioScience, 55(7), 573-582.

97. Poly, F., Wertz, S., Brothier, E., Degrange, V. (2008). First exploration of Nitrobacter diversity in soils by a PCR cloning-sequencing approach targeting functional gene nxrA. *FEMS microbiology ecology*, *63*(1), 132-140.

98. Pommering-Röser, A., Rath, G., Koops, H.P. (1996). Phylogenetic diversity within the genus Nitrosomonas. *System. Appl. Microbiol*. 19: 344-351.

99. Ponte, S., and Gibbon, P. (2005). Quality standards, conventions and the governance of global value chains. *Economy and society*, *34*(1), 1-31.

100. Pratt, P., & Castellanos, J. (1981). Available nitrogen from animal manures. California Agriculture, 35(7), 24-24.

101. Premuzic, Z., Bargiela, M., Garcia, A., Rendina, A., & Iorio, A. (1998). Calcium, iron, potassium, phosphorus, and vitamin C content of organic and hydroponic tomatoes. *HortScience*, *33*(2), 255-257.

102. Purkhold, U., Pommerening-Röser, A., Juretschko, S., Schmid, M.C., Koops, H.P., Wagner, M. (2000). Phylogeny of all recognized species of ammonia oxidizers based on comparative 16S rRNA and amoA sequence analysis: implications for molecular diversity surveys. *Applied and environmental microbiology*, *66*(12), 5368-5382.

103. Qian, P., Schoenau, J.J. (2002). Availability of nitrogen in solid manure amendments with different C: N ratios. Canadian Journal of Soil Science, 82(2), 219-225.

104. Reider, C.R., Herdman, W.R., Drinkwater, L.E., & Janke, R. (2000). Yields and nutrient budgets under composts, raw dairy manure and mineral fertilizer. Compost Science & Utilization, 8(4), 328-339.

105. Resh, H.M. (2012). Hydroponic food production: a definitive guidebook for the advanced home gardener and the commercial hydroponic grower. CRC Press.

106. Rippy, J.F., Peet, M.M., Louws, F.J., Nelson, P.V., Orr, D.B., & Sorensen, K.A. (2004). Plant development and harvest yields of greenhouse tomatoes in six organic growing systems. *Hortscience*, *39*(2), 223-229.

107. Roberts, T.L. (2008). Improving nutrient use efficiency. *Turkish Journal of Agriculture and Forestry*, *32*(3), 177-182.

108. Rotthauwe, J.H., Witzel, K.P., Liesack, W. (1997). The ammonia monooxygenase structural gene amoA as a functional marker: molecular fine-scale analysis of natural ammonia-oxidizing populations. *Applied and environmental microbiology*, *63*(12), 4704-4712.

109. Rubio, G., Walk, T., Ge, Z., Yan, X., Liao, H., & Lynch, J.P. (2001). Root gravitropism and below-ground competition among neibouring plants: a modelling approach. *Annals of Botany, 88*(5), 929-940.

110. Rughöft, S., Herrmann, M., Lazar, C.S., Cesarz, S., Levick, S.R., Trumbore, S.E., Küsel K. (2016). Community composition and abundance of bacterial, archaeal and nitrifying populations in savanna soils on contrasting bedrock material in Kruger National Park, South Africa. *Frontiers in Microbiology*, 7 (OCT), art. no. 1638.

111. Saha, S.K. (2010). Soilless Cultivation for Landless People: An Alternative Livelihood Practice through Indigenous Hydroponic Agriculture in Flood-prone Bangladesh.

112. Saijai, S., Ando, A., Inukai, R., Shinohara, M., Ogawa, J. (2016). Analysis of microbial community and nitrogen transition with enriched nitrifying soil microbes for organic hydroponics. Bioscience, Biotechnology, and Biochemistry, 80(11), 2247-2254.

113. Sakamoto, Y., Watanabe, S., Nakashima, T., and Okano, K. (1999). Effects of salinity at two ripening stages on the fruit quality of single-truss tomato grown in hydroponics. *The Journal of Horticultural Science and Biotechnology*, *74*(6), 690-693.

114. Salisbury, F.B., & Ross, C.W. (1992). Plant physiology. 4th edition. Wadsworth Pub. Co.

115. Sanchez, P.A. (2002). Soil fertility and hunger in Africa. *Science(Washington), 295*(5562), 2019-2020.

116. Sansoucy, R. (1995). Livestock-a driving force for food security and sustainable development. World, 3074(5389), 1035.

117. Savvas, D. 2003. Hydroponics: A modern technology supporting the application of intergrated crop management in greenhouse. *Journal of food agriculture and environment, 1,* 80-86.

118. Scalbert, A., Manach, C., Morand, C., Remesy, C. (2005). Dietary polyphenols and the prevention of diseases. *Crit. Rev. Food Sci. Nutr*. 45: 287-306.

119. Schnitzler, W.H., Sharma, A.K., Gruda, N.S., and Heuberger, H.T. (2001). A low-tech hydroponic system for bell pepper (Capsicum Annuum L.) production. In *International Symposium on Growing Media and Hydroponics 644* (pp. 47-53).

120. Sgherri, C., Cecconami, S., Pinzino, C., Navari-Izzo, F., Izzo, R. (2010). Levels of antioxidants and nutraceuticals in basil grown in hydroponics and soil. *Food Chemistry 123*, 416–422.

121. Shah, M.C., Kansara, S.R., & Shilpkar, P.G. (2015). Suitability of Goat Dung and Calotropis Gigantea Leaves for Composting. International Journal of Chemical Sciences, 13(1).

122. Shah, S.H., Munir, S.U., & Shah, S.H. (2011). Evaluation of two nutrient solutions for growing tomatoes in a non-circulating hydroponics system. Sarhad Journal of Agriculture (Pakistan).

123. Sheikh, B.A. (2006). Hydroponics: key to sustain agriculture in water stressed and urban environment. Pak. J. Agric., Agril. Eng., Vet. Sci. 22, 53–57.

124. Shi, J., and Maguer, M.L. (2000). Lycopene in tomatoes: chemical and physical properties affected by food processing. *Critical reviews in food science and nutrition, 40*(1), 1-42.

125. Shinohara, M., Aoyama, C., Fujiwara, K., Watanabe, A., Ohmori, H., Uehara, Y., & Takano, M. (2011). Microbial mineralization of organic nitrogen into nitrate to allow the use of organic fertilizer in hydroponics. Soil science and plant nutrition, 57(2), 190-203.

126. Sneh, M. (1995). History of fertigation in Israel. *International Water & Irrigation Review*, *15*(3), 17-20.

127. Starkenburg, S.R, Chain, P.S., Sayavedra-Soto, L.A., Hauser, L., Larimer, F.W., Hickey, W.J. (2006). Genome sequence of the chemolithoautotrophic nitrite-oxidizing bacterium Nitrobacter winogradskyi Nb-255. *Applied and environmental microbiology*, *72*(3), 2050-2063.

128. Strayer, R.F., Finger, B.W., and Alazraki, M.P. (1997). Evaluation of an anaerobic digestion system for processing CELSS crop residues for resource recovery. *Advances in Space Research*, *20*(10), 2009-2015.

129. Tchegueni, S., Koriko, M., Koledzi, E., Bodjona, M.B., Kili, K.A., Tchangbedji, G., & Hafidi, M. (2013). Physicochemical characterization of organic matter during co-composting of shea-nut cake with goat manure. African Journal of Biotechnology, 12(22).

130. Tennakoon, N.A. (1990). Goat manure as a soil 'ameliorant' and yield 'stimulant' in coconut. COCOS, The Coconut Research Institute of Sri Lanka 8,26-32.

131. Thippayarugs, S., Suzuki, K., Katsuta, Y., Yoshida, A., Matsumoto, N., Kabaki, N., & Wongwiwatchai, C. (2001). Vegetable production using energy-saving

hydroponics systems in Northeast Thailand. JIRCAS Journal for Scientific Papers (Japan).

132. Toor, R., Savage, G., and Heeb, A. (2006). Influence of different types of fertilizers on the major antioxidant components of tomatoes. *Journal of Food Composition and Analysis*, *19*(1), 20-27.

133. Torsvik, V., Goksoyr J., Daae, F.L. (1990). High diversity in DNA of soil bacteria. *Appl. Environ. Microbiol*. 56: 782-787.

134. Trejo-Téllez, L.I., & Gómez-Merino, F.C. (2012). Nutrient solutions for hydroponic systems. In Hydroponics-A Standard Methodology for Plant Biological Researches. InTech.

135. Tyson, R.V. (2002). Off the shelf hydroponic gardens. In Proc. Fla. State Hort. Soc (Vol. 115, pp. 159-160).

136. UNDP. 2015. Hydroponics Vegetable Farming. SGP The GEF Small Grants Programme.

137. Warncke, D.D., & Krauskopf, D.M. (1983). Greenhouse growth media: Testing and nutrition guidelines. Extension Bulletin E, 1736.

138. Wertz, S., Goyer, C., Zebarth, B.J., Burton, D.L., Tatti, E., Chantigny, M.H., Filion, M. (2013). Effects of temperatures near the freezing point on N2O emissions, denitrification and on the abundance and structure of nitrifying and denitrifying soil communities. *FEMS Microbiology Ecology*, 83: (1) 242-254

139. Willuweit, T., Söll, P., Müller, R. (2008). U.S. Patent No. 7,384,777. Washington, DC: U.S. Patent and Trademark Office.

140. Woese, C.R., Weisburg, W.G., Paster, B.J., Hahn, C.M., Tanner, R.S., Krieg, N.R., Koops, H.P., Harms, H., Stackebrandt, E. (1984). The phylogeny of the purple bacteria: the beta subdivision. System. Appl. Microbiol. 5: 327-336.

141. Xu, H.L., Wang, R., and Mridha, M.A.U. (2001). Effects of organic fertilizers and a microbial inoculant on leaf photosynthesis and fruit yield and quality of tomato plants. *Journal of Crop Production*, *3*(1), 173-182.

142. Zaller, J.G. (2007). Vermicompost as a substitute for peat in potting media: Effects on germination, biomass allocation, yields and fruit quality of three tomato varieties. *Scientia Horticulturae*, *112*(2), 191-199.

143. Zhou, X., Fornara, D., Wasson, E.A., Wang, D., Ren, G., Christie, P., Jia, Z. (2015). Effects of 44 years of chronic nitrogen fertilization on the soil nitrifying community of permanent grassland. Soil Biology and Biochemistry 91: 76-83

144. Zotarelli, L., Dukes, M.D., Scholberg, J.M.S., Munoz-Carpena, R., and Icerman, J. (2009). Tomato nitrogen accumulation and fertilizer use efficiency on a sandy soil, as affected by nitrogen rate and irrigation scheduling. *Agricultural Water Management*, *96*(8), 1247-1258.

# CHAPTER 3: FORMULATION OF AN ORGANIC HYDROPONIC NUTRIENT SOLUTION FROM GOAT MANURE

## ABSTRACT

For many years, the generation of nitrates from organic sources in order to create nutrient solutions for hydroponics had proved a challenge until lately when microorganisms were introduced to perform this task. The objective of the current study therefore, was to use local microbial consortium to nitrify goat manure in water and to determine microbial diversity in the inoculated consortium. Therefore, microorganisms were sourced from garden soil and natural compost at the Sam Nujoma Marine Resources Research Centre (SANUMARC) in Henties Bay Namibia to convert organic nitrogen in goat manure from Utuseb farm near Walvis Bay into nitrates. Results show that microbial consortium from the compost source produced significantly ($P< 0.05$) more nitrates followed by the garden-soil source, suggesting that it is necessary to add inoculum in order to generate nitrate from goat manure. The ammonia oxidizing bacteria (AOB) community from the compost samples was dominated by uncultured ammonia-oxidizing species followed by uncultured bacterium (both not identified), with the least being *Nitrosomonas* species. The AOB community from the garden source was dominated by uncultured bacterium, followed by uncultured ammonia-oxidizing species and the least being *Nitrosomonas* species. NOB community from the compost source was dominated by uncultured bacterium, followed by *Nitrobacter winogradskyi* and *Nitobacter vulgaris* with the least being *Nitrococcus mobilis* and *Nitrospira moscovensis.* For the garden soil microbial source, uncultured nitrite oxidizing bacteria dominated followed by uncultured bacterium, whereas the least species were *N. moscovensis* and *Nitrobacter alkalicus.* Moreover, community composition of the compost sample was more diverse than the community from the garden sample. These results maintain that there are other unculturable yet important microbes doing the same job if not better than the known ones, in this case suggesting that there may be other local nitrite-oxidizing bacteria responsible for oxidizing ammonia other than the traditionally known *Nitrobacter, Nitrospira* and *Nitrococcus* species.

## KEYWORDS

Hydroponics, ammonification, nitrification, ammonia-oxidizing bacteria, nitrite-oxidizing bacteria.

## 3.1 INTRODUCTION

The first part of this gives the problem statements, objectives and hypothesis of the chapter. The second part of this chapter then details the procedure or materials and methods used to collect and analyse data for the objective of this chapter. The last part presents the results found from the collection and analysis of data for this chapter as well as the discussion and conclusion of the chapter.

### 3.1.1 Statement of the Problem

The gap arising from literature on hydroponics is that though hydroponics method of crop production is suited for countries like Namibia, it is not up-taken to full use due to the inaccessibility of fertilisers currently used to most potential hydroponic farmers. Hence there is a need to find an alternative means of the hydroponic technology that will be accessible to many in Namibia in general and Namibia's desert and coastal communities in particular. A potential alternative is goat manure, which is easily accessible at Namibia's desert and coastal communities. The challenge with animal manure being used hydroponically is that nitrogen in animal manure is largely in organic forms which makes it inaccessible to plants. Recent discoveries by Shinohara *et al* (2011), however, reveal the possibility to use nitrifying microorganisms from soils to release the organic nitrogen in manure into plant available inorganic forms. Such studies focused on fish-based organic fertilizer, but there has not been any study focusing on domestic animal manure like goat

manure, using similar procedures to produce organic hydroponic nutrient solutions for plant growth. To this end, in Africa, there has never been a documented study on organic hydroponics of any kind, apart from one done by Mowa (2015), where the levels of nutrients in the hydroponic solution were not established.

### 3.1.2 Objective

The objective of the current study was to formulate hydroponic nutrient solution from Goat manure.

### 3.1.3 Research hypothesis

Hydroponic nutrient solution can be formulated from goat manure.

## 3.2 MATERIALS AND METHODS

### 3.2.1 Study site

The current study was conducted at the Sam Nujoma Marine Resources Research Centre (SANUMARC) 22.1135˚ S, 14.2832˚ E, Henties Bay, Namibia (Figure 2).


**Figure 2: Map showing study area (Arroukatchee, 2017)**

### *3.2.2 Manure collection and preparation*

Dry manure was sampled according to Lupwayi *et al* (2000), whereby samples were collected from Utuseb communal farm in the Erongo region, where the manure was stored for at least three months in an open storage. Goat manure was chosen because it is abundantly available due to dominance of small stock farming in the western regions of

Namibia which contain the coastal towns and villages including Henties Bay. Utuseb communal farm was chosen due to its proximity to Henties Bay. The manure was homogenized, air-dried for 1 week at 30°C, ground and sieved (< 2 mm) before use as per Qian & Schoenau (2002).

In order to determine levels of mineral nutrients in the collected manure before formulating the nutrient solution, this prepared manure was then added at 1g/L to 1L of water, stirred for 24 hours and the supernatant was filtered and macro-elemental analysis were done as follows: Total Nitrogen – using modified Kjeldahl method (ISO 11261) (Nikodemus, 2013) Sulphate – using automated methylene blue method (detection limit of 1mg/L) (Biati *et al.,* 2010), Nitrate – using automated Cd reduction (detection limit of 0.1 mg/L) (Shinohara, 2011), Phosphate – using spectrophotometric method (detection limit of 0.03 mg/L) (Hussain & Shah, 2014), Magnesium – using ICP-OES (detection limit of 0.1 mg/L) (Sen *et al.,* 2011), Potassium - using ICP-OES (detection limit of 0.1 mg/L) (Sen *et al.,* 2011), Calcium - using ICP-OES (detection limit of 0.1 mg/L) (Moir *et al.,* 1996). The results were expressed on a dry-weight basis.

Data reported by Bandara & Tennakoon (2003); Moreno-Caselles *et al* (2002); Azeez & Van Averbeke, (2010) indeed showed that in the minimum range, goat manure had 1040 mg/kg Iron (Fe), 20 mg/kg Copper (Cu), 246 mg/kg Manganese (Mn), 112 mg/kg Zinc (Zn), 2 mg/kg Cadmium (Cd), and 29 mg/kg Boron. In the maximum range, goat manure had 18% Fe, 13% Mn, 6% Zn, and 38% Boron more than cattle manure for example. Hence it was judged that goat manure has acceptable composition of the mineral micronutrients. Therefore, given the comparisons of the various sources of manure to

goats (Bandara & Tennakoon, 2003; Moreno-Caselles *et al*., 2002; Azeez & Van Averbeke, 2010) mineral micronutrients in the goat manure were presumed to be adequate for formulating hydroponic nutrient solution without engaging considerable time and financial resources for analysing mineral micronutrient information.

### 3.2.3 Source of microorganisms

The following sources were used: Sam Nujoma Marine Resources Research Centre (SANUMARC) garden soil and SANUMARC compost. SANUMARC garden soil was soil extracted from garden plots in the shaded-net garden of SANUMARC at Henties Bay. SANUMARC compost was soil collected from a compost heap at SANUMARC where grass clippings and plant leaves have been heaped for longer than 3 months. In order to determine the best source of microorganisms to mineralize organic N, samples from the above sources were investigated as shown in Table 3. Each treatment was added to separate one-litre flasks of distilled water containing 1 g/L of goat manure. Each treatment was triplicated. The flasks were shaken (120 strokes/min) for 20 days at 25˚C, and the $NO_3^-$concentrations were then determined using the Hach DR5000 spectrophotometer which uses Cadmium metals to reduce nitrates in the sample to nitrite. The Nitrite ion reacts in an acidic medium with Sulfunic acid to form an intermediate diazonium salt. The salt couples with gentisic acid to form an amber coloured solution and the results are then measured at 500nm.

**Table 3: treatments for investigating optimal source of microorganisms mineralising organic nitrogen**

| Treatments | Inoculum added (g/l) |
|---|---|
| Treatment 1: Compost added (compost essentially a farmyard manure). Tends to have one of the highest mineral micronutrients, but less than of goat. | 5 |
| Treatment 2:<br>Garden soil added (not regarded as farmyard manure, but a plain soil) | 5 |
| Treatment 3: Control<br>Manure + water only. No inoculum added | 0 |

### 3.2.4 Identification of microbes

In order to understand microbial composition of the 2 inocula from the source of microorganisms experiment**,** identification was done. Therefore, DNA was extracted from both the garden soil and the compost from SANUMARC. The DNA extraction was performed according to manufacturer's recommendations: based on direct cell lysis with subsequent recovery and purification of nucleic acids. Up to 250 mg of soil was added to the lysis tubes, and the samples were homogenized using a standard benchtop vortex for 20 minutes. The DNA was then amplified using functional genes targeting primers (*NorA* and *AmoA*) using standard PCR procedure at SANUMARC.

Genomic DNA was prepared and used as a template. 305bp *NxrA* genes and 490bp *AmoA* genes were amplified using NorA and AmoA genes. The sequence of the amoA primers was: amoA-f 5'- GGGGTTTCTACTGGTGGT-3′, amoA-r 5′- CCCCTCKGGAAAGCCTTCTTC-3′ (Rotthauwe et al., 1997; Purkhold et al.,2000). The sequence for the NXR primers was: norA-f 5′ CAGACCGACGTGTGCGAAAG3′, norA-r 5′-TCYACAAGGAACGGAAGGTC-3′ (Poly *et al.*, 2008). The PCR mixture included 0.5 µM of each primer, 23.5 µl of nuclease free water, 25 µl master mix and 1

µl of DNA template, making up a total of 50 µl. The negative control contained nuclease free water in place of DNA template.

Using the thermos-cycler, the PCR reaction was carried out as follows:

NXRA - 1 cycle of pre-denaturation at 94ºC for 5 minutes, 30 cycles of denaturation at 94ºC for 30 seconds, annealing at 60ºC for 30 seconds, extension at 72ºC for 45 seconds, final extension at 72ºC for 5 minutes.

AmoA- 1 cycle of pre-denaturation at 94ºC for 5 minutes, annealing at 60ºC for 1 minute 30 seconds, extension at 72ºC for 1 minute 30 seconds, 42 cycles of denaturation at 94ºC for 1 minute, annealing at 60ºC for 1 minute 30 seconds, final extension at 72ºC for 10 minutes.

Amplified PCR products were visualised on 1% agarose gel with a 1kb ladder (Inqaba Biotechnology Industries, SA). Further sequencing of the PCR products was done (South Africa) and the reads were merged, clustered and then a BLAST search was done against Genbank in order to identify nitrifying species from the sequence data.

### 3.2.5 Concentration of microorganisms

Microorganisms were sourced from natural compost at the Sam Nujoma Marine & Coastal Research Centre in Henties Bay Namibia, where compost was sampled and used as nitrifying microbial inoculum in order to establish an optimum concentration level of the inoculum. Therefore, inoculum was added at 1g, 5g & 10g/L to 100-mL flasks (n=3 per treatment) of distilled water containing 0.5 g of goat manure. No inoculum was added to

the control. The flasks were then shaken (120 strokes/min) for 21 days at 25°C, and then the nitrate concentrations were determined using the Hach DR5000 spectrophotometer.

### 3.2.6 Manure for optimal nitrate generation by inoculum

Since 10g/L of inoculum in the previous experiment (3.2.5) was carried forward, goat manure was added as follows: 0.25, 0.5, or 1 g/L daily for 15 days to 2 L of water containing the 10g/L microbial inoculum (Shinohara *et al.*, 2011). This was done to ferment and establish conducive ecosystem in which nitrifying bacteria would thrive and function optimally. Once this got established, 1g/L of manure was added daily for the next 30 days to 6 L of distilled water plus 1 L of the established ecosystem in 7 L buckets. The pH was maintained between 7.3 and 8.0, whereas temperature was maintained at 28°C throughout the experiment using a Plant Growth Chamber (Labcon – double door). Dissolved oxygen was kept above 80% saturation by aeration (20kPa) with Aleas Double Outlet Air Pumps. Nitrates were determined and recorded after 7, 12, 15, 20, 30, 40 and 50 days.

In order to establish levels of major elements required for plant growth in formulated nutrient solution, after Nitrates reached 198mg/L in the nutrient solution, the following elements were determined using the DR5000 HACH spectrophotometer: Phosphorus (P), Sulphur (S). The following were determined using DR890 colorimeter: Potassium (K), Nitrates (NO3). Calcium (Ca) and Magnesium (Mg) were determined using ICP-OES method. The pH of the nutrient solution was then adjusted using citric acid to lower it. The prepared goat manure was added to the formulated nutrient solution in order to

increase the levels of the other mineral nutrients, considering they were found to be relatively low. Recommended mineral nutrition for tomatoes are generally: nitrate (NO3) 200 – 210 mg/L, Phosphate (P) 46.7 – 50 mg/L, Potassium (K) 371.5 – 420 mg/L, Magnesium (Mg) 58.3 – 75 mg/L, Sulphur (S) 120 – 140.9 mg/L, and Calcium (Ca) 190 – 216 mg/L (Peet & Welles, 2005; Wamcke & Krauskopf, 1983; Adams, 1986).

The recommended pH is between 6.0 -7,3 (Buck *et al.,* 2004), whereas the recommended Electrical Conductivity (EC) is between 1 – 4.5 dS $m^{-1}$ (Schwarz & Kuchenbuch, 1998; Buck *et al.,* 2004).

### 3.2.7 Data analysis

Means comparisons and correlations analysis were done whereby, a One-way Analysis of Variance (ANOVA), followed by mean separation by Duncan's Multiple Range Test was used when ANOVA determined that the effects of the treatments were significant ($P \leq 0.05$ for F-test).

$$Yij = \mu + \tau i + \beta j + \gamma ij + \epsilon ij$$

Where $\mu$ is the overall mean response, $\tau i$ is the effect due to the $i$ -th level of factor A, $\beta j$ is the effect due to the$j$ -th level of factor B and $\gamma ij$ is the effect due to any interaction between the $i$ -th level of A and the $j$ -th level of B, $\epsilon ij$ is the error term where the error terms are independent observations.

## 3.3 RESULTS

### 3.3.1. Levels of nutrients in unprocessed goat manure

The levels of mineral nutrients in the collected manure before it was used to formulate the nutrient solutions were found as highlighted in Table 4 below.

**Table 4: Levels of mineral nutrients in goat manure collected from Utuseb farm*, Erongo Region, Namibia.**

| Element | Symbol | mg/kg (Level in conventional solution)**? | Mg/kg (Obtained) | Magnitude of observed |
|---|---|---|---|---|
| Nitrogen | N | 15,000 | 22000 | x1.4-fold |
| Potassium | K | 10,000 | 11950 | x1.2-fold |
| Calcium | Ca | 5,000 | 14740 | x2.9-fold |
| Magnesium | Mg | 2,000 | 2720 | x1.4-fold |
| Phosphorus | P | 2,000 | 3180 | x1.6-fold |
| Sulfate | S | 1,000 | 46000 | x46-fold |
| Nitrate | NO3 | 223.9 mg/L | 0.072 | |
| Electrical conductivity | EC | ***1 – 4.5 dS $m^{-1}$ | 1.87 dS $m^{-1}$ | x2 fold |
| pH | 1.5 | ***recommended 6.0 -7,3 | 7.4 | Within similar range |

*Sample collected from where most goat farmers inhabit close to Henties Bay. **?= the expected would be dependent on whether or not a given source is undergoing nitrification; ***Schwarz & Kuchenbuch, 1998; Buck *et al.*, 2004.

### 3.3.2. Nitrate levels from three sources of microorganisms

The garden soil and compost soil that had been chosen as inoculum sources for the microorganisms needed for mineralization and nitrification of organic nitrogen into nitrate plus goat manure as a nitrogen source were found to be different (Fig 3).

The control (addition of only goat manure without inoculum in water) resulted in the lowest nitrate generation (Table 5). The finding suggests the importance of the presence

of the nitrifying organisms. In contrast, the microbial consortium from the compost source, however, significantly ($P \leq 0.05$) produced more nitrates than the garden-soil source and the control where no inoculum had been added (Fig 3).

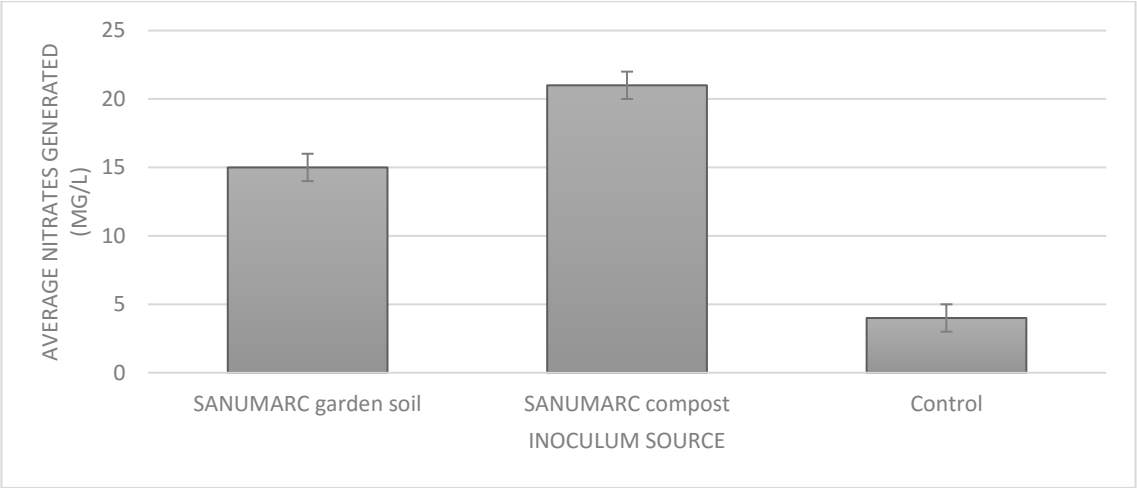



**Figure 3: Mineralization of organic nitrogen into nitrate by the addition of microbial inocula from various sources.**

**Table 5: Descriptive statistics for source of microorganisms between the 3 treatments**

| Treatment | N | Mean | Std. Deviation | Std. Error | CV | Std. Error |
|---|---|---|---|---|---|---|
| Garden soil | 6 | 17 | .894 | .365 | 5,25 | .365 |
| Compost | 6 | 20.78 | 1.470 | .600 | 7.07 | .600 |
| Control | 6 | 4.67 | .516 | .211 | 11 | .211 |

### 3.3.3 Identification of microbes

The two sources of inoculum (i.e. garden vs compost) contained microbial species capable of oxidising ammonia (Table 6a). The pie charts represent the proportion of the community of micro-organisms peculiar to each of environments (i.e. C & G) and are thus

amplifying the information in Table 6a. The highest preponderance of bacterial amoA gene in the Compost (i.e. C) (Figure 4c) was in an uncultured ammonia-oxidizing species (64%) followed by uncultured bacterium (21%). In contrast, in the garden soil (i.e. G) (Figure 4g) uncultured bacterium was 51% followed by uncultured ammonia-oxidizing species (34%). The Garden soil with a 34% preponderance of AOB compared to 64% of AOB in Compost would appear to suggest the oxidizing beneficial effect of AOB in the latter for generating nitrites from nitrogen (see Figure 3). It can be noted that the uncultured bacterium of 51% preponderance in contrast was reduced to 21% in Compost soil.

**Table 6a: Ammonia Oxidising Bacteria (AOB) species composition from the 2 sources of inoculum and species diversity**

| **Community of microbial organisms as distinguished from *metagenomic analysis** | **Inoculum source (numbers of possible microbial organisms)** | | |
|---|---|---|---|
| | **Garden** | **Compost** | **Magnitude of increase in compost** |
| 1. uncultured ammonia-oxidising | 529 | 2469 | x 4.7-fold |
| 2. Nitrosomonas sp. | 1 | 14 | x 14-fold |
| 3. uncultured Nitrosomonadales | 12 | 0 | x 0-fold |
| 4. Nitrosospira sp. | 4 | 537 | x 134-fold |
| 5. uncultured bacterium | 807 | 823 | x 1-fold |
| 6. uncultured beta | 181 | 34 | x 0.2-fold |
| 7. uncultured Nitrosospira | 47 | 0 | x 0-fold |
| **SUM** | **1581** | **3877** | **x 2.5-fold** |
| Simpson diversity index | **0.61** | **0,53** | |

*see Appendix 1 (A & B)

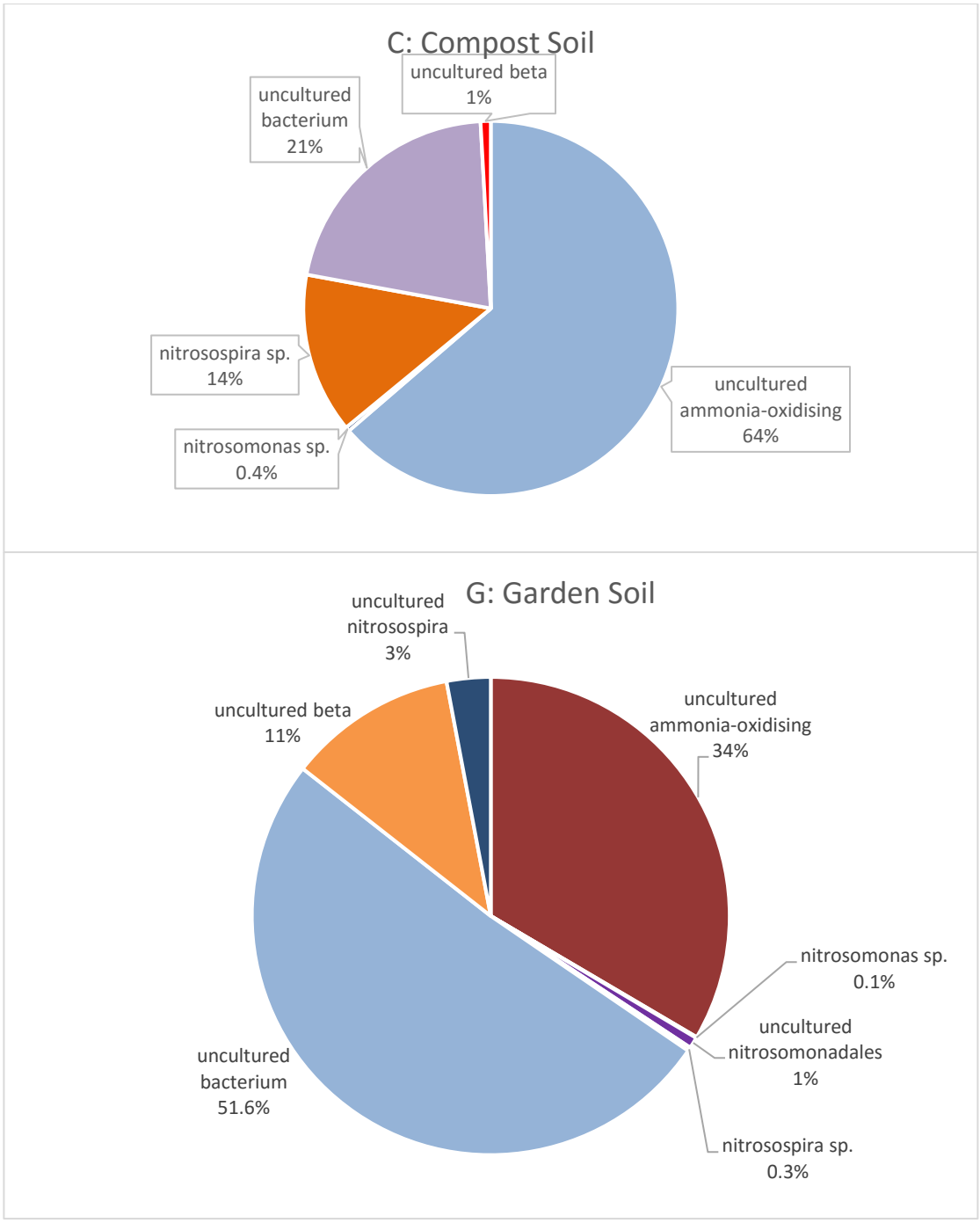



**Figure 4c & 4g: Relative abundance of the various communities of the total Ammonia-Oxidising Bacteria (AOB) metagenome isolated from the 2 main environments (Environment C: compost soil; G: garden soil).**

*Nitrospira moscovensis* (NOB-Nitrosp) was 0.01% in the Compost inoculum. In the same inoculum, Nitrosomonas species (AOB-Nitrosom) was 0.4%. The numerical ratio of the NOB-Nitrosp to AOB-Nitrosom is 1:40. In the Garden inoculum, the numerical ratio was 1:10. In effect, the differences might seem to suggest that there was ammonia oxidising effect in the compost inoculum (Figures 5c & 5g).

The total Nitrobacter metagenome (responsible for using nitrite substrate for conversion into nitrates in compost inoculum was 43% (Figure 5c) compared to 20% from the garden inoculum. There is no doubt, therefore, that the predominant Nitrobacter activity in the Compost inoculum seems to account for the higher generation of nitrates (see Figure 3).

The pH of the garden soil was 7.8 (range which is slight to moderate alkalinity) whereas for compost soil pH was 8.6 (strong alkalinity). pH optimum conditions under which nitrifying microorganisms operate in range of 7.5 – 9 pH.

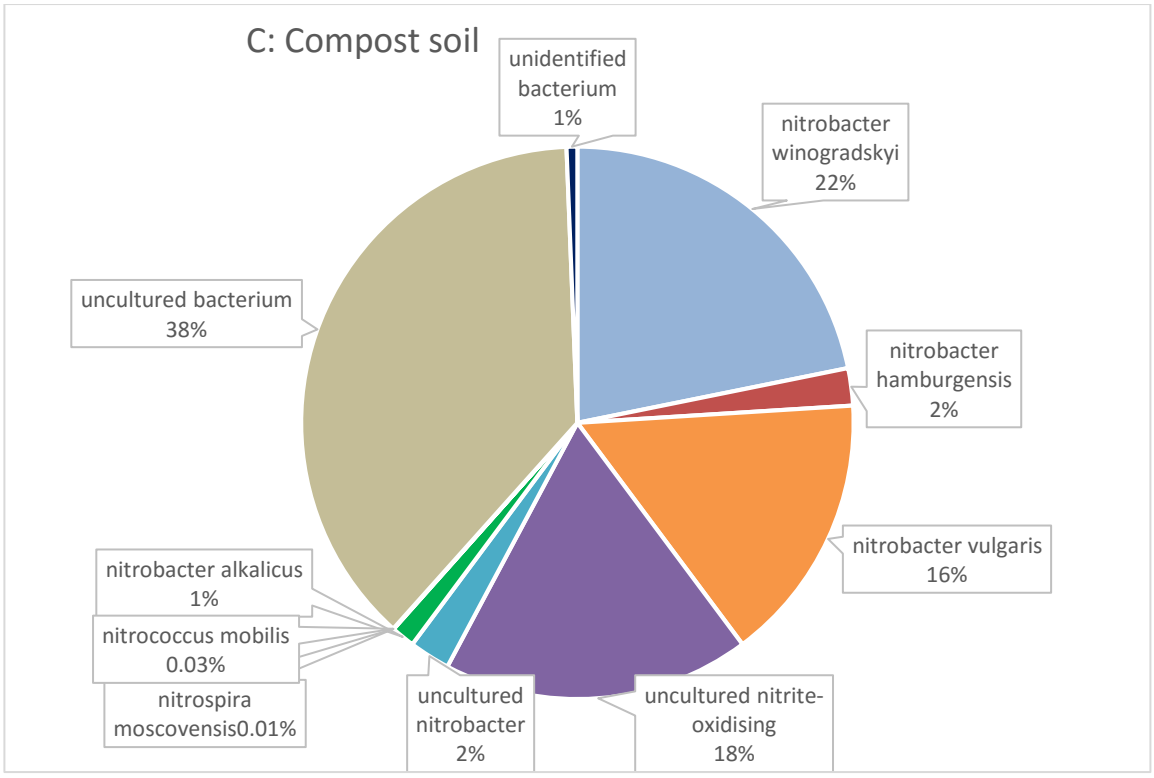
The mean daytime soil temperature ranged between 14.8°C and 25°C. The lower daytime temperature was recorded on days when it was cold and misty, whereas the higher daytime temperature was recorded on days when it was sunny and warm. The water field capacity (WFC) for garden soil was 73% and 65% for compost soil. The latter is expected to be more porous than the former in terms of water saturation.

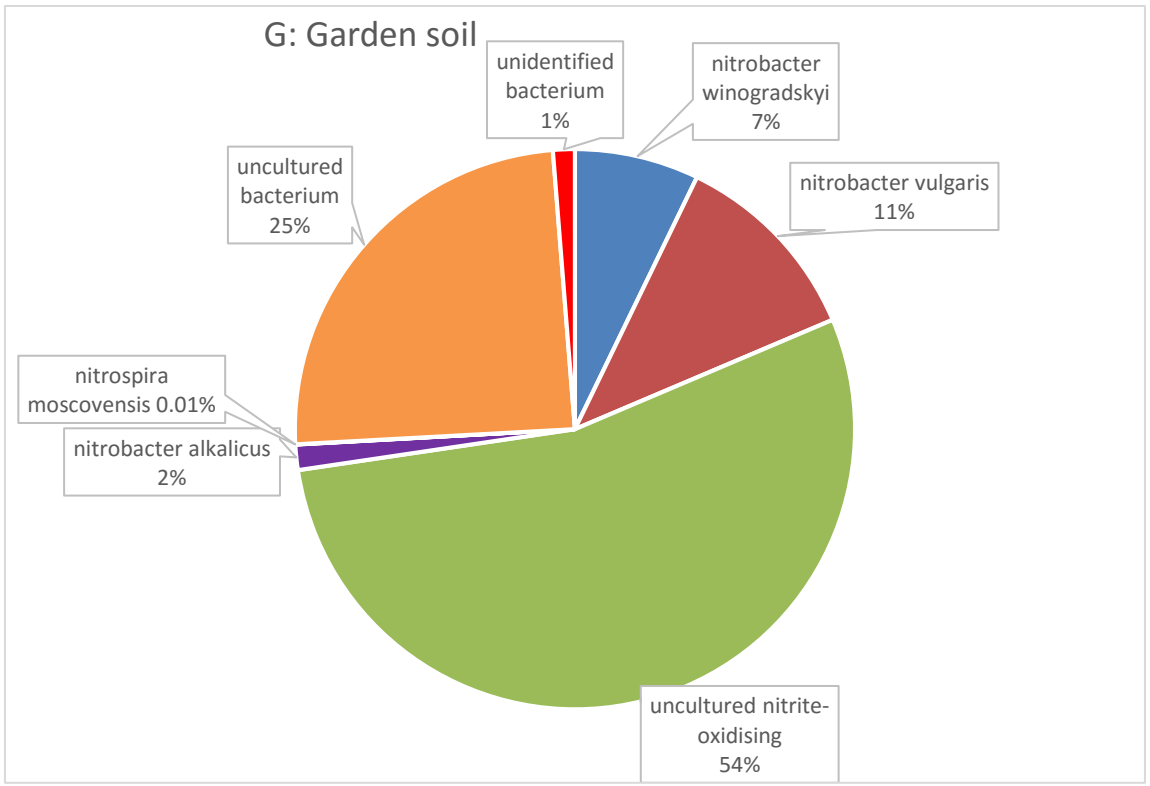
Table 6b below indicates that the two sources of inoculum (i.e. garden vs compost) contained the microbial species as shown.

**Table 6b: Nitrite Oxidizing Bacteria (NOB) species composition from the 2 sources of inoculum and species diversity**

| Community of microbial organisms as distinguished from *metagenomic analysis | Inoculum source | | |
|---|---|---|---|
| | Compost | Garden | Magnitude of increase in compost |
| *Nitrobacter winogradskyi* | 3150 | 1021 | x 3.08-fold |
| *Nitrobacter hamburgensis* | 315 | | x 315-fold |
| *Nitrobacter vulgaris* | 2281 | 1626 | x 1.4-fold |
| *uncultured nitrite-oxidising* | 2598 | 7687 | x 0.3-fold |
| *uncultured nitrobacter* | 346 | | x 346-fold |
| *Nitrobacter alkalicus* | 200 | 208 | x 0.9-fold |
| *Nitrospira moscovensis* | 4 | 2 | x 2-fold |
| *Nitrococcus mobilis* | 1 | | x 1-fold |
| *uncultured bacterium* | 5450 | 3499 | x 1.6-fold |
| *unidentified bacterium* | 91 | 179 | x 0.5-fold |
| **SUM** | **14436** | **14222** | |
| **Simpson's diversity index** | **0,75** | **0,63** | |

*see Appendix 1 (C & D)




**Figures 5c&5g: Relative abundance of the various communities of the total Nitrite-Oxidising Bacteria (NOB) metagenome isolated from the compost source (C) and from the garden soil source (G).**

### 3.3.4. Concentration of microorganisms

As an initial step, interest was in getting to know the density of inoculum in varying quantities in compost soil of which amounts are shown as high (10g), medium (5g), and low (1g) quantity treatments (Figure 6). According to Figure 6 and Table 7, a high inoculum treatment (10g) compared to the medium (5g) and low (1g) gave significantly ($P \leq 0.05$) the highest nitrate density in 21 days.

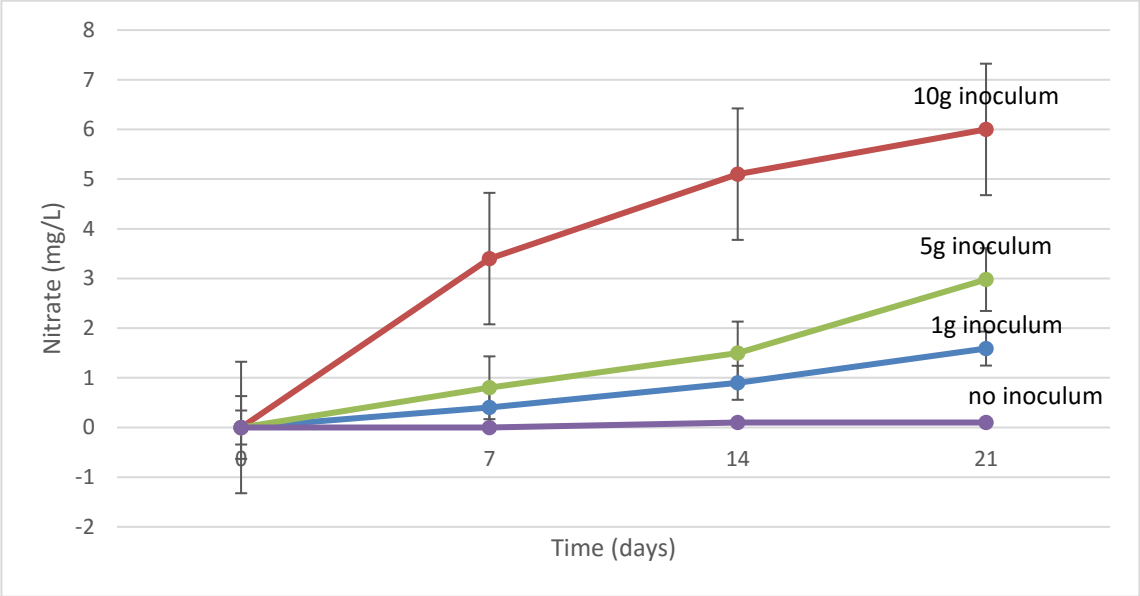



**Figure 6: Inoculum requirements with SANUMARC compost.**

**Table 7: Descriptive statistics for source of microorganisms between the 3 treatments**

| Treatment | N | Mean | Std. Deviation | CV | Std. Error |
|---|---|---|---|---|---|
| No inoculum | 6 | .133 | .0516 | 38.79699 | .0211 |
| 1g inoculum | 6 | 1.517 | .0753 | 4.963744 | .0307 |
| 5g inoculum | 6 | 2.900 | .2280 | 7.862069 | .0931 |
| 10g inoculum | 6 | 6.000 | .6325 | 10.54167 | .2582 |

### 3.3.5. Manure for optimal nitrate generation by inoculum

The first goat manure density was 0.25g/L added daily for 15 days to the 10g/L inoculum density (Figure 7). The others were done in densities of 0.5g/L and 1g/L added daily for 15 days. A noticeable effect of the nitrate release started to appear at 12 days after the addition of goat manure (Figure 7). More significantly ($P \leq 0.05$) the 0.25g/L manure was highest in generating nitrates up to day 50 after the start of trial.

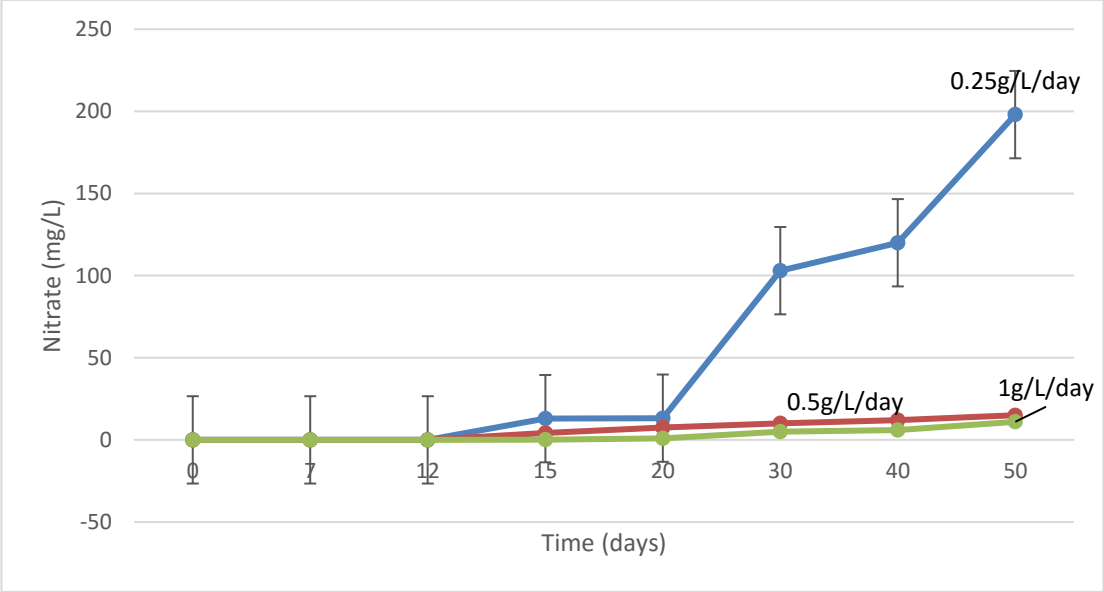



**Figure 7: Optimum amount of organic fertilizer.**

**Table 8: Descriptive statistics for optimum concentration of manure between the 3 treatments**

| Treatment | N | Mean | Std. Deviation | CV | Std. Error |
|---|---|---|---|---|---|
| 0.25g/L/day manure | 6 | 197.17 | 1.169 | 0.592889 | .477 |
| 0.50 g/L/day manure | 6 | 15.00 | .894 | 5.96 | .365 |
| 1.00 g/L/day manure | 6 | 10.50 | .548 | 5.219048 | .224 |

At the end of the above trial levels of mineral nutrients were bio-assayed as shown in Table 9. It can be noticed that in contrast to a very low nitrate density in garden soil, nitrates accumulation in the Compost inoculum plus goat manure was found to be in the usual range of the nitrate expected (198mg/L as obtained in this study vs 224mg/L in conventional hydroponic nutrient solution).

**Table 9: Levels of mineral nutrients in the formulated organic nutrient solution**

| Element | Symbol | Mg/L *(as reported elsewhere) | Mg/L **(Obtained) | Magnitude of observed |
|---|---|---|---|---|
| Potassium | K | 371.5 | 360 | x 0.9-fold |
| Calcium | Ca | 216.5 | 250 | x 1.2-fold |
| Magnesium | Mg | 58.3 | 67 | x 1.2-fold |
| Phosphorus | P | 46.5 | 42.1 | x 0.9-fold |
| Sulfur | S | 140.9 | 198 | x 1.4-fold |
| Nitrate | NO3 | 223.9 | 198 | Not unusual variation |
| Electrical conductivity | EC | *1 – 4.5 dS $m^{-1}$ | 3.2 dS $m^{-1}$ | |
| pH | 1.5 slurry | *recommended 6.0 -7,3 | 6.5 | Within similar range |

*Schwarz & Kuchenbuch, 1998; Buck *et al.,* 2004. ** results from current study.

The implications of the mineral nutrients in relation to the pH are as presented in Table 10. The initial pH in the nutrient solution was 7.4 and it was adjusted to the recommended pH 6.5 (Buck *et al.,* 2004) by adding citric acid.

**Table 10: Implications of pH levels on macro-nutrients availability as a reflection of hydroponic nutrient status**

| This study's analyses | *Standard reference [A] **(mol/L):** | This study's pH compared to $*10^{-7}$ & $10^{-6}$ | Remarks |
|---|---|---|---|
| S mg/L | | 6.5 | |
| K mg/L | | 6.5 | K was almost completely present as a free ion in this study's hydroponic nutrient solution since it can be accommodated within a pH range of 2 and 9. |
| Mg mg/L | | 6.5 | Available to plants in a wide range of pH |
| Ca mg/L | | 6.5 | Available to plants in a wide range of pH |
| NO3 mg/L | | 6.5 | |
| P mg/L | | 6.5 | It is imputed that at this study's pH of 6.5 lying between 5.97 & 6.4 will be expected to have between 94.5 & 74.5% [$H_2PO-4$]/[A]. However, it is assumed that the pH is close to p$K_{a1}$, giving an equimolar mixture of $H_3PO_4$ and $H_2PO-4$ according to Trejo-Téllez & Gómez-Merino (2012). At this study's pH the nutrient solution seems to have predominated as the HPO4-2 ion according to the expectation of the last 2 rows (Trejo-Téllez & Gómez-Merino, 2012). |
| | $*10^{-7}$; | 6.74 | [$H_2PO-4$]/[A]: 74.5%; [$HPO-4$]/[A]:25.5%; [$PO_3-4$]/[A]: 3.02 x $10^{-5}$;% |
| | $*10^{-6}$; | 5.97 | [$H_2PO-4$]/[A]: 94.5%; [$HPO-4$]/[A]:5.5%; [$PO_3-4$]/[A]: 1.1. x $10^{-6}$;% |

*As tabulated by Trejo-Téllez & Gómez-Merino (2012) in order to evaluate the ionic status of a given hydroponic nutrient. pKa is a measure of a compound's ability to lose a hydrogen ion. The two are related but distinct, and for the most part cannot be calculated from the other without additional information.

## 3.4 DISCUSSION

The current study's result which showed that the observed levels of mineral nutrients in the goat manure collected from Utuseb farm in the Erongo region were at good levels

(Table 4) as such this was a good resource to be used for formulating an organic hydroponic nutrient solution considering that all the mineral nutrients were sufficiently available to meet minimum requirements for plant production, in exception of nitrogen which was predominantly organic than plant available nitrate (0.072 mg/L). In comparison to goat manure from other places such as South Africa (Azeez & Van Averbeke, 2010), Sri Lanka (Bandara & Tennakoon, 2003), the United States of America (Pratt & Castellanos, 1981), and Spain (Moreno-Caselles *et al.*, 2002), the current findings suggest that goat manure in these countries has the same macronutrient levels indicating similarities in diets. Therefore, results from the present study will be applicable to goat manure from such countries with similar goat feeds.

The present study's result show that Sulfur was more in the observed compared to levels reported elsewhere may be attributed to the fact of proximity of Utuseb farms to the Atlantic Ocean (30-40km), whereby such areas in the Namib desert like Utuseb have been known to get the marine Sulfur accumulates that are moved and deposited by wind as dust (Soderberg *et al.*, 2014). Therefore, Sulfate levels may have accumulated as they got deposited on the manure over time, considering the manure was kept over 3 months.

Indeed, the result of the current study show that there was more nitrate generated with the presence of inoculum as opposed to when there was no inoculum implying that locally sourced microorganisms can be used for the use as inoculum to convert organic nitrogen into nitrates. This is important in that there is no requirement of bringing microorganisms from different environments with different settings such as temperature, moisture, pH and

EC which may have required acclimatisation of such microorganisms before they are used.

The result also show that Compost inoculum was the best source of nitrifying bacteria over garden soil is attributed to the fact that there was a high preponderance of ammonia-oxidizing species (64%) in the Compost inoculum than the Garden inoculum where, in contrast there was rather a high preponderance of uncultured bacterium (51%). This means there were more microorganisms converting ammonia into nitrites in the ammonia-oxidising species found in the Compost source than in the uncultured bacterium found in the Garden source. This is in agreement with Rughöft *et al* (2016) whose work indicated that preponderance of ammonia-oxidising bacteria was positively associated with nitrate generation. Furthermore, Saijai *et al* (2016) have also shown that preponderance of Nitrobacter species in media is also positively correlated with nitrate generation. Moreover, the current results may mean that either the uncultured bacterium (unidentified species) were either competed out of the micro-organismic community (metagenome) or were reduced to their minimum in relation to ammonia oxidizing activity.

Furthermore, in addition to the preponderant ammonia-oxidising species in the Compost source, a group of microorganisms that are known to convert nitrite into nitrates were also found more preponderant in the Compost source than in the Garden source. These are the *Nitrobacter* metagenome (43% in Compost source compared to 20% from the Garden source. Moreover, the results may mean that there are other unculturable yet important microbes doing the same job if not better than the known ones, in this case suggesting that there may be other local ammonia-oxidising bacteria responsible for oxidizing ammonia

other than the traditionally known *Nitrosomonas* and *Nitrosospira* species. This is considering that nitrifying bacteria picked up by the *Amo*A gene encoding the active site polypeptide of the ammonia monooxygenase (*Amo*A) in the samples are dominated by species unidentified. Given these results therefore, it is not prudent to use Garden soil over Compost soil.

The present study found that the higher inoculum density treatment of 10g/L produced more nitrates than other densities. This may mean that the more concentrated the inoculum is, the more efficient it will be to generating nitrates from goat manure. Preponderance is therefore, associated with density. This is important because without knowing how much of the inoculum to use on goat manure, resources may be wasted in several trials using less concentration.

Finally, the present study also found that at the initial fermentation stage, adding small amounts of manure (0.25 g/L) compared to large amounts of manure encourages the establishment of the microbial ecosystem required for subsequent mineralisation of organic nitrogen compounds. This therefore, may mean that nitrification can be hindered by excessive amounts of goat manure during the fermentation stage. Shinohara *et al* (2011) found similar results when mineralising organic nitrogen in fish-based fertiliser, where they found that lower doses of manure (less than 0.5g/L) in the first days of fermentation encouraged the establishment of microbial consortia which would later mineralise large amounts of organic nitrogen from fish-based fertiliser into sufficient nitrates for plant growth.

The levels of mineral nutrients found in this study are similar to those reported by other researchers such as Shinohara (2011), Peet & Welles (2005); Wamcke & Krauskopf (1983); and Adams (1986). Though the EC in the formulated nutrient solution is relatively higher than that of conventional hydroponic fertiliser (HAIFA), it is still within tolerable range for crops such as tomato considering the highest range recommended by Buck *et al* (2004) was 4.5 dS $m^{-1}$ for tomato yield in contrast to using 2.6 dS $m^{-1}$. Moreover, the initial levels of other nutrients in the raw unprocessed manure changed during the processing, resulting in levels comparable to those from the conventional hydroponic nutrient solutions. This means that processing of manure in the manner it was done is a requirement to get nutrient levels required for plant performance. Therefore, based on the macro nutritional profile of the formulated nutrient solution from this study, a good alternative hydroponic nutrient solution has been created and available for growth of crops such as tomato. According to Liebig's law of the minimum, nitrogen which was the limiting nutrient in the nutrient solution from goat manure has now been provided in sufficient amounts to render this organic nutrient solution suitable for plant production (Shinohara, 2011).

## 3.5 CONCLUSION

Based on the current results, it can be concluded that using local microbial consortia from SANUMARC natural compost (Erongo region), it is possible to formulate an organic hydroponic nutrient solution from local goat manure that is alternative to the conventional hydroponic nutrient solution. Such organic hydroponic nutrient solution may be used to grow tomato plants in the Erongo region.

Garden soil and compost are appropriate sources of microorganisms to generate nitrate from goat manure in water. There are other unknown local microorganisms oxidising ammonia and nitrite and the presence of these nitrifying bacteria in addition to the presence of the known ammonia-oxidizing species in the garden soil and compost samples infers their roles in the observed nitrification. Therefore, nitrifying microbial consortia that would degrade goat manure for use in organic hydroponics can be sourced from any place with similar environment (temperature, pH and soil) to Henties Bay. Further research can consider determining ammonium levels as well from nitrification of goat manure and establish the optimal rate of nitrification by a certain amount of soil containing nitrifying microbial consortia.

## REFERENCES


1. Adams, P. (1986). Mineral nutrition. In *The tomato crop* (pp. 281-334).
2. Arroukatchee (2017). Map of Henties Bay. http://www.southern-africa.arroukatchee.fr/namibia/towns/henties-bay.htm
3. Azeez, J.O., and Van Averbeke, W. (2010). Nitrogen mineralization potential of three animal manures applied on a sandy clay loam soil. *Bioresource technology*, *101*(14), 5645-5651.
4. Bandara, S.D.H., and Tennakoon, N.A. (2003). Nutrient content of some locally available organic materials and their potential as alternative sources of nutrients for coconut. *COCOS, 15*, 23-30.
5. Biati, A., Karbassi, A.R., Hassani, A.H., Monavari, S.M., and Moattar, F. (2010). Role of metal species in flocculation rate during estuarine mixing. *International Journal of Environmental Science & Technology*, *7*(2), 327-336.
6. Buck, J.S., Kubota, C., Wu, M. (2004). Effects of nutrient solution EC, plant microclimate and cultivars on fruit quality and yield of hydroponic tomatoes (Lycopersicon esculentum). In *VII International Symposium on Protected Cultivation in Mild Winter Climates: Production, Pest Management and Global Competition* 659 (pp. 541-547)
7. HAIFA. (n.d.). Nutritional recommendations for tomato in open-field, tunnels and greenhouse. Retrived from URL: www.haifa-group.com/files/Guides/tomato/Tomato.pdf.
8. Hussain, F., and Shah, S.Z. (2014). Direct effects of phosphates concentration on the microalgal growth in Malakand Pakistan. *Pak. J. Weed Sci. Res*, *20*(2), 199-206.

9. Lupwayi, N.Z., Girma, M., and Haque, I. (2000). Plant nutrient contents of cattle manures from small-scale farms and experimental stations in the Ethiopian highlands. *Agriculture, ecosystems & environment, 78* (1), 57-63.
10. Moir, C.M., Freedman, B., and McCurdy, R. (1996). Metal mobilization from water-distribution systems of buildings serviced by lead-pipe mains. *Canadian water resources journal*, *21*(1), 45-52.
11. Moreno-Caselles, J., Moral, R., Perez-Murcia, M., Perez-Espinosa, A., & Rufete, B. (2002). Nutrient value of animal manures in front of environmental hazards. *Communications in Soil Science and Plant Analysis*, *33*(15-18), 3023-3032.
12. Mowa, E. (2015). Organic manure for vegetable production under hydroponic conditions in arid Namibia. International Science & Technology Journal of Namibia, 2015, 5:3-12.
13. Nikodemus, O., Kasparinskis, R., & Kukuls, I. (2013). Influence of afforestation on soil genesis, morphology and properties in glacial till deposits. *Archives of Agronomy and Soil Science*, *59*(3), 449-465.
14. Peet, M.M., and Welles, G.W.H. (2005). Greenhouse tomato production. *Crop production science in horticulture, 13,* 257.
15. Poly, F., Wertz, S., Brothier, E., Degrange, V. (2008). First exploration of Nitrobacter diversity in soils by a PCR cloning-sequencing approach targeting functional gene nxrA. *FEMS microbiology ecology, 63*(1), 132-140.
16. Pratt, P.F., and Castellanos, J.Z. (1981). Available nitrogen from animal manures. *Califonia Agriculture,* 35(7), 24-24.
17. Purkhold, U., Pommerening-Röser, A., Juretschko, S., Schmid, M.C., Koops, H.P., Wagner, M. (2000). Phylogeny of all recognized species of ammonia

oxidizers based on comparative 16S rRNA and amoA sequence analysis: implications for molecular diversity surveys. *Applied and environmental microbiology, 66*(12), 5368-5382.

18. Qian, P., Schoenau, J.J. (2002). Availability of nitrogen in solid manure amendments with different C: N ratios. *Canadian Journal of Soil Science, 82*(2), 219-225.
19. Rotthauwe, J.H., Witzel, K.P., Liesack, W. (1997). The ammonia monooxygenase structural gene amoA as a functional marker: molecular fine-scale analysis of natural ammonia-oxidizing populations. *Applied and environmental microbiology, 63*(12), 4704-4712.
20. Rughöft, S., Herrmann, M., Lazar, C.S., Cesarz, S., Levick, S.R., Trumbore, S.E., Küsel K. (2016). Community composition and abundance of bacterial, archaeal and nitrifying populations in savanna soils on contrasting bedrock material in Kruger National Park, South Africa. *Frontiers in Microbiology*, 7 (OCT), art. no. 1638.
21. Saijai, S., Ando, A., Inukai, R., Shinohara, M., Ogawa, J. (2016). Analysis of microbial community and nitrogen transition with enriched nitrifying soil microbes for organic hydroponics. Bioscience, Biotechnology, and Biochemistry, 80(11), 2247-2254.
22. Schwarz, D., and Kuchenbuch, R. (1998). Water uptake by tomato plants grown in closed hydroponic systems dependent on the EC-level. In *International Symposium on Water Quality & Quality-Greenhouse 458* (pp. 323-328).
23. Sen, I., Shandil, A., and Shrivastava, V.S. (2011). Study for determination of heavy metals in fish species of the river Yamuna (Delhi) by inductively coupled

plasma – optical emission spectroscopy (ICP-OES). *Advance in Applied Science Research, 2* (2), 161-166.

24. Shinohara, M., Aoyama, C., Fujiwara, K., Watanabe, A., Ohmori, H., Uehara, Y., & Takano, M. (2011). Microbial mineralization of organic nitrogen into nitrate to allow the use of organic fertilizer in hydroponics. *Soil science and plant nutrition, 57*(2), 190-203.
25. Soderberg, K., Henschel, J., Swap, R.J., and Macko, S.A. (2014). Sulphur isotopes in the central Namib Desert ecosystem. *Transactions of the Royal Society of South Africa*, *69*(3), 217-223.
26. Trejo-Téllez, L.I., and Gómez-Merino, F.C. (2012). Nutrient solutions for hydroponic systems. In *Hydroponics-A Standard Methodology for Plant Biological Researches*. InTech.
27. Wamcke, D.D., and Krauskopf, D.M. (1983). Greenhouse growth media: Testing and nutrition guidelines. *Extension Bulletin E, 1736.*
28. Zhang, M., Lawlor, P.G., Lynch, B., and Zhan, X. (2011). Partial nitrification and nutrient removal in intermittently aerated sequencing batch reactors treating separated digested liquid after anaerobic digestion of pig manure. *Bioprocess and Biosystems engineering,* 34(9), 1049-1056.

# CHAPTER 4: CROP PERFOMANCE RESPONSE TO APPLIED FORMULATED NUTRIENT SOLUTION


## ABSTRACT

The influence of organic hydroponic nutrient solution on tomato growth and yield was studied using a randomized complete block design (RCBD) with three treatments (goat manure-derived hydroponic nutrient solution, commercial hydroponic nutrient solution and tap water) replicated three times. Determinate tomato seeds were germinated and transplanted into the 3 treatments. Twenty days after transplanting, vegetative response variables were recorded at fifteen days intervals from each treatment. Vegetative response variables included plant height, number of leaves and stem diameter. The reproductive parameters included number of flowers per plant, number of fruits per plant, average fruit weight (g), yield (kg) per plant and yield (kg) per treatment. Furthermore, the organic nutrient solution formulated from goat manure positively improved plant growth and yield performance of a tomato crop, and provided a technique feasible and alternative to conventional hydroponics. These results open further possibilities for other crops currently grown in hydroponics using conventional fertilizers.



## KEYWORDS

Goat-derived hydroponic nutrient solution, tomato, vegetative, reproductive, growth.


## 4.1 INTRODUCTION

The first part of this chapter gives the problem statements, objectives and hypothesis of the chapter. The second part of this chapter then details the procedure or materials and methods used to collect and analyse data for the objective of this chapter. The last part of this chapter presents the results found from the collection and analysis of data for this chapter as well as the discussion and conclusion of the chapter.

### 4.1.1 Statement of the Problem

The gap arising from the literature review (chapter 2) is that an alternative hydroponic nutrient solution formulated from goat manure has never been tested on plant performance in terms of growth and reproduction. This is considering the goat manure-derived hydroponic nutrient solution formulated in chapter 3 of this study needed to be tested on a crop in order to determine its efficacy. Moreover, considering the current study is the first of its kind in Africa and Namibia, there is a need to establish plant performance in terms of nutrient use efficiency of nitrate nitrogen available in the formulated nutrient solution from goat manure.

### 4.1.2 Objective

To determine performance of tomato due to the developed hydroponic solution

### 4.1.3 Hypothesis

Goat manure-derived hydroponic nutrient solution will positively influence tomato plant growth and reproductive performance.

## 4.2 MATERIALS AND METHODS

### 4.2.1 Experimental setup

To determine the effect of the organic hydroponic nutrient solution on growth and yield of tomato, hydroponic tables (22.8 cm X 250 cm X 150 cm) constructed from planks and

black sheeting were used in a greenhouse at the Sam Nujoma Marine and Coastal Research Centre (SANUMARC), Henties Bay. Styrofoam that completely cover the hydroponic tables yet float on water in the table were added. Each hydroponic table covered an area of 3.75 $m^2$. A randomized complete block design (RCBD) with three treatments each triplicated (organic nutrient solution, conventional hydroponic fertiliser (as positive control) and plain tap water (as negative control). Each replicate was a hydroponic table of 6 plants and served as an experimental unit. Organic nutrient solution was formulated in 2016 at SANUMARC, Henties Bay using natural compost containing Ammonia and Nitrite Oxidising Bacteria to convert organic Nitrogen into nitrates from goat manure. The positive control treatment was a nutrient solution made from the conventional hydroponic fertilisers; Calcium Nitrate and Hygrotech. The final nutrient solution made from the 2 fertilisers consisted of: Calcium (Ca)217 mg/L, Nitrate (N) 225 mg/L, Phosphate (P) 46.5 mg/L, Potassium (K) 372 mg/L, Magnesium (Mg) 58 mg/L, Sulphur (S) 141 mg/L. Organic nutrient solution had Nitrate (N) 198mg/L, Phosphate (P) 42.1mg/L, Potassium (K) 360mg/L, Magnesium (Mg) 67 mg/L, Sulphur (S) 198 mg/L, Calcium (Ca) 250. The negative control used was plain tap water from Henties Bay municipality with $\leq$ 0.05mg/L nitrate levels.

The formulated organic hydroponic nutrient solution was therefore, compared with the conventional nutrient solution according to the method by Shinohara *et al* (2011) and the negative control (Table 11). Determinate tomato seeds (Settler) were germinated on coconut husks in polystyrene trays on 23 February 2017 and transplanted into 3 treatments (conventional fertiliser & organic nutrient solution and plain water) on 24 March 2017.

**Table 11: treatments for investigating performance of tomatoes**

| Treatment | Nitrate concentration (mg/L) |
|---|---|
| Treatment 1: Negative control<br>(Tap water without fertiliser) | ≤0.05 |
| Treatment 2: Positive control<br>(Commercial (conventional) fertilizer @ 225mg/L NO3-) | 223.9 |
| Treatment 3: Manure nutrient solution<br>(Formulated organic nutrient solution @ 198mg/L NO3-) | 198 |

Twenty days after transplanting, vegetative response variables were recorded at fifteen days' intervalsfrom each treatment (Liu *et al.,* 2014). The nutrient solutions were used once for the entire experiment without changing from transplanting to harvesting. Therefore, between the 2 experiments, separate nutrient solutions were formulated and used only once and disposed off thereafter.

### 4.2.2 Vegetative growth

Response variables included plant height (cm) measured from the ground to the main apex, number of leaves and stem diameter measured 10 cm above the root level. A RCBD was used whereby each treatment was triplicated with 6 plants per replicate for vegetative growth.

### 4.2.3 Reproductive growth

The reproductive parameters included number of flowers per plant, number of fruits per plant, average fruit weight (g), yield (kg) per plant and yield (kg) per treatment. In order to determine yield, tomato fruits were picked and weighed when they had reached full

maturity at harvest time (Mehdizadeh *et al.*, 2013). In addition, number of days to first flowering and number of days to first harvest were also recorded in accordance to Shah *et al* (2011). Therefore, a RCBD was used whereby each treatment was triplicated with 6 plants per replicate for reproductive growth. The whole experiment was repeated twice at different time intervals.

### 4.2.4. Nutrient Use Efficiency

Nutrient use efficiency (NUE) was evaluated according to Zotarelli *et al* (2009) whereby NUE was defined as nitrate nitrogen recovery efficiency, nitrate nitrogen removed from the nutrient solution divided by the total amount of nitrate nitrogen supplied at the initial planting stage in the nutrient solution (Atiyeh *et al.*, 2001; Zotarelli *et al.*, 2009; Grewal *et al.*, 2011). An RCBD was used whereby each treatment was triplicated with 6 plants per replicate for NUE. Furthermore, NUE was defined as agrophysiological efficiency whereby the yield obtained per unit of nutrient up-taken was recorded (Baligar *et al.*, 2001).

### 4.2.5 Plant dry biomass

Plant dry biomass was determined according to Atiyeh *et al* (2000) and Zaller (2007), whereby five (5) tomato plants (whole plant, above-ground parts and roots) per treatment were randomly selected and harvested at the last fruit harvest (at the end of the experiment) and oven-dried at 80°C for at least 24 hours to determine total plant dry weights.

### 4.2.6 Statistical analysis

Means comparisons analysis were applied whereby, a One-way Analysis of Variance (ANOVA), followed by mean separation by Least Significant Difference (LSD) was used when ANOVA determined that the effects of the treatments were significant ($P \leq 0.05$ for F-test).

## 4.3 RESULTS

### 4.3.1 Vegetative growth

Tomato plant height increased over time and was high in plants growing under Goat Manure Derived Hydroponic Nutrient Solution (GMDHNS) which produced the same tomato plant height as the Commercial Hydroponic Nutrient Solution (CHNS). In relation to the Tap Water (TW) treatment, GMDHNS and CHN significantly ($P \leq 0.05$) increased tomato plant height (Figure 8 and Table 12).

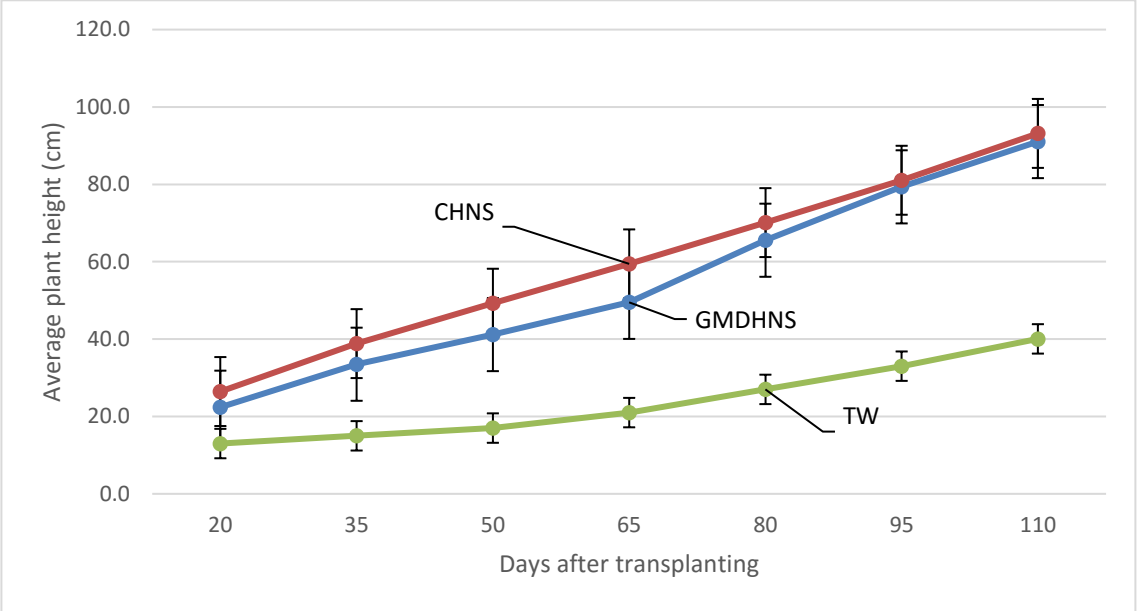



**Figure 8: Effect on plant height of tomatoes grown in Commercial Hydroponic Nutrient Solution (CHNS), Goat Manure Derived Hydroponic Nutrient Solution (GMDHNS), & Tap Water (TW) nutrient solutions.**

**Table 12: Descriptive statistics for tomato plant height in the 3 different treatments**

| Treatment | N | Mean | Std. Deviation | CV | Std. Error |
|---|---|---|---|---|---|
| CHNS | 18 | 93.17 | 2.640 | 2.83 | .622 |
| GMDHNS | 18 | 91.06 | 4.795 | 5.26 | 1.130 |
| TW | 18 | 40.06 | 7.658 | 19.1 | 1.805 |

GMDHNS produced 19% more leaves than the TW treatment whereas CHNS produced 8% more than GMDHNS. Therefore, tomato plants produced significantly ($P \leq 0.05$) more leaves when grown in the goat manure derived hydroponic nutrient solution than in tap water even though less than in the commercial hydroponic nutrient solution (Figure 9 and Table 13).

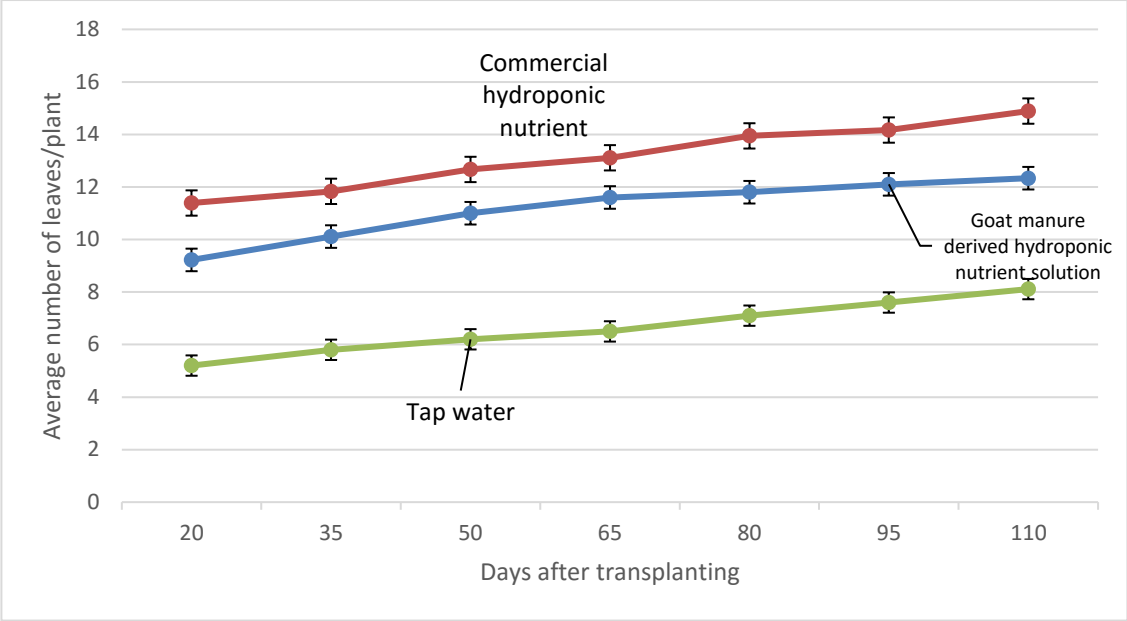



**Figure 9: Effect of different nutrient solutions on the number of leaves produced by tomato plants**

**Table 13: Descriptive statistics for the number of leaves per plant between the 3 treatments**

| Treatment | N | Mean | Std. Deviation | CV | Std. Error |
|---|---|---|---|---|---|
| CHNS | 18 | 14.89 | 4.185 | 28.1 | .986 |
| GMDHNS | 18 | 12.33 | 2.301 | 18.6 | .542 |
| TW | 18 | 8.11 | 1.023 | 12.5 | .241 |

Growing tomato plants in either manure derived hydroponic nutrient solution or in the commercial hydroponic nutrient solution produced the same plant stem diameter. In contrast growing tomato plants in tap water produced significantly ($P \leq 0.05$) less stem diameter (less than 2 cm) compared to both goat manure-derived hydroponic nutrient solution and the commercial hydroponic nutrient solution (Figure 10 and Table 14).

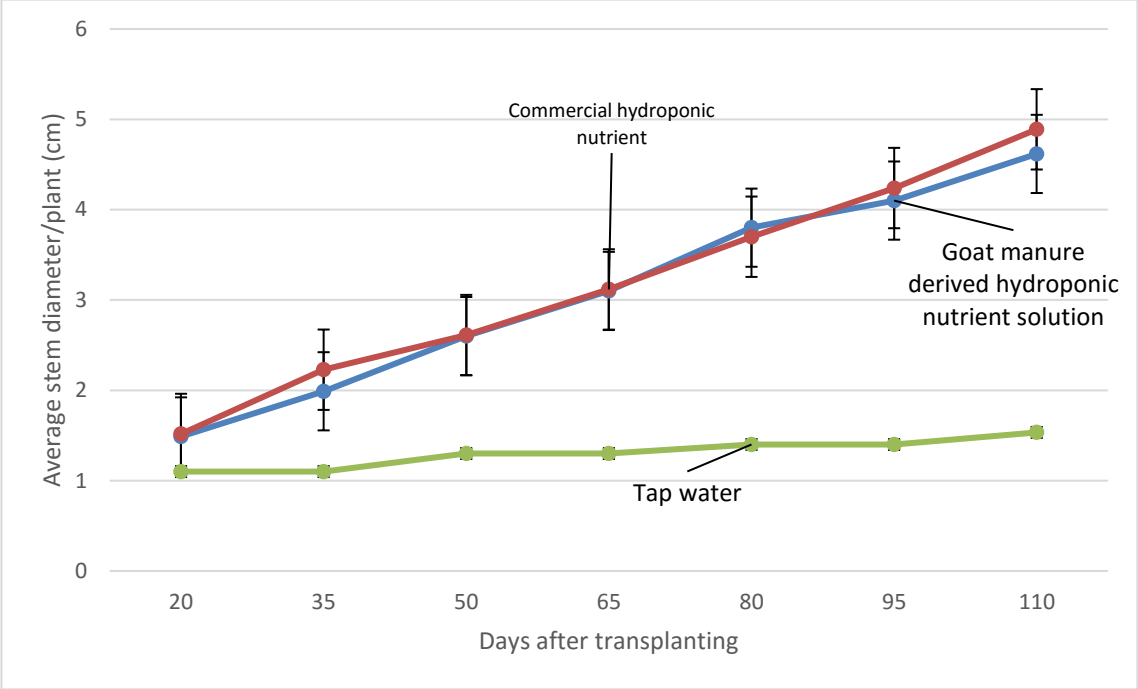



**Figure 10: Effect of different nutrient solutions on stem diameter of tomato plants**

**Table 14: Descriptive statistics for tomato plant stem diameter between the 3 treatments**

| Treatment | N | Mean | Std. Deviation | CV | Std. Error |
|---|---|---|---|---|---|
| CHNS | 18 | 4.888889 | 1.1318329 | 23.1 | .2667756 |
| GMDHNS | 18 | 4.616667 | .5283159 | 11.4 | .1245252 |
| TW | 18 | 1.533333 | .3662208 | 24.4 | .0863191 |

### 4.3.2 Reproductive growth

Tomato plants grown in Goat Manure Derived Hydroponic Nutrient Solution (GMSHNS) produced 34% more flowers than the Tap Water (TW) treatment, whereas Commercial Hydroponic Nutrient Solution (CHNS) produced 44% more flowers than the TW treatment (Figure 11). ANOVA shows that the results displayed in Figure 10 and Table 15 were significantly different.

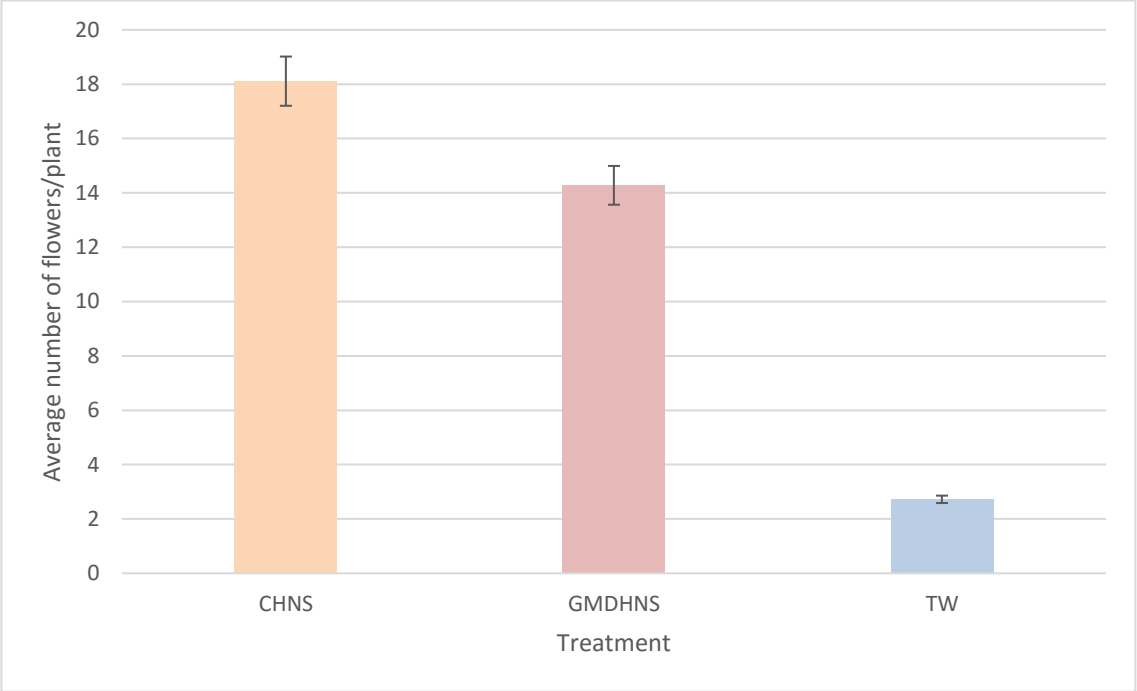



**Figure 11: Average number of flowers produced by tomato plants according to different treatments (CHNS- Commercial Hydroponic Nutrient Solution; GMDHNS – Goat Manure Derived Hydroponic Nutrient Solution; TW – Tap Water)**

**Table 15: Descriptive statistics for the number of flowers per plant between the 3 treatments**

| Treatment | N | Mean | Std. Deviation | CV | Std. Error |
|---|---|---|---|---|---|
| CHNS | 18 | 18.11 | 5.234 | 29.07 | 1.234 |
| GMDHNS | 18 | 14.28 | 5.998 | 42 | 1.414 |
| TW | 18 | 2.72 | 1.638 | 59.9 | .386 |

Growing tomato plants in the GMDHNS produced 36% more fruits than growing tomato plants in TW. CHNS produced 57% more fruits the TW treatment. ANOVA results uphold the observed trend in Figure 12 and Table 16, by indicating that these differences were significant ($P \leq 0.05$).

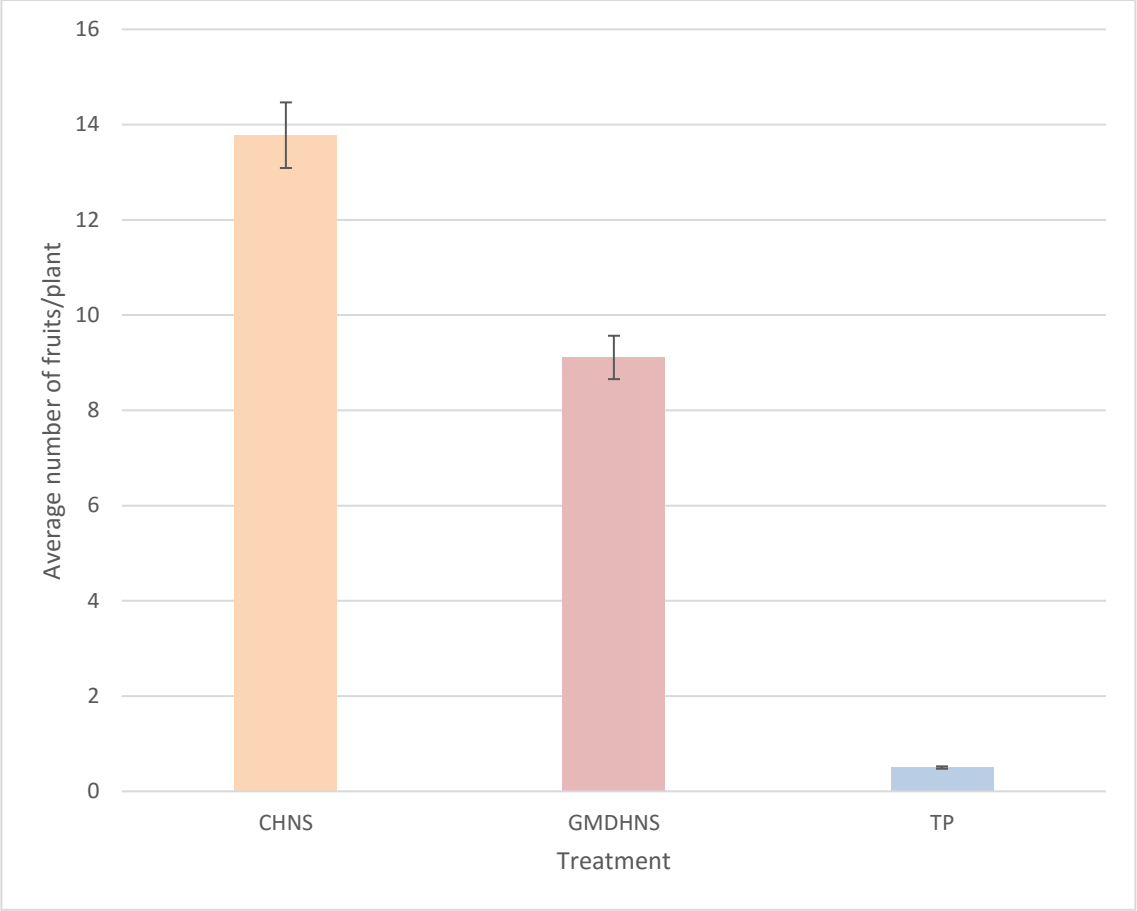



**Figure 12: Average number of fruits produced by each tomato plant according to different treatments (CHNS- Commercial Hydroponic Nutrient Solution; GMDHNS – Goat Manure Derived Hydroponic Nutrient Solution; TW – Tap Water)**

**Table 16: Descriptive statistics for the number of fruits per plant between the 3 treatments**

| Treatment | N | Mean | Std. Deviation | CV | Std. Error |
|---|---|---|---|---|---|
| CHNS | 18 | 13.78 | 2.365 | 17.1 | .558 |
| GMDHNS | 18 | 9.11 | 1.605 | 17.6 | .378 |
| TW | 18 | .50 | .098 | 19.6 | .146 |

Tomato plants grown in the GMDHNS produced a significantly ($P \leq 0.05$) higher yield than those grown in TW. Whereas tomato plants grown in CHNS produced a significantly ($P \leq 0.05$) higher yield than TW and GMDHNS (Figure 13 and Table 17).

With yields from the current study, 1050 grams of tomato can be produced per tomato plant grown in conventional hydroponic fertilizer compared to 851 grams that can be produced per tomato plant grown in manure nutrient solution. Therefore, under the current study's set-up, overall yield from the manure treatment will be 13616 kilograms per hectare or 13,6 t/ha, whereas overall yield from the conventional fertilizer will be 16800 kilograms per hectare or 16,8 t/ha.

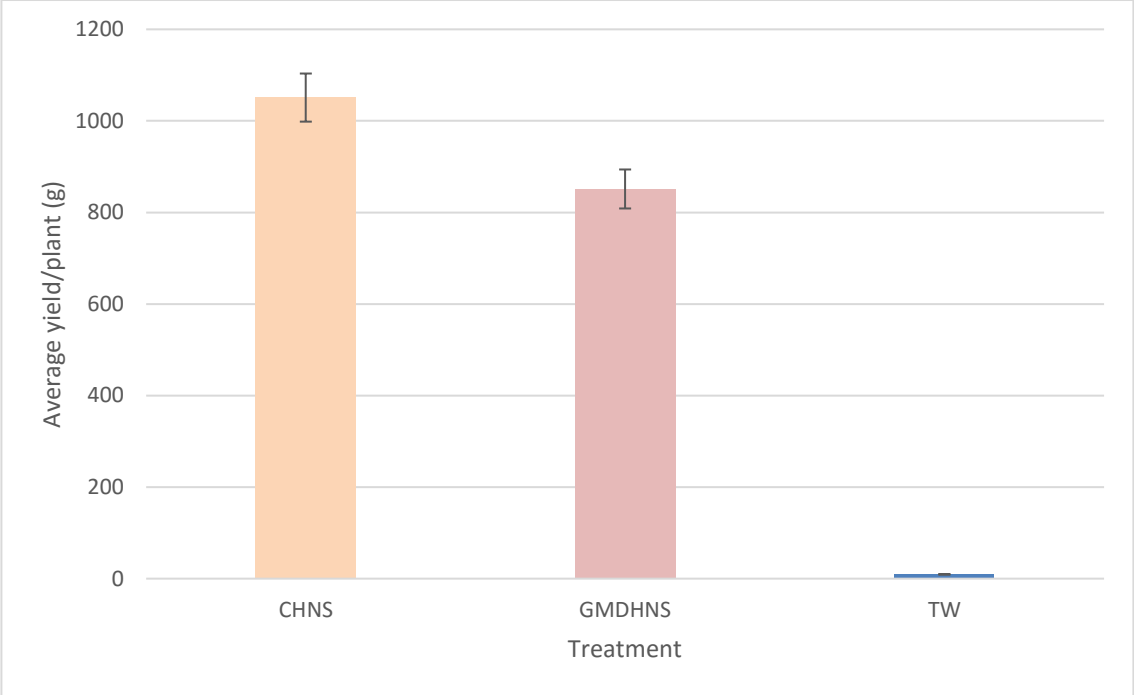



**Figure 13: Average yield per tomato plant as influenced by different nutrient solution (CHNS-Commercial Hydroponic Nutrient Solution; GMDHNS – Goat Manure Derived Hydroponic Nutrient Solution; TW – Tap Water).**

**Table 17: Descriptive for yield per plant between the 3 treatments**

| Treatment | N | Mean | Std. Deviation | CV | Std. Error |
|---|---|---|---|---|---|
| CHNS | 18 | 1050.905556 | 160.0739332 | 15.2 | 37.7297879 |
| GMDHNS | 18 | 851.272222 | 150.5382219 | 17.6 | 35.4821992 |
| TW | 18 | 9.388889 | 11.5867007 | 124 | 2.7310116 |

According to Figure 14 and Table 18, growing tomato plants in the goat manure derived hydroponic nutrient solution produced tomato fruits with a significantly ($P \leq 0.05$) higher mass (43% more) than fruits from plants grown in the tap water treatment. Growing tomato plants in the commercial hydroponic nutrient solution produced 49% more single fruit mass more than TW.

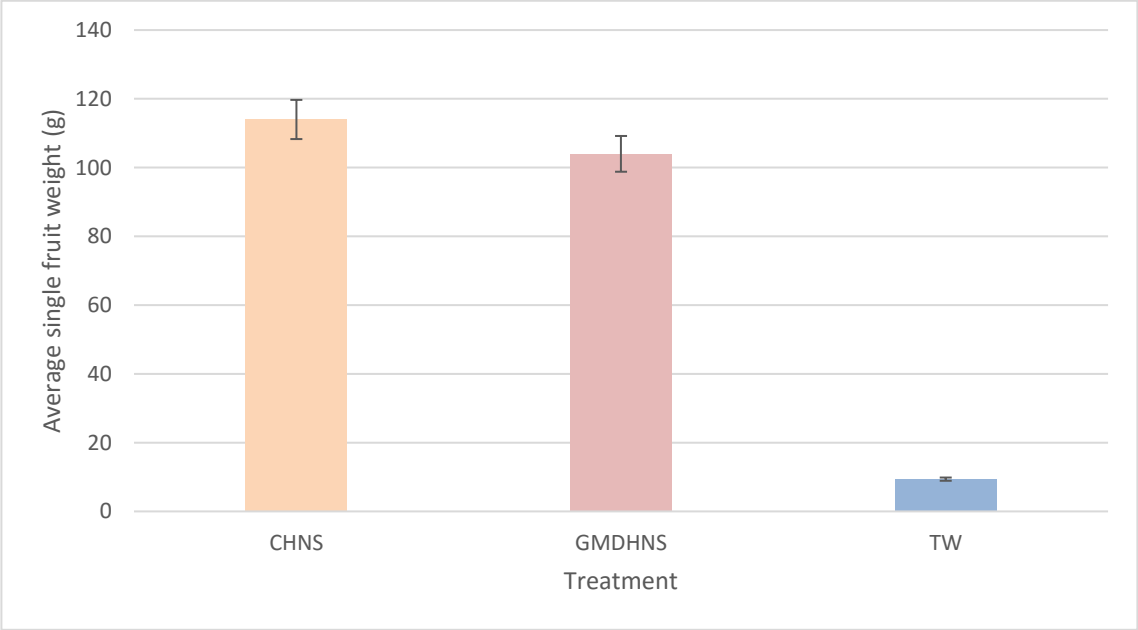



**Figure 14: Average weight of a single fruit produced by tomato plants as influenced by nutrient solution (CHNS- Commercial Hydroponic Nutrient Solution; GMDHNS – Goat Manure Derived Hydroponic Nutrient Solution; TW – Tap Water)**

**Table 18: Descriptive statistics for single fruit weight between the 3 treatments**

| Treatment | N | Mean | Std. Deviation | CV | Std. Error |
|---|---|---|---|---|---|
| CHNS | 18 | 112.256 | 6.6303 | 5.8 | 1.5628 |
| GDHNS | 18 | 106.678 | 5.9704 | 5.6 | 1.4072 |
| TW | 18 | 9.389 | 11.5867 | 122 | 2.7310 |

It took significantly ($P \leq 0.05$) more days for tomato plants to flower when grown in the tap water treatment than when grown in the goat manure derived hydroponic nutrient solution and the commercial hydroponic nutrient solution (Figure 15).

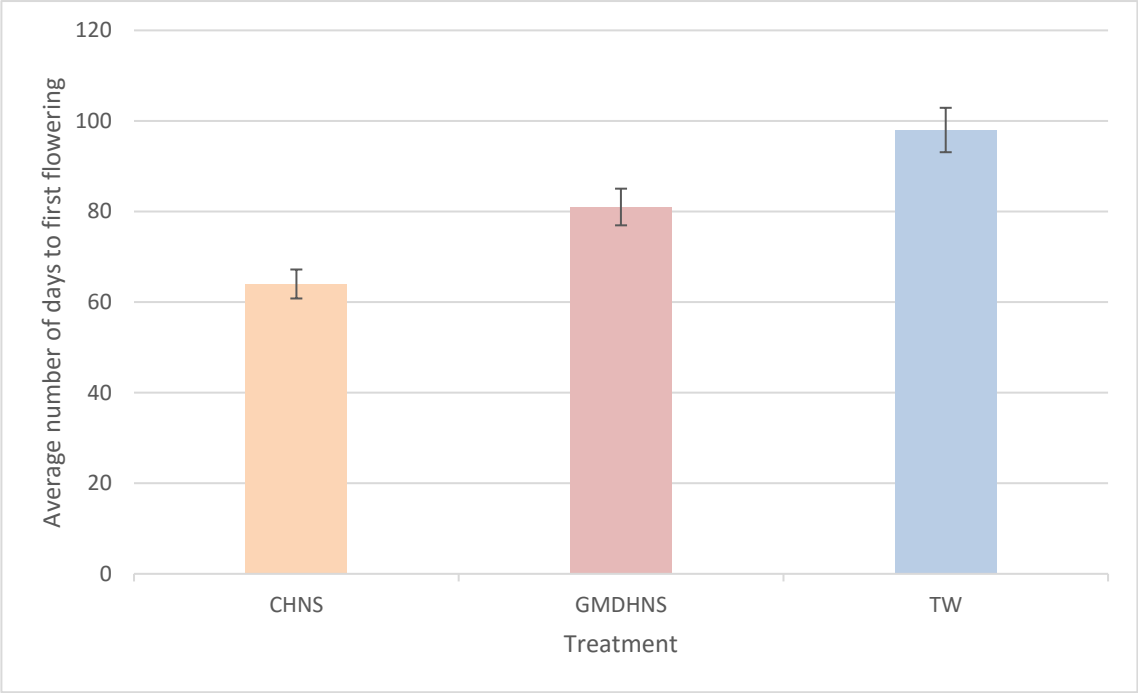



**Figure 15: Average number of days it takes for tomato plants to flower under different treatments**

On average, it took significantly longer ($P \leq 0.05$) for fruits on tomato plants grown in the TW treatment to ripen to the harvest stage compared to plants grown in the GMDHNS and in the CHNS that matured 16 days and 32 days earlier respectively (Figure 16).

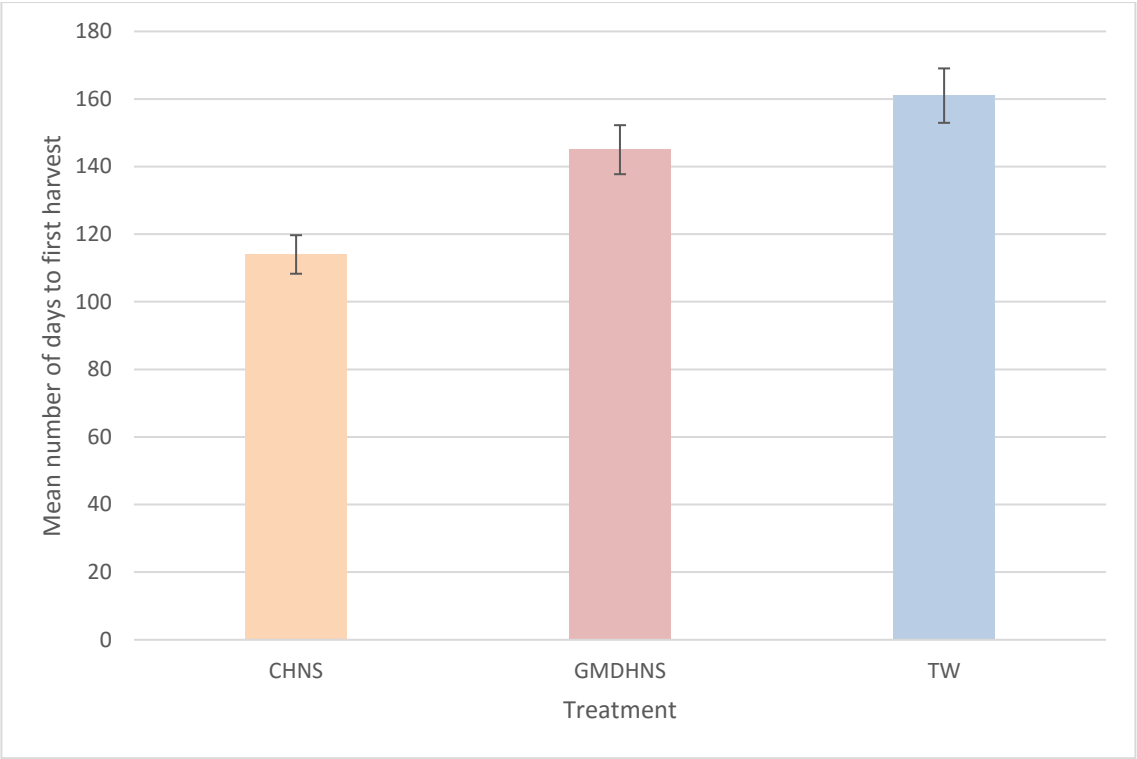



**Figure 16: Average number of days it takes to first harvests from tomato plants grown in different treatments**

### 4.3.3 Plant biomass

Tomato biomass weight at the last harvest after seeding, were greatest in the goat manure derived hydroponic nutrient solution than in the other treatments (Table 19). The difference in plant biomass was significant ($P \leq 0.05$) between tomato plants grown in the goat manure derived hydroponic nutrient solution and the tap water treatment.

### 4.3.4 Nutrient Use Efficiency

According to Table 19, there was no significant difference in terms of Nutrient Use Efficiency by tomato plants when grown in each of the three different treatments.

Furthermore, as indicated in Table 9, there was a significant ($P \leq 0.05$) difference between goat manure derived hydroponic nutrient solution and tap water in yield obtained per unit of nitrate up-taken. There was, however, no significant difference between the goat manure derived hydroponic nutrient solution and the commercial hydroponic nutrient solution in yield obtained per unit of nitrate up-taken.

**Table 19: Comparison of the three nutrient solutions for growing tomato in hydroponic conditions**

| Treatment | Mean plant biomass (g) | Nutrient use efficiency (%) | Yield (g) per nitrate up-taken |
|---|---|---|---|
| Goat manure derived hydroponic nutrient solution | 139 | 88.9 | 9.6 |
| Tap water | 15 | 82 | 0.1 |
| Commercial hydroponic nutrient solution | 139 | 94.7 | 11.9 |

## 4.4 DISCUSSION

### 4.4.1 Vegetative rresponse of tomato plants to goat manure-derived hydroponic nutrient solution

The current study's results reveal that Goat Manure Derived Hydroponic Nutrient Solution (GMDHNS) increased vegetative performance of tomato plants in comparison to using Tap Water (TW) and that there was similar vegetative performance of tomato plants grown in Commercial Hydroponic Nutrient Solution (CHNS). These results may mean that the formulated GMDHNS meets the minimum required levels of nutrients for plant vegetative growth.

This makes sense because plant vegetative growth has been known to be influenced by levels of mineral nutrients available to the plant. Using other organic sources processed

in like manner to the current study's GMDHNS, similar results have been reported by Shinohara *et al* (2011) and Kawamura-Aoyama *et al* (2014) who reported that when mineral nutrients in the organic hydroponic nutrient solutions were comparable to those of the CHNS, vegetative growth was increased. Vegetative growth is important to the plant in that it is correlated to yield. As an example, plant stem diameter was established to be correlated with yield (Glendinning, 1966). These results further suggest that the minimum amounts of other nutrients required for tomato plant growth were sufficient in the organic nutrient solution to result in similar growth patterns as those from the commercial hydroponic nutrient solution. Considering that goat manure had never been used hydroponically from available published literature, these results bring a new contribution to existing knowledge

### 4.4.2 Reproductive rresponse of tomato plants to goat manure-derived hydroponic nutrient solution

Results from the current study show that reproductive growth was high in tomato plants grown in Goat Manure-Derived Hydroponic Nutrient Solution (GMDHNS) compared to the Tap Water (TW) treatment. This means that tomatoes grown in GMDHNS received favourable amounts of nutrients in comparison to those grown in TW. This makes sense because plants grown in TW had less mineral nutrients available compared to those grown in the GMDHNS where mineral nutrients were comparable to those in the Commercial Hydroponic Nutrient Solution (CHNS). Tomato plants can therefore, grow in TW hydroponically to reach maturity, though the size of fruits will be very small and less in numbers. This means the tomato plants in TW were just surviving on the least minimum

nutrients to just keep the plants alive. Similarly, tomato reproductive performance was more when using CHNS than GMDHNS because mineral nutrients were balanced and the pH and conductance of the CHNS were buffered. During experimentation, the pH and conductance kept fluctuating from time to time and even though they were adjusted each time they changed, such changes may have had influence on the reproductive performance. The yield from manure nutrient solution is still more than most yields from organic fertilization applied on soil systems such as those found by Ghorbani *et al* (2008). Even though reproductive performance was high in the CHNS than GMDHNS, the results are comparable and when we consider gross margins economics of less than 1 million (Figure 1) it is evident that GMDHNS was formulated with less cost compared to CHNS that requires complex and costly resources to be produced. Therefore, on such merits GMDHNS benefits the investor more if it goes through the artefactual and local farmers thereafter upscale GMDHNS due to cost and accessibility. These findings from the present study are also supportive of earlier findings indicating that manures and other organic sources of plant required nutrients, indeed provide sufficient nutrients to promote plant growth and yield (Atiyeh *et al.,* 2002; Ojeniyi *et al.,* 2008; Mehdizadeh *et al.,* 2013; Wilkinson, 1979).

### 4.4.3 Nutrient Use Efficiency and plant biomass

The current results indicate that there was no significant difference in Nutrient Use Efficiency (NUE) by tomato plants between the three (3) different treatments. Such results may mean that tomato plants were able to absorb the available nutrients including nitrates as much as they are availed in the media, hence from the least available nitrates

in the tap water treatment to the most available nitrates in the commercial hydroponic nutrient solution and the goat manure derived hydroponic nutrient solution, tomato plants were able to acquire nutrients from the growth mediums. These results are supported by Zaller (2007) who indicated that the higher the nutrient concentration or availability, the higher the use of such nutrients by plants as per the plant demand. Hence it is noted from the current results that when higher concentrations of nitrates were availed, plant biomass increased, whereas when less nitrates were availed, plant biomass was also reduced, because it was determined by how much nutrients, in particular, nitrates were available for use by plants.

With regards Nutrient Use Efficiency in terms of agronomic efficiency, the current results show that there was no significant difference in yield obtained per unit of nitrate up-taken between the GMDHNS and the CHNS treatment, whereas there was a significant difference between the TW treatment and the other 2 treatments (GMDHNS & CHNS). This could be attributed to the fact that nutrients availability, in particular nitrates, was sufficient in the GMDHNS and in the CHNS treatments compared to the undesirable levels in the TW treatment. Nitrates were optimized in the GMDHNS and in the CHNS treatments to ranges that are desirable for tomato plants. The desirable range of nitrate for tomato plants is 200 mg/L (Jones, 2007) and the manure and positive control treatments were optimized to such levels and were efficiently used by tomato plants.

These results are supported by Aerts (1990) who found that when mineral nutrients in the growth media are efficiently used by plants, vegetative and reproductive plant performances are enhanced. Hence, we see the reproductive performance in this regard was enhanced because nutrients in the organic nutrient solution were efficiently usable

making the GMDHNS an optimized nutrient solution with potential to maximize yields. In hydroponic systems such as the one applied in the current study, using organic fertilization has now an advantage of increasing plant nutrient use considering the fact that nutrients are availed to plants for prolonged periods thereby allowing plants enough time to use the available nutrients. Moreover, enhancing crop yield is every farmer's goal, therefore, with the formulated organic nutrient solution from goat manure, crop yield for every nitrate in the nutrient solution provides prospects for a new tomato value chain.

The current results indicate that tomato biomass weight at the last harvest after seeding, were greatest in the Goat Manure Derived Hydroponic Nutrient Solution (GMDHNS) and in the Commercial Hydroponic Nutrient Solution (CHNS), and least in the Tap Water (TW) treatment. This can be attributed to nitrate concentration and availability in the GMDHNS and in the CHNS treatments where nitrate levels were higher than in the TW treatment. The present study's results are supported by Baligar & Duncan (1990) who indicated that nutrient availability in the growth media influence the total dry matter accumulation which in turn influences nutrient demand. Therefore, the nutrient solution formulated from goat manure contain sufficient nitrates that are efficiently usable by tomato plants from the available pool of nutrients. The nutrient solution formulated from goat manure with regards plant biomass, is as good as using commercial fertilizer as it stimulated comparable total plant biomass in tomato plants. Other studies such as that by Zaller (2007) found similar results as for the current study, where organic fertilization produced higher tomato plant biomass with higher concentrations whereas lower organic fertilizer concentration produced less tomato plant biomass. Therefore, the TW treatment with less nitrate concentration subsequently produced less tomato plant biomass, whereas,

the GMDHNS and CHNS treatments with comparable higher concentrations produced higher tomato plant biomass. Therefore, even though tomato plants grown in TW treatment reached maturity, they remained short and thin plants with very small sized fruits that would be undesirable to a farmer in comparison to the other treatments. This is also attributed to less nutrients in the growth media available for plant use.

## 4.5 CONCLUSION

The organic nutrient solution formulated from goat manure positively improved plant growth and yield performance of a tomato crop, and provides a technique feasible and alternative to conventional hydroponics. The locally formulated hydroponic nutrient solution (GMDHNS) compared well with the conventional nutrient solution (CHNS) and even though GMDHNS may require further optimization, it is a good and cheaper alternative in that locals would easily adopt it because it relies on adding value to their own resources. This is particularly significant to communities living in Namibian areas where fertilizer access, water or fertile soil are limiting factors to horticultural production in that locally available manure resources can be added value to and used as the main input for horticultural production. These results open further possibilities for other crops currently grown in hydroponics using conventional fertilisers and being baseline for Africa in terms of organic hydroponics, provide a foundation for further research in this regard.

## REFERENCES


1. Aerts, R. (1990). Nutrient use efficiency in evergreen and deciduous species from heathlands. *Oecologia, 84* (3) 391-397.
2. Atiyeh, R.M., Arancon, N., Edwards, C.A., and Metzger, J.D. (2000). Influence of earthworm-processed pig manure on the growth and yield of greenhouse tomatoes. *Bioresource Technology*, *75*(3), 175-180.
3. Atiyeh, R.M., Arancon, N.Q., Edwards, C.A., Metzger, J.D. (2002). The influence of earthworm-processed pig manure on the growth and productivity of marigolds. *Bioresource technology*, *81*(2), 103-108.
4. Baligar, V. C., R. R. Duncan, and N. K. Fageria. 1990. Soil-plant interaction on nutrient use efficiency in plants: An overview. pp 351–373. In: V C Baligar and R R Duncan (eds.), Crops as Enhancers of Nutrient Use. Academic Press, San Diego, CA.
5. Baligar, V.C., Fageria, N.K., and He, Z.L. (2001). Nutrient use efficiency in plants. *Communications in Soil Science and Plant Analysis*, *32*(7-8), 921-950.
6. Ghorbani, R., Koocheki, A., Jahan, M., and Asadi, G.A. (2008). Impact of organic amendments and compost extracts on tomato production and storability in agroecological systems. *Agronomy for sustainable development*, *28*(2), 307-311.
7. Glendinning, D.R. (1966). Further observations on the relationship between growth and yield in cocoa varieties. *Euphytica*, *15*(1), 116-127.

8. Grewal, H.S., Maheshwari, B., and Parks, S.E. (2011). Water and nutrient use efficiency of a low-cost hydroponic greenhouse for a cucumber crop: An Australian case study. *Agricultural Water Management*, *98*(5), 841-846.
9. Jones, J.B. (2007). Tomato Plant Culture-In the Field, Greenhouse, and Home Garden. CRC Press, New York.
10. Kawamura-Aoyama, C., Fujiwara, K., Shinohara, M., Takano, M. (2014). Study on the hydroponic culture of lettuce with microbially degraded solid food waste as a nitrate source. *Japan Agricultural Research Quarterly: JARQ*, *48*(1), 71-76.
11. Liu, T., Cheng, Z., Meng, H., Ahmad, I., Zhao, H. (2014). Growth, yield and quality of spring tomato and physicochemical properties of medium in a tomato/garlic intercropping system under plastic tunnel organic medium cultivation. *Scientia Horticulturae*, *170*, 159-168.
12. Mehdizadeh, M., Darbandi, E.I., Naseri-Rad, H., Tobeh, A. (2013). Growth and yield of tomato (Lycopersicon esculentum Mill.) as influenced by different organic fertilizers. *International journal of Agronomy and plant production*, *4*(4), 734-738.
13. Ojeniyi, S.O. (2008). Effect of poultry manure on selected soil physical and chemical properties, growth, yield and nutrient status of tomato. *African Journal of Agricultural Research*, *3*(9), 612-616.
14. Shah, S.H., Munir, S.U., and Shah, S.H. (2011). Evaluation of two nutrient solutions for growing tomatoes in a non-circulating hydroponics system. *Sarhad Journal of Agriculture (Pakistan)*.

15. Shinohara, M., Aoyama, C., Fujiwara, K., Watanabe, A., Ohmori, H., Uehara, Y., Takano, M. (2011). Microbial mineralization of organic nitrogen into nitrate to allow the use of organic fertilizer in hydroponics. *Soil science and plant nutrition*, *57*(2), 190-203.
16. Wilkinson, S.R. (1979). Plant Nutrient and Economic Valus of Animal Manures. *Journal of animal Science*, *48*(1), 121-133.
17. Zaller, J.G. (2007). Vermicompost as a substitute for peat in potting media: Effects on germination, biomass allocation, yields and fruit quality of three tomato varieties. *Scientia Horticulturae*, *112*(2), 191-199.
18. Zotarelli, L., Dukes, M.D., Scholberg, J.M.S., Munoz-Carpena, R., and Icerman, J. (2009). Tomato nitrogen accumulation and fertilizer use efficiency on a sandy soil, as affected by nitrogen rate and irrigation scheduling. *Agricultural Water Management*, *96*(8), 1247-1258.

# CHAPTER 5: FRUIT QUALITY DUE TO THE FORMULATED NUTRIENT SOLUTION


## ABSTRACT

The influence of goat manure-derived hydroponic nutrient solution on tomato fruit quality was studied using a randomized complete block design (RCBD) with three treatments (goat manure-derived hydroponic nutrient solution (GMDHNS), commercial hydroponic nutrient solution (CHNS) and tap water (TW)) replicated three times. Tomato fruits from the three treatments were collected and analyzed for Total Soluble Solids (TSS), weight and size, and lycopene content. Though GMDHNS results were comparable, they were both significantly ($P \leq 0.05$) more than TP. Results found on fruit weight and size were that CHNS significantly ($P \leq 0.05$) increased fruit weight by 48%, whereas GMDHNS significantly ($P \leq 0.05$) increased fruit weight by 42% more than TW. CHNS had significantly ($P \leq 0.05$) larger tomatoes more than the other treatments, whereas GMDHNS had significantly ($P \leq 0.05$) larger tomatoes than TW. Lycopene content results indicated that GMDHNS had significantly ($P \leq 0.05$) higher lycopene content than CHNS. Overall the current study's results indicate that GMDHNS produces higher quality fruits even more than CHNS.



## KEYWORDS

Fruit quality, size, weight, Brix values, Total Soluble Solids, lycopene.


## 5.1 INTRODUCTION

The first part of this chapter gives the problem statements, objectives and hypothesis of the chapter. The second part of this chapter then details the procedure or materials and methods used to collect and analyse data for the objective of this chapter. The last part of the chapter presents the results found from the collection and analysis of data for this chapter as well as the discussion and conclusion of the chapter.

### 5.1.1 Problem statement

With the advent of healthy lifestyles leading to diet transformations in recent years, consumers now determine acceptable standards of quality for foods they get. With many now preferring organically grown vegetables over conventionally grown ones on account of health benefits that are associated with quality attributes from organically grown vegetables. For semi-arid countries such as Namibia where hydroponic growing system has been introduced, the commercial nutrient solution has always been the only way to practice hydroponics in Namibia, until recently when Mowa (2015) established that it was possible to grow vegetables hydroponically using organic sources in Namibia. Such establishments have since left a lot of aspects such as quality concerning vegetables produced from organic hydroponics. The current study therefore, looked at the quality of tomatoes produced from organic hydroponics.

### 5.1.2 Objective

To determine the quality of tomatoes produced from organic hydroponic nutrients.

### 5.1.3 Research hypothesis

The organic nutrient solution formulated from goat manure will improve tomato fruit quality (weight, size, Brix value and lycopene).

## 5.2 MATERIALS AND METHODS

For this study, fruit quality was product oriented as opposed to customer oriented, whereby quality was evaluated through fruit size and weight, total soluble solids content, and lycopene content. Treatment 1 was tomato fruits from plants grown in the organic nutrient solution formulated from goat manure. Treatment 2 (positive control) was tomato fruits from plants grown in the conventional hydroponic nutrient solution. Treatment 3 (negative control) was tomato plants grown in plain tap water, but did not produce fruits sufficient for analysis and comparison to other treatments. Hydroponic tables (22.8 cm X 250 cm X 150 cm) constructed from planks and black sheeting were used in a greenhouse at the Sam Nujoma Marine and Coastal Research Centre (SANUMARC), Henties Bay. Styrofoam that completely cover the hydroponic tables yet float on water in the table were added. Each hydroponic table covered an area of 3.75 $m^2$. A randomized complete block design (RCBD) with three treatments each triplicated (organic nutrient solution, conventional hydroponic fertiliser (as positive control) and plain tap water (as negative control). Each replicate was a hydroponic table of 6 plants and served as an experimental unit.

### 5.2.1 Fruit weight and size data collection and analysis

Fruit size and weight were determined according to Mena-Violante & Olalde-Portugal (2007). Fruits were harvested manually from 10 randomly selected plants for each treatment, then washed in tap water and carefully selected to ensure good uniformity in maturity and size. Nine (9) tomato fruits from each treatment were then separated into three sizes according to Atiyeh *et al* (1973) whereby tomato sizes were classified based

on the following diameters: small (<5:8 cm), medium (between 5.8 and 6.4 cm), and large (>6:4 cm). Percentages of tomatoes in each size category were then determined. Eighteen (18) tomato fruits were then weighed on a semi-analytical scale (Mettler Toledo) as whole fruits individually and results expressed in grams (g). Size measurements were made with a handheld Vernier callipers and expressed in centimetres (cm).

Means comparisons and correlations analysis were done whereby, a One-way Analysis of Variance (ANOVA) at the 0.05 confidence levels was used.

$$Yij = \mu+\tau i + \beta j + \gamma ij + \epsilon ij$$

Where $\mu$ is the overall mean response, $\tau i$ is the effect due to the $i$ -th level of factor A, $\beta j$ is the effect due to the$j$ -th level of factor B and $\gamma ij$ is the effect due to any interaction between the $i$ -th level of A and the $j$ -th level of B, $\epsilon ij$ is the error term where the error terms are independent observations.

### 5.2.2 Brix value data collection and analysis

A Brix test was applied to determine Total Soluble Solids (TSS) content as affected by the organic nutrient solution. Therefore, Brix values were determined according to Helyes *et.al* (2006), where the value was measured using a refractometer (Multi Fruits PAL-BX|ACID F5). Ripened sample fruits from six randomly selected plants per treatment were collected randomly by hand with 1 kg per treatment. Fruit samples from each treatment were washed and homogenised using a blender and the serum samples were then analysed. The tap water treatment did not produce sufficient quantities of tomato to warrant Brix analysis.

Means comparisons using the ANOVA test was used on data for Brix values to determine whether the effects of the treatments were significant ($P \leq 0.05$ for F-test).

### 5.2.3 Lycopene data collection and analysis

#### 5.2.3.1 Tomato fruit harvest

Ten (10) tomato fruits were randomly hand harvested from the two treatments (goat manure derived hydroponic nutrient solution and commercial hydroponic nutrient solution) at the last harvest when they had reached the mature red-ripe stage. Harvested tomatoes were washed and cut into halves and homogenized into a paste in a blender for1min before being used for lycopene content analysis. Tap Water did not produce sufficient fruits to warrant HPLC analysis.

#### 5.2.3.2 Samples

Approximately 50 g of the homogenous paste from the blender for each treatment was prepared for processing for lycopene extraction.

#### 5.2.3.3 Reagents and standards

All solvents: methanol, Dimethyl sulfoxide, hexane, acetone and ethanol) used for separation of tomato lycopene were of HPLC grade and purchased from Biodynamics Namibia. The lycopene standard was purchased from Sigma-Aldrich Chemicals (St Louis, MO, USA).

#### 5.2.3.4 HPLC Analysis of Lycopene Instrumentation

The HPLC apparatus (Perkin Elmer Flexar LC) was equipped with a quaternary Isopump Exchange System (G1310A), Degasser (G1379A), ALS autosampler (G1329A), ALS Therm thermostat (G1330B) column heater and a Thermo Separation Spectra Series UV/Visible diode-array detector (G1315A). The column used for separation was Column Develosil 5µ C30-UG 100A Phenomenex (150 x 4.6 mm). Chemicals/Reagents and Standards of lycopene from tomato were obtained from Sigma Company (St. Louis, MO, USA), HPLC grade solvents were supplied by Biodynamics Namibia. De-mineralized water was prepared regularly and supplied to the laboratory through pipeline.

#### 5.2.3.5 Preparation of Standard Solutions Lycopene

The trans-lycopene (all-E'-lycopene) standard solution was prepared by diluting 1 mg commercial lycopene standard from tomato with hexane to desired concentration and the solutions were transferred to a volumetric flask. Sample Preparation Lycopene extraction from tomato was based on modifications and optimization of Barba *et al* (2006) method, with solvents mixtures of hexane/acetone/ethanol (50:25:25, v/v/v) for extraction. Approximately 50 g of homogeneous tomato paste was precisely weighed into a 250 mL Erlenmeyer flask, tightly wrapped in aluminium foil to protect it from light and then mixed with 150 mL of extraction solvent. The mixture was stirred on a magnetic stirring plate during 30 min. 15mL of de-mineralized water was added to the extract, and mechanically agitated. The solution was subsequently separated into distinct phases of polar and non-polar layers. From lycopene bearing upper hexane layer, 10mL was placed in a round-

bottomed flask and dried in rotary evaporator (Heidolph 36000310) at ~34°C. Different compositions of solvents (THF/ACN/MeOH) were assayed for redissolution of the dry extract: (1) THF/ACN/MeOH (15:30:55, v/v/v), (2) DMSO/MeOH (50:50/v/v) and, (3) dissolving in THF and then ACN/MeOH followed (15:30:55, v/v/v). In all the solvents containing Methanol and Acetonitrile, ultrasonic agitation for few minutes was used to make sure that the solvents were mixed homogeneously. The lycopene residue was dissolved to final volume of 3mL with DMSO/MeOH (50:50/v/v for HPLC analysis.

#### 5.2.3.6 Chromatographic Conditions

Tomato extracts were analysed for lycopene content by separation followed by quantitative determination using reversed-phase HPLC system consisting of UV/visible diode-array detector separation module, auto-injector, and column temperature regulator. The C30 column was then conditioned with the elution solvent at flow rate of 0.8 mL/min with detection at 472 nm. Other flow rates including 0.9 and 1.5mL/min have also been tested. Based on Barba *et al*. (2006), separation of carotenoids was attempted isocratically using various compositions of ISP (Isopropyl) and ACN (acetonitrile) as mobile phase: (1) ISP /ACN (40:60, v/v). Injection volume of 5 µL was used to avoid overloading on column during analysis. The peak responses were measured at 472 nm for lycopene (Figure 17). The identification of the peaks was carried out by comparing the retention times with those obtained with a standard solution of all-trans lycopene (Figure 1). The quantification was performed using calibration curves made with different injected amounts of external standard, all-trans-lycopene; in a similar proportion as in the samples. Peak Identification and Quantification Identification of carotenoids was carried out by

comparison of HPLC retention times with corresponding standards and chromatographic properties with standards of lycopene.

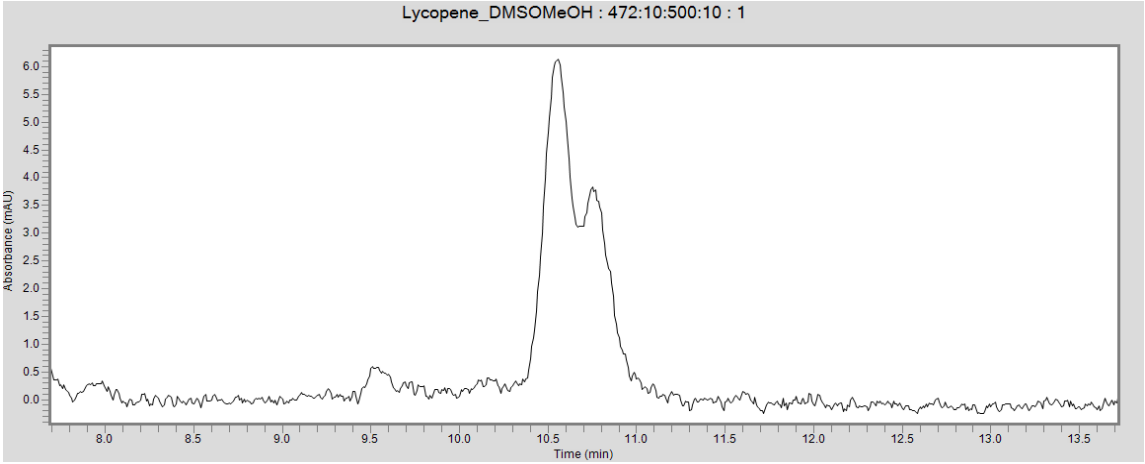



**Figure 17: Chromatogram of lycopene standard solution under chromatographic condition: Column Develosil 5µ C30-UG 100A Phenomenex (150 x 4.6 mm); mobile phase ISP/ACN (40:60, v/v); flow rate 0.8 ml/min; column temperature 30 ˚C; λdetection = 472 nm.**

In general, quantitative analysis of carotenoids is carried out by external calibration curve method, generated from measurements made with many concentrations of pure standards (0.039-2.8 µg/mL). Then lycopene content in tomato was calculated according to the formula here below.

$$Lycopene\ [mg/g] = \frac{A_{Sa}\, V_T\, V_2}{m.\, RF.\, V_1}$$

Where, **Asa**: Peak areas of lycopene from sample solution [mAU], **m**: Amount of sample [g], **RF**: Response factor [mAU/mg], **VT**: Total volume of the upper lycopene bearing hexane layer [mL], **V1**: Volume of extract, which is evaporated [mL] and **V2**: Volume of final sample solution [mL].

The result was then expressed as milligram of lycopene per 100g fresh weight of the tomato analysed. Validation of method for repeatability was done by repeating analysis on the same samples for three days.

### 5.3.4 Statistical Analysis

All tomato data from HPLC-analysis were subjected to analysis of variance (ANOVA) to test treatment differences at $P \leq 0.05$ using SPSS® version 24.

## 5.3 RESULTS

### 5.3.1 Fruit weight and size

According to Figure 18 and Table 20, GMDHNS significantly ($P \leq 0.05$) increased single tomato fruit weight by 42% more than the TW treatment, whereas CHNS significantly ($P \leq 0.05$) increased single tomato fruit weight by 48% more than the TW treatment where no fertiliser was added. Therefore, the usage of goat manure derived hydroponic nutrient solution yields more single fruit weight comparable to using commercial hydroponic nutrient solution.

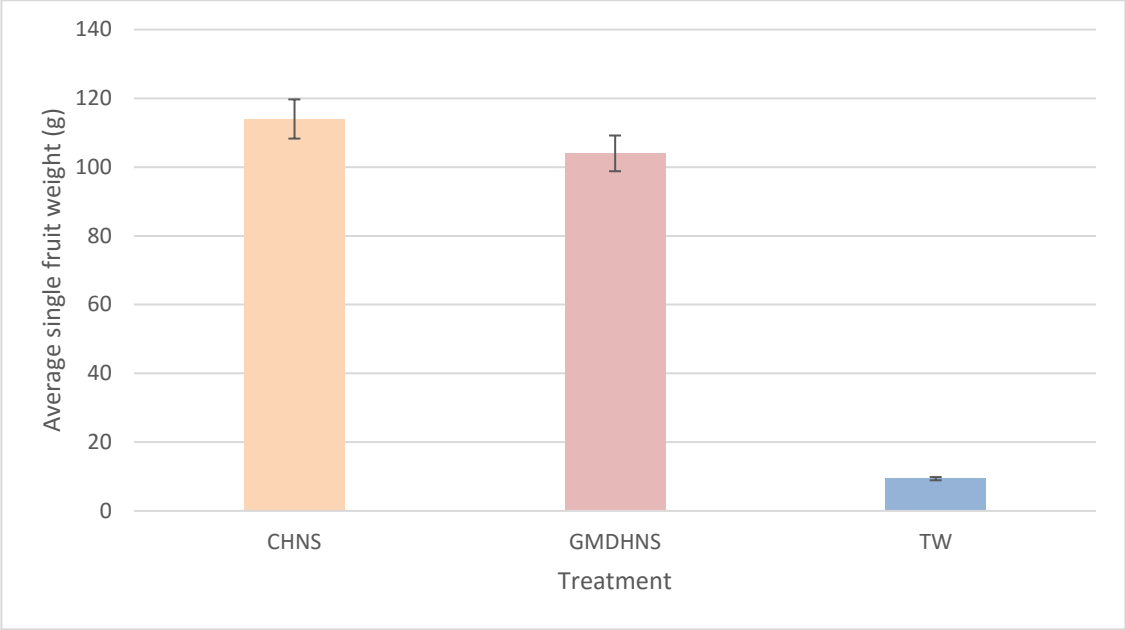



**Figure 18: Average weight of a single tomato fruit as influenced by nutrient solutions: CHNS – Commercial Hydroponic Nutrient Solution; GMDHNS – Goat Manure Derived Hydroponic Nutrient Solution; TW – Tap Water**

**Table 20: Descriptive statistics for single fruit weight between the 3 treatments**

| Treatment | N | Mean | Std. Deviation | CV | Std. Error |
|---|---|---|---|---|---|
| CHNS | 18 | 112.256 | 6.6303 | 5.8 | 1.5628 |
| GDHNS | 18 | 106.678 | 5.9704 | 5.6 | 1.4072 |
| TW | 18 | 9.389 | 11.5867 | 122 | 2.7310 |

Growing tomato plants in the GMDHNS resulted in more medium sized fruits (diameter 5.8 - 6.4) than small sized fruits (diameter <5.8) and also more than large sized tomato fruits (diameter >6.4). In contrast, growing tomato plants in the CHNS resulted in more large fruits than medium sized and small sized fruits (Figure 19 and Table 21). Growing tomato plants in the TW treatment resulted in all small sized fruits. Analysis of variance shows that these results were significantly different ($P \leq 0.05$) at confidence levels of 0.5.

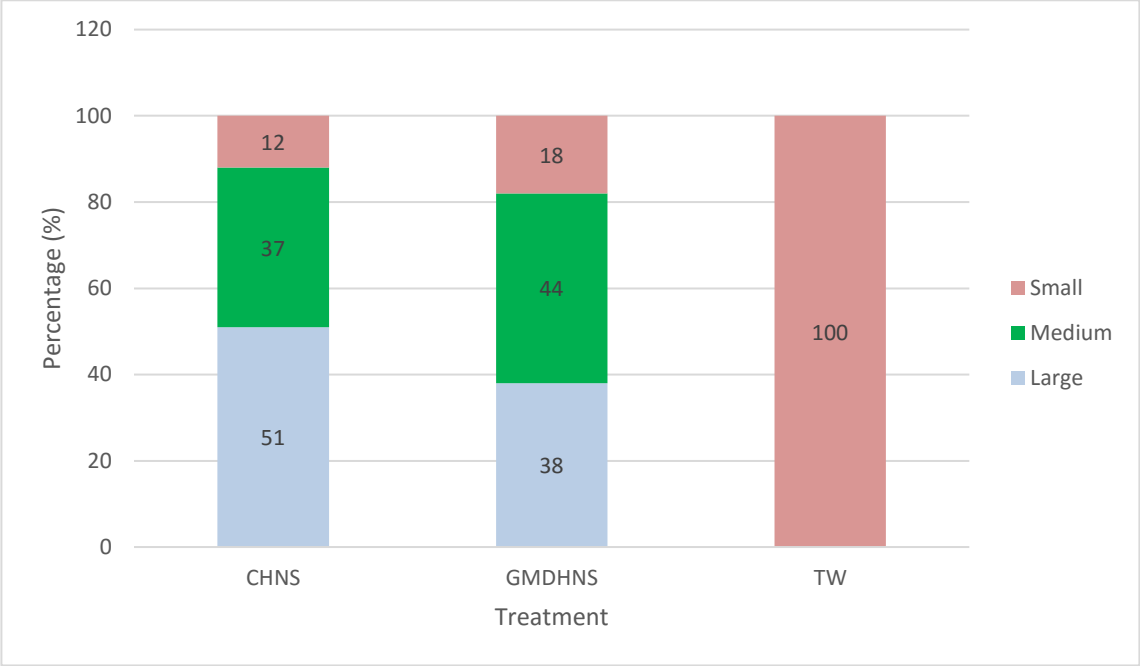



**Figure 19 Sizes of tomato fruits produced from a standard Commercial Hydroponic Nutrient Solutiom (CHNS) compared with those produced from Goat Manure Derived Hydroponic Nutrient Solution (GMDHNS.)**

**Table 21: Descriptive statistics for tomato fruit sizes between the 3 treatments**

| Treatment | N | Mean | Std. Deviation | CV | Std. Error |
|---|---|---|---|---|---|
| CHNS | 9 | 6.077778 | .5403188 | 8.8 | .1801063 |
| GMDHNS | 9 | 5.144444 | .8001736 | 15.6 | .2667245 |
| TW | 9 | 1.588889 | .3407508 | 22.6 | .1135836 |

### 5.3.2 Brix values

Total soluble solids (Brix) content of tomato fruits from plants grown in the goat manure derived hydroponic nutrient solution were found to be significantly higher ($P \leq 0.05$) than those from fruits grown in the commercial hydroponic nutrient solution as can be seen in Tables 22 and 23 below.

**Table 22: Brix values from tomatoes in the 2 nutrient solutions**

| Treatment | Brix (°) |
|---|---|
| Goat manure derived hydroponic nutrient solution | 7.4 |
| Commercial hydroponic nutrient solution | 5.6 |

**Table 23: Descriptive statistics for Brix values of tomatoes in the 2 nutrient solutions**

| | N | Mean | Std. Deviation | CV | Std. Error |
|---|---|---|---|---|---|
| CHNS | 15 | 5.653333 | .8741254 | 15.4 | .2256982 |
| GMDHNS | 15 | 6.446667 | .6812454 | 10.5 | .1758968 |

## 5.3.3 Lycopene content

### *5.3.3.1 Linearity and calibration curve*

Tomato lycopene standards were dissolved at different concentrations in the same conditions as mentioned in the method, after which they were injected in the chromatographic system. The resulting calibration curve is depicted in figure 20.

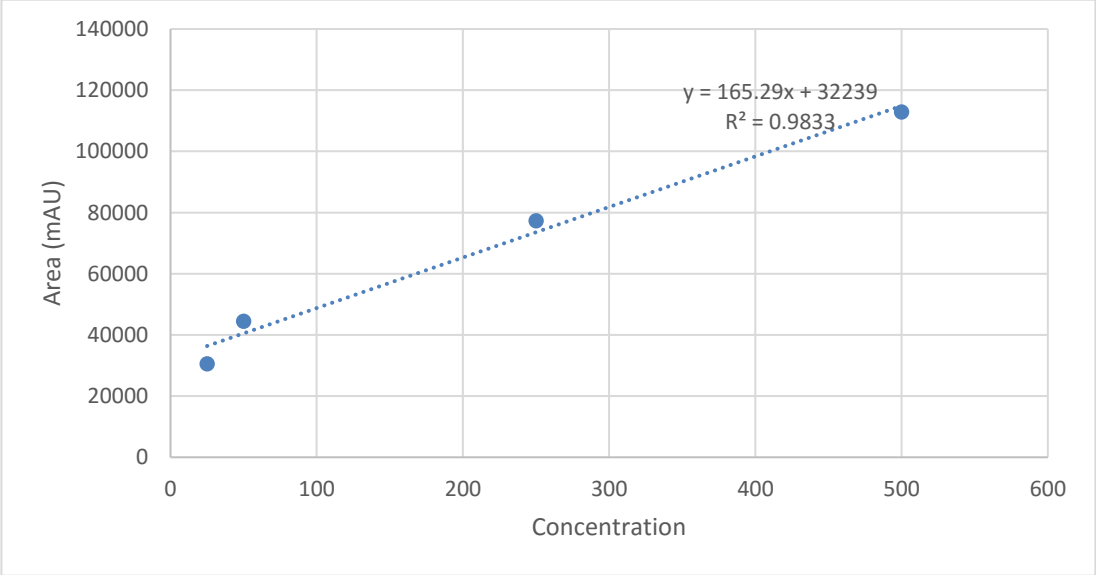



**Figure 20: Four-pointed (n=2) calibration curve for lycopene standard showing area vs concentration**

Tomato lycopene standards were dissolved at different concentrations in the same conditions as mentioned in the methodology section, after which they were injected in the chromatographic system. The resulting calibration curve is depicted in figure 20.

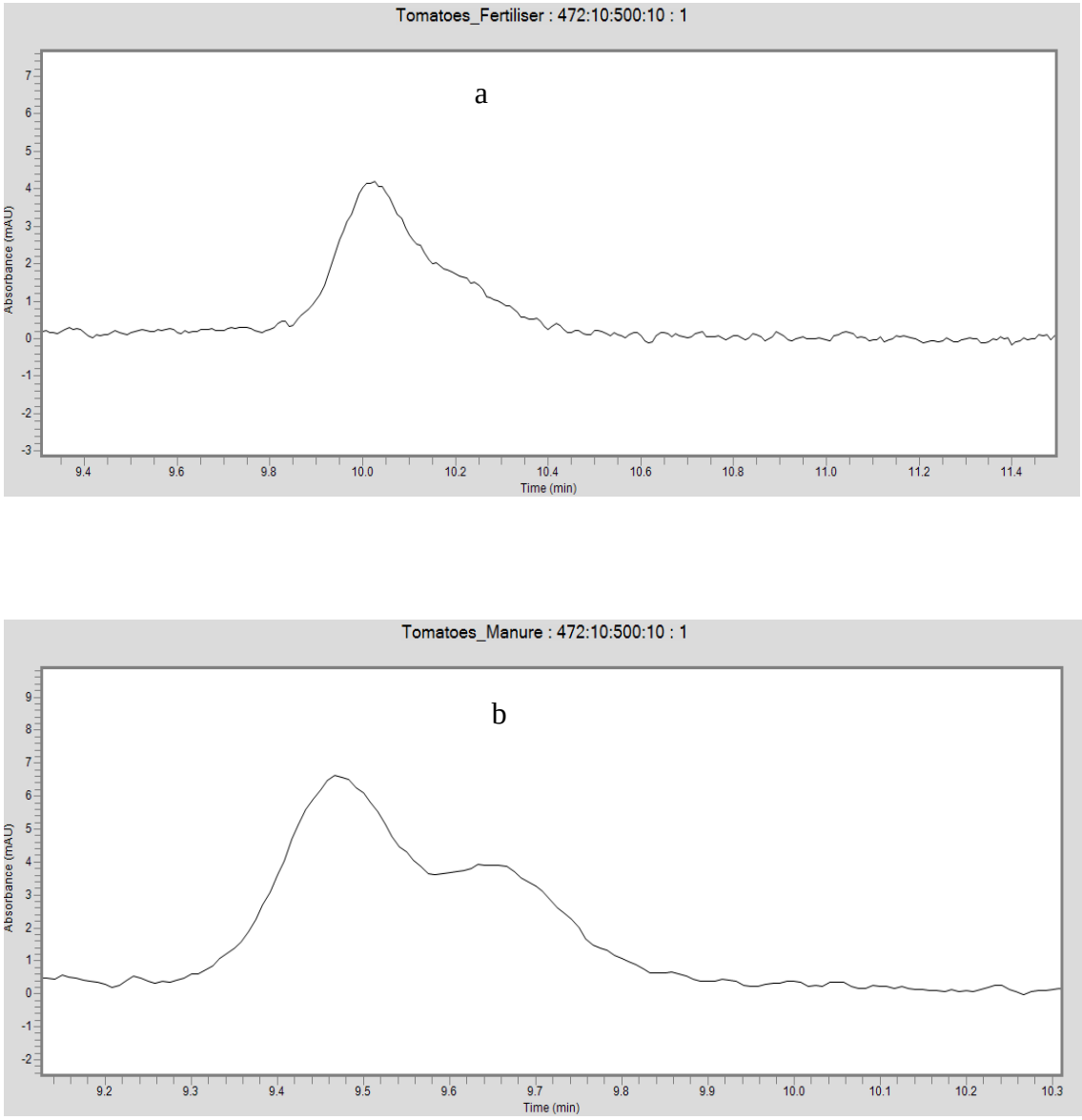



**Figure 21: Chromatogram of lycopene in tomatoes from conventional (CHNS) (a) and manure nutrient (GMDHNS) (b) solutions under chromatographic condition: Column Develosil 5µ C30-UG 100A Phenomenex (150 x 4.6 mm); mobile phase ISP/ACN (40:60, v/v); flow rate 0.8 ml/min; column temperature 30 ˚C; λdetection = 472 nm.**

HPLC results show that there was no other carotenoid displayed from the current results other than lycopene shown in Figure 21(a) and 21(b). The highest lycopene concentration obtained was in tomato fruits from the Goat Manure Derived Hydroponic Nutrient Solution, whereas the lowest lycopene concentration was in tomato fruits from the conventional treatment (Figure 22 & Table 24).

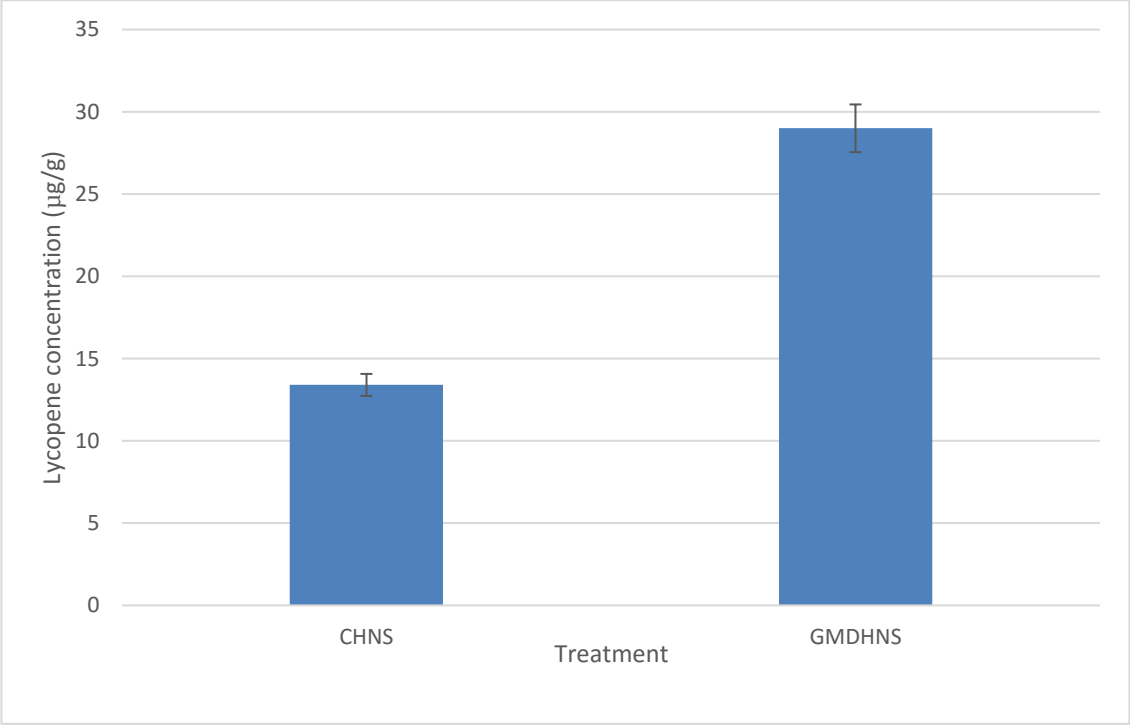



**Figure 2: Lycopene concentration levels in tomatoes in the 2 treatments (CHNS – Commercial Hydroponic Nutrient Solutiom; GMDHNS – Goat Manure Derived Hydroponic Nutrient Solution)**

**Table 24: Descriptive statistics for lycopene content of tomatoes in the 2 nutrient solutions**

| Treatment | N | Mean | Std. Deviation | CV | Std. Error |
|---|---|---|---|---|---|
| GMDHNS | 3 | 28.600 | 1.4422 | 4.8 | .8327 |
| CHNS | 3 | 12.800 | .7211 | 5.6 | .4163 |
| Total | 6 | 20.700 | 8.7139 | | 3.5574 |

## 5.4 DISCUSSION

### 5.4.1 Weight and size

The Goat Manure Derived Hydroponic Nutrient Solution (GMDHNS) increased single tomato fruit weight more than the Tap Water (TW) treatment and was comparable to the Commercial Hydroponic Nutrient Solution (CHNS). This means that the locally formulated GMDHNS meets the minimum mineral nutrient requirements for quality tomato fruit weights. These results are further attributed to the fact that fruits from the GMDHNS were bigger and fleshier than those from the TW treatment, whereas fruits from the CHNS were generally bigger and fleshier with more weight in them compared to fruits from other treatments. This weight and size increment was therefore, influenced by the availability of mineral nutrients in the different nutrient solution which is limited by stressing conditions. Pieper & Barrett (2009) support these findings by arguing that tomato plants with less stressing conditions will result in tomato fruits with higher moisture content which in turn determine the size and weight of tomato fruits. This explains why tomato plants in tap water yielded only smaller sized fruits because tap water alone has less mineral nutrients hence such plants were stressed on account of less available nutrients. Stressing conditions can also explain why there were larger sized fruits in the commercial hydroponic nutrient solution than in the goat manure derived hydroponic nutrient solution. From the current study, the pH and EC in the organic nutrient solution were fluctuating despite the adjustments made during the growing period, which may have led to inconsistent levels of nutrients available to the tomato plants at all times. Buck *et al* (2004) recommend the pH in the hydroponic nutrient solution which is optimal to be 6.5, whereas the pH in the GMDHNS kept fluctuating because it was not

buffered in contrast to the buffered conventional nutrient solution (CHNS). There was also a further difference in Electrical Conductivity (EC) between the organic nutrient solution (GMDHNS) and the conventional nutrient solution (CHNS) which may have brought further stress to plants grown organically. Dorai *et al* (2001) established that when EC in the nutrient solution was high, tomato fruits tended to be smaller in size compared to a balanced recommended EC of <3,5 $mS \times cm^{-1}$. Therefore, the absence of these potential stressors in the conventional nutrient solution may have led to development of more fleshy tomato fruits in the conventional nutrient solution compared to the organic nutrient solution. Zushi *et al* (2009) also reported that stressing conditions in organic growing systems on the tomato plant lead to lower mass in the fruit in contrast to conventional fertilisers where such stressors are absent.

### 5.4.2 Brix values

The results from the current study reveal that Brix values were higher in tomato fruits from the organic nutrient solution than from tomato fruits from the conventional nutrient solution. This could be attributed to stress and EC levels that were in the organic nutrient solution in contrast to the balanced and buffered conventional nutrient solution. Helyes *et al* (2006) support these findings as they reported that Brix values in tomatoes were influenced by the cultivation method used, which in the current study were the organic nutrient solution and the conventional nutrient solution. Oliveira *et al* (2013) also support these findings as they reported that total soluble solids (Brix) are increased in fruits from organic farming systems over conventional farming systems.

Hence the organic nutrient solution from the current study resulted in high Brix values because it is a system with no buffered EC values which may introduce stressing conditions to the tomato plants. Kleinhenz & Bumgarner (2012) also support these findings as they reported that stressing tomato plants during fruit development can increase fruit soluble solids content but lower total fruit yield. They further reported that increased electrical conductivity levels in irrigation water lead to increased °Brix levels in greenhouse-grown tomatoes.

### 5.4.3 Lycopene content

The current study found that lycopene content was high in organically grown tomatoes compared to conventionally grown tomatoes. This means that locally formulated GMDHNS has the attributes to produce quality tomato fruits with health benefits such as reducing various cancer risks. These results are supported by Brandt & Mølgaard (2001) who reported that lycopene was high in organically grown fruits compared to conventionally grown fruits. These differences in lycopene content from the current study could be attributed to firstly, the dilution effect. Tomato fruits with more water content will have less lycopene per unit (grams) compared to tomatoes with less water content on the same units. Hence the lycopene would have been diluted more in the presence of more water in the fruit. Pieper & Barrett (2009) reported similar results and they maintained that the higher moisture content in conventional tomato fruits was responsible for lower lycopene and soluble solids contents due to dilution. This is because conventional tomato fruits contain more water which dilutes the concentration per unit. This may be attributed to the current study's results as well, considering that there were more large-sized fruits

from conventional nutrient solution than from the organic nutrient solution where there were more medium sized fruits. Yet higher lycopene content was recorded in the organic nutrient solution.

Secondly, the higher lycopene content in fruits from organic nutrient solution (GMDHNS) can be attributed to time to fruit maturity. It was established in the current study (chapter 4) that fruits from tomato plants grown in the organic nutrient solution took longer to reach maturity. This aspect has been reported by Benbrook (2005) who indicated that the superior quality in organic foods was linked to the pace of crop physiological development which is relatively slower in organic production systems than that from conventional production systems.

Thirdly, the higher lycopene content in fruits from organic nutrient solution can be attributed to defence to stress. pH and EC values of the organic nutrient solution kept fluctuating throughout the study in contrast to the buffered conventional nutrient solution. Such inconsistencies may have introduced stress to tomato plants, which can react by producing more antioxidants when stressed. Carbonaro *et al* (2002) support these findings as they reported similar results, where they established that polyphenol and antioxidant levels are higher in the organic treatments compared to the conventionally grown crops, because improvement in the antioxidant defense system of the plant occurs as a consequence of the organic cultivation practice where there is more stress compared to conventional practices.

## 5.5 CONCLUSION

Based on the current results, it can be concluded that the formulated Goat Derived Hydroponic Nutrient Solution (GMDHNS) improves tomato fruit quality to high qualities over tomato fruits from the Commercial Hydroponic Nutrient Solution (CHNS). The quality introduced by the (GMDHNS) makes it worthwhile to go into artefactual and field framed before reaching the level of business uptake in order to realise a new tomato value chain that will then be up scaled by locals initially and the wider community eventually. This is because the new tomato value chain is based on quality standards of a tomato product instead of just mass quantity production.

## REFERENCES


1. Atiyeh, R.M., Arancon, N., Edwards, C.A., and Metzger, J.D. (2000). Influence of earthworm-processed pig manure on the growth and yield of greenhouse tomatoes. *Bioresource Technology*, *75*(3), 175-180.
2. Barba, A.O., Hurtado, M.C., Mata, M.S., Ruiz, V.F., and De Tejada, M.L.S. (2006). Application of a UV-vs detection-HPLC method for a rapid determination of lycopene and β carotene in vegetables. *Food chemistry*, *95*(2), 328-336.
3. Benbrook, C.M. (2005). Elevating antioxidant levels in food through organic farming and food processing. *Organic Center, State of Science Review, January*.
4. Brandt, K., and Mølgaard, J.P. (2001). Organic agriculture: does it enhance or reduce the nutritional value of plant foods? *Journal of the Science of Food and Agriculture*, *81*(9), 924-931.
5. Buck, J.S., Kubota, C., Wu, M. (2004). Effects of nutrient solution EC, plant microclimate and cultivars on fruit quality and yield of hydroponic tomatoes (Lycopersicon esculentum). In *VII International Symposium on Protected Cultivation in Mild Winter Climates: Production, Pest Management and Global Competition* 659 (pp. 541-547)
6. Carbonaro, M., Mattera, M., Nicoli, S., Bergamo, P., & Cappelloni, M. (2002). Modulation of antioxidant compounds in organic vs conventional fruit (peach, Prunus persica L., and pear, Pyrus communis L.). *Journal of agricultural and food chemistry*, *50*(19), 5458-5462.
7. Dorai, M., Papadopoulos, A., and Gosselin, A. (2001). Influence of electric conductivity management on greenhouse tomato yield and fruit quality. *Agronomie*, *21*(4), 367-383.

8. Helyes, L., Dimény, J., Pék, Z., and Lugasi, A. (2006). Effect of the variety and growing methods as well as cultivation conditions on the composition of tomato (Lycopersicon lycopersicum (L.) Karsten) fruit. In *IV International Conference on Managing Quality in Chains-The Integrated View on Fruits and Vegetables Quality 712* (pp. 511-516).
9. Kleinhenz, M.D., and Bumgarner, N.R. (2012). Using Brix as an indicator of vegetable quality. *Linking measured values to crop management. Fact Sheet. Agriculture and Natural Resources. The Ohio State University, Columbus, OH*.
10. Mena-Violante, H.G., and Olalde-Portugal, V. 2007. Alteration of tomato fruit quality by root inoculation with plant growth-promoting rhizobacteria (PGPR): Bacillus subtilis BEB-13bs. *Scientia Horticulturae*, *113*(1), 103-106.
11. Mowa, E. (2015). Organic manure for vegetable production under hydroponic conditions in arid Namibia. *International Science and Technology Journal of Namibia*, *5*:3-12.
12. Oliveira, A.B., Moura, C.F., Gomes-Filho, E., Marco, C.A., Urban, L., and Miranda, M.R.A. (2013). The impact of organic farming on quality of tomatoes is associated to increased oxidative stress during fruit development. *PLoS One*, *8*(2), e56354.
13. Pieper, J.R., and Barrett, D.M. (2009). Effects of organic and conventional production systems on quality and nutritional parameters of processing tomatoes. *Journal of the Science of Food and Agriculture*, *89*(2), 177-194.
14. Zushi, K., Matsuzoe, N., Kitano, M. (2009) Developmental and tissue-specific changes in oxidative parameters and antioxidant systems in tomato fruits grown under salt stress. *Sci Hortic* 122: 362–368.

# CHAPTER 6: OVERALL CONCLUSIONS & RECCOMMENDATIONS

## 6.1 CONCLUSIONS

This study concludes as follows on the research objectives:

1. The first objective was to formulate hydroponic nutrient solution from Goat Manure. Findings from the current study suggest that there are unique sites in Namibia like Utuseb, where raw goat manure is enriched with mineral nutrients for hydroponic nutrient solution formulation. The current study's findings also suggest that there are unique sites like the Sam Nujoma Marine and Coastal Resources Research Centre (SANUMARC) where sufficient nitrifying microorganisms can be sourced to convert organic nitrogen in animal manure like goat manure into nitrates. The present study's findings further suggest that nitrification of manure such as goat manure is possible with locally sourced microorganisms leading to a quality organic hydroponic nutrient solution. Thus, it can be concluded that a hydroponic nutrient solution can be formulated from goat manure.

2. The second objective was to determine performance of tomato due to the developed hydroponic solution.

   Findings from the current study suggest that tomato production using local Goat Manure Derived Hydroponic Nutrient Solution (GMDHNS) positively influenced tomato growth and reproduction significantly more than when using Tap Water (TW). In fact, tomato growth and reproduction of GMDHNS compared relatively well with that of the Commercial Hydroponic Nutrient Solution (CHNS). Thus, it

can be concluded that the locally formulated hydroponic nutrient solution enhances growth and reproductive performance of tomato, even though it may still need further optimization.

3. The third objective was to determine the quality of tomatoes produced from organic hydroponic nutrients.

   Findings from the current study suggest that total soluble solids and lycopene content were higher in tomato fruits from the locally formulated hydroponic nutrient solution, even though the quantity was higher in the conventional hydroponic solution. Thus, it can be concluded that locally formulated hydroponic solutions in horticultural production improve quality of produce even though not to the same level of quantity as that of conventional hydroponic nutrient solution.

Therefore, all the objectives that were set for this study have been met at the end of the study. In addition, the study concludes that a new tomato value chain has been created with the locally formulated nutrient solution being the entry point in the value chain which will be dictated by a market of the current generation on reform to healthful diets. The quality of tomato fruits produced from the locally formulated nutrient solution is more beneficial than the quantity produced in this regard.

Overall, the contribution of the findings of this study to new knowledge is that the formulated hydroponic nutrient solution from goat manure is an input to artefactual (lab in the field) that leads to a new value chain. Furthermore, considering that this study was

the first of its kind in Namibia and Africa, the findings presented here serve as a baseline for further studies.

## 6.2 RECCOMENDATIONS

The present study was a baseline study for Africa, considering there has never been a similar study in Africa in general and Namibia in particular, infact there has never been a documented study on goat manure hydroponically even at the global level. Hence there is a need for further studies to follow-up on the results of this study so that a complete understanding on all aspects related to organic nutrient solutions hydroponically will be found. Such aspects will include buffering pH and EC, degradation rates of organic nitrogen into nitrates from different manures and many others.

The findings from the current study are recommended to be a contribution for the green economy policy framework where they can be taken into artefactual and field framed leading to a new tomato value chain for Namibia. This is provided that an enabling environment is availed by the Government of Namibia through the Ministry of Agriculture, Water and Forestry and other relevant agencies to involve all potential farmers from urban to rural areas. This will make Namibia a production hub even in arid conditions, just as Israel has done for years by producing enough for own consumption and surplus fruits and vegetables for export to other countries, yet Israel is an arid country. Once this innovation gets upscaled by business investors then it will eventually reach the targeted end users, the people who will do mass production of tomatoes using the

formulated nutrient solution. Therefore, the next step is to market the innovation to investors.

Based on the findings of the present study, it is recommended that;

1. Further studies on shelf life of the formulated nutrient solution from goat manure be done as part of the Artefactual which will appeal to business investors and lead to business adoption of the current innovation.
2. Extension of the current results to cattle manure is a further study recommended especially in areas of Namibia where crop production is not possible due to water and fertile soils yet cattle production is intense, leaving cattle manure without value in such areas. Such a practice will not conflict the current field practices in areas with sufficient water where cattle manure gets applied to soils to provide organic matter and fertility. Therefore, open field cultivation will coexist with hydroponics cultivation because their geographic areas of operation will be different in Namibia.
3. Soils with abundant nitrifying microorganisms be sought and used as sources of microbial consortia to degrade organic nitrogen into nitrates from goat manure. Such soils include natural composting sites and legume soils.
4. To minimise on input costs, materials for construction of hydroponic tables should include simple items such planks, plastic sheeting and styroforms or other cheaper alternatives, without using air pumps and other sophisticated equipment.
5. Quality end product should be the focus of the upcoming farmers who will upscale the findings of the present research to higher levels.

# APPENDICES

## APPENDIX 1 (A): Metagenomic BLAST results for the Garden inoculum source: amoA gene

| Organism/HIT | Accession | e-value | Fastq header |
|---|---|---|---|
| No hits | None | 0 | M01232:27:000000000-ARHCV:1:1119:19319:4876 |
| uncultured bacterium | gi\|692348421\|gb\|KM030899.1\| Uncultured bacterium clone CT3B28 ammonia monooxygenase (amoA) gene, partial cds | 2.89043e-121 | M01232:27:000000000-ARHCV:1:1114:18794:4073 |
| uncultured ammonia-oxidizing | gi\|913469413\|gb\|KP781186.1\| Uncultured ammonia-oxidizing bacterium clone SBE1 ammonia monooxygenase subunit A (amoA) gene, partial cds | 2.79966e-146 | M01232:27:000000000-ARHCV:1:2107:12053:10313 |
| uncultured beta | gi\|910275809\|gb\|KP783383.1\| Uncultured beta proteobacterium clone G5B17 ammonia monooxygenase subunit A (amoA) gene, partial cds | 4.24588e-51 | M01232:27:000000000-ARHCV:1:1112:16821:22160 |
| pseudomonas aeruginosa | gi\|861670927\|gb\|CP012001.1\| Pseudomonas aeruginosa DSM 50071, complete genome | 2.77993e-151 | M01232:27:000000000-ARHCV:1:2108:9616:13803 |
| uncultured nitrosospira | gi\|78057483\|gb\|DQ208958.1\| Uncultured Nitrosospira sp. isolate DGGE gel band SD03 AmoA (amoA) gene, partial cds | 2.7953e-48 | M01232:27:000000000-ARHCV:1:1103:7480:12275 |
| janthinobacterium agaricidamnosum | gi\|571265423\|emb\|HG322949.1\| Janthinobacterium agaricidamnosum NBRC 102515 = DSM 9628, complete genome | 1.51227e-38 | M01232:27:000000000-ARHCV:1:1112:25613:21598 |
| gordonia sp. | gi\|403643428\|gb\|CP002907.1\| Gordonia sp. KTR9, complete genome | 6.84449e-24 | M01232:27:000000000-ARHCV:1:1106:21868:23716 |
| homo sapiens | gi\|584458501\|ref\|NG_033930.1\| Homo sapiens SH3-domain binding protein 4 (SH3BP4), RefSeqGene on chromosome 2 | 0.0012278 2 | M01232:27:000000000-ARHCV:1:2103:24967:22356 |

| | | | |
|---|---|---|---|
| uncultured nitrosomonadales | gi\|685453658\|gb\|KJ949143.1\| Uncultured Nitrosomonadales bacterium clone ZY-HF-1 ammonia monooxygenase (amoA) gene, partial cds >gi\|685453660\|gb\|KJ949144.1\| Uncultured Nitrosomonadales bacterium clone JD-HF-1 ammonia monooxygenase (amoA) gene, partial cds >gi\|685453662\|gb\|KJ949145.1\| Uncultured Nitrosomonadales bacterium clone LZ-HF-1 ammonia monooxygenase (amoA) gene, partial cds >gi\|685453664\|gb\|KJ949146.1\| Uncultured Nitrosomonadales bacterium clone JX-HF-1 ammonia monooxygenase (amoA) gene, partial cds | 2.83956e-136 | M01232:27:000000000-ARHCV:1:2113:20863:11882 |
| thiobacillus denitrificans | gi\|74055513\|gb\|CP000116.1\| Thiobacillus denitrificans ATCC 25259, complete genome | 5.43908e-38 | M01232:27:000000000-ARHCV:1:1113:14243:18941 |
| archangium gephyra | gi\|827418478\|gb\|CP011509.1\| Archangium gephyra strain DSM 2261, complete genome | 3.32119e-37 | M01232:27:000000000-ARHCV:1:1108:27202:7646 |
| african horse | gi\|768677459\|gb\|KP009697.1\| African horse sickness virus isolate HS 29/00 segment 7, complete sequence | 5.66381e-08 | M01232:27:000000000-ARHCV:1:2109:24296:13710 |
| influenza a | gi\|398324780\|gb\|JX273568.1\| Influenza A virus (A/teal/Northern Ireland/14567-10-5257/2007(H9N1)) segment 4 hemagglutinin (HA) gene, complete cds | 6.22313e-93 | M01232:27:000000000-ARHCV:1:1103:24499:15954 |
| erythrobacter litoralis | gi\|84785911\|gb\|CP000157.1\| Erythrobacter litoralis HTCC2594, complete genome | 2.13418e-14 | M01232:27:000000000-ARHCV:1:1117:18742:6238 |
| blastococcus saxobsidens | gi\|378781357\|emb\|FO117623.1\| Blastococcus saxobsidens DD2 complete genome | 3.63491e-29 | M01232:27:000000000-ARHCV:1:1117:27942:7558 |
| triticum aestivum | gi\|669026884\|emb\|HG670306.1\| Triticum aestivum chromosome 3B, genomic scaffold, cultivar Chinese Spring | 3.41298e-06 | M01232:27:000000000-ARHCV:1:1107:25773:5980 |

| | | | |
|---|---|---|---|
| trypanosoma cruzi | gi\|71652421\|ref\|XM_809775.1\| Trypanosoma cruzi strain CL Brener vacuolar-type proton translocating pyrophosphatase 1 partial mRNA | 2.11649e-05 | M01232:27:000000000-ARHCV:1:1106:10280:7900 |
| amycolatopsis methanolica | gi\|672354214\|gb\|CP009110.1\| Amycolatopsis methanolica 239, complete genome | 3.76552e-15 | M01232:27:000000000-ARHCV:1:2118:9484:6986 |
| nitrosospira sp. | gi\|81251131\|gb\|DQ228455.1\| Nitrosospira sp. 9SS1 ammonia monooxygenase subunit A (amoA) gene, partial cds | 6.21258e-123 | M01232:27:000000000-ARHCV:1:1103:11329:11668 |
| echinostoma caproni | gi\|688640536\|emb\|LL269994.1\| Echinostoma caproni genome assembly E_caproni_Egypt ,scaffold ECPE_scaffold0034352 | 3.23871e-09 | M01232:27:000000000-ARHCV:1:1117:11798:22283 |
| mycobacterium smegmatis | gi\|777214100\|emb\|LN831039.1\| Mycobacterium smegmatis genome assembly NCTC8159, chromosome : 1 | 1.71612e-31 | M01232:27:000000000-ARHCV:1:1110:9502:15329 |
| heliobacterium modesticaldum | gi\|171696369\|gb\|CP000930.2\| Heliobacterium modesticaldum Ice1, complete genome | 1.06941e-25 | M01232:27:000000000-ARHCV:1:1115:21954:6601 |
| dermacoccus nishinomiyaensis | gi\|664687059\|gb\|CP008889.1\| Dermacoccus nishinomiyaensis strain M25, complete genome | 2.175e-35 | M01232:27:000000000-ARHCV:1:2104:21407:17681 |
| methylobacterium extorquens | gi\|254265931\|emb\|FP103042.2\| Methylobacterium extorquens DM4 str. DM4 chromosome, complete genome | 6.08508e-10 | M01232:27:000000000-ARHCV:1:2112:21029:23893 |
| predicted: pantholops | gi\|556737072\|ref\|XM_005964065.1\| PREDICTED: Pantholops hodgsonii COBW domain-containing protein DDB_G0274527-like (LOC102333664), mRNA | 3.43835e-05 | M01232:27:000000000-ARHCV:1:1118:5755:11621 |
| infectious bronchitis | gi\|925169335\|gb\|KP868573.1\| Infectious bronchitis virus strain CK/CH/LJL/130908, complete genome | 2.14883e-59 | M01232:27:000000000-ARHCV:1:2114:20716:21363 |
| apteryx australis | gi\|840011424\|emb\|LK064761.1\| Apteryx australis mantelli genome assembly AptMant0, scaffold scaffold101 | 2.83039e-75 | M01232:27:000000000-ARHCV:1:1106:23484:13532 |

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|---|---|---|---|
| pseudomonas stutzeri | gi\|431823496\|gb\|CP003071.1\| Pseudomonas stutzeri RCH2, complete genome | 1.05508e-59 | M01232:27:000000000-ARHCV:1:1117:20583:19980 |
| bradyrhizobium japonicum | gi\|736032532\|gb\|CP010313.1\| Bradyrhizobium japonicum strain E109, complete genome | 1.91048e-53 | M01232:27:000000000-ARHCV:1:1119:28520:13147 |
| geodermatophilus obscurus | gi\|284061874\|gb\|CP001867.1\| Geodermatophilus obscurus DSM 43160, complete genome | 9.68935e-05 | M01232:27:000000000-ARHCV:1:2103:27256:13247 |
| sinorhizobium meliloti | gi\|334098883\|gb\|CP002783.1\| Sinorhizobium meliloti AK83 chromosome 3, complete sequence | 3.0919e-61 | M01232:27:000000000-ARHCV:1:1101:21564:8374 |
| microbacterium sp. | gi\|914697494\|gb\|CP012299.1\| Microbacterium sp. CGR1, complete genome | 4.75249e-13 | M01232:27:000000000-ARHCV:1:2102:23130:23613 |
| rubrivivax gelatinosus | gi\|381376528\|dbj\|AP012320.1\| Rubrivivax gelatinosus IL144 DNA, complete genome | 2.93613e-10 | M01232:27:000000000-ARHCV:1:1117:15625:3329 |
| sphingomonas sp. | gi\|469477505\|gb\|CP004036.1\| Sphingomonas sp. MM-1, complete genome | 3.29203e-18 | M01232:27:000000000-ARHCV:1:2108:14191:18017 |
| cedecea neteri | gi\|689262542\|gb\|CP009451.1\| Cedecea neteri strain SSMD04, complete genome | 7.33573e-08 | M01232:27:000000000-ARHCV:1:2110:15976:21362 |
| uncultured nitrite-oxidizing | gi\|724471095\|gb\|KM408671.1\| Uncultured nitrite-oxidizing bacterium clone IB2-2 nitrite oxidoreductase alpha subunit (nxrA) gene, partial cds | 1.77382e-103 | M01232:27:000000000-ARHCV:1:2111:21088:24323 |
| uncultured bacteroidales | gi\|192792311\|gb\|EU794260.1\| Uncultured Bacteroidales bacterium clone EMP_F18 16S ribosomal RNA gene, partial sequence | 1.3118e-139 | M01232:27:000000000-ARHCV:1:1110:8483:9760 |
| predicted: musa | gi\|695051105\|ref\|XM_009415291.1\| PREDICTED: Musa acuminata subsp. malaccensis adenosylhomocysteinase-like (LOC103994848), mRNA | 1.1735e-07 | M01232:27:000000000-ARHCV:1:1106:6945:14483 |

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|---|---|---|---|
| rhodopseudomonas palustris | gi\|315599110\|gb\|CP002418.1\| Rhodopseudomonas palustris DX-1, complete genome | 2.10278e-07 | M01232:27:000000000-ARHCV:1:2108:25795:7667 |
| paracoccus denitrificans | gi\|119376152\|gb\|CP000490.1\| Paracoccus denitrificans PD1222 chromosome 2, complete sequence | 3.1862e-05 | M01232:27:000000000-ARHCV:1:2102:9863:16949 |
| ovis canadensis | gi\|850492351\|gb\|CP011904.1\| Ovis canadensis canadensis isolate 43U chromosome 19 sequence | 3.85668e-14 | M01232:27:000000000-ARHCV:1:2101:19104:12507 |
| hoeflea sp. | gi\|822663067\|gb\|CP011479.1\| Hoeflea sp. IMCC20628, complete genome | 1.22619e-24 | M01232:27:000000000-ARHCV:1:1112:26091:20125 |
| devosia sp. | gi\|901895891\|gb\|CP011300.1\| Devosia sp. H5989, complete genome | 1.34941e-13 | M01232:27:000000000-ARHCV:1:2113:16579:5810 |
| fusarium pseudograminearum | gi\|685851168\|ref\|XM_009254342.1\| Fusarium pseudograminearum CS3096 hypothetical protein partial mRNA | 0.000476508 | M01232:27:000000000-ARHCV:1:2118:12988:16290 |
| luteimonas mephitis | gi\|672710205\|emb\|LM994050.1\| Luteimonas mephitis partial groEL gene for chaperonin, strain CIP 107229 | 9.42247e-12 | M01232:27:000000000-ARHCV:1:2102:22813:24857 |
| polymorphum gilvum | gi\|326411376\|gb\|CP002568.1\| Polymorphum gilvum SL003B-26A1, complete genome | 1.95508e-06 | M01232:27:000000000-ARHCV:1:1111:15440:11324 |
| gordonia bronchialis | gi\|262083393\|gb\|CP001802.1\| Gordonia bronchialis DSM 43247, complete genome | 3.26197e-15 | M01232:27:000000000-ARHCV:1:1109:26739:20079 |
| uncultured firmicutes | gi\|154757251\|gb\|EU029484.1\| Uncultured Firmicutes bacterium clone T4266 16S ribosomal RNA gene, partial sequence | 6.16878e-128 | M01232:27:000000000-ARHCV:1:2116:27086:20899 |
| geobacter uraniireducens | gi\|146395585\|gb\|CP000698.1\| Geobacter uraniireducens Rf4, complete genome | 0.00338821 | M01232:27:000000000-ARHCV:1:1105:21842:12781 |

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|---|---|---|---|
| nitrobacter winogradskyi | gi\|542214432\|gb\|KF437381.1\| Nitrobacter winogradskyi isolate DGGE gel band 5-1 NorA (norA) gene, partial cds | 2.19131e-137 | M01232:27:000000000-ARHCV:1:2104:7604:20013 |
| methylocella silvestris | gi\|217501576\|gb\|CP001280.1\| Methylocella silvestris BL2, complete genome | 2.14685e-20 | M01232:27:000000000-ARHCV:1:2111:9373:16764 |
| nitrosomonas sp. | gi\|338803667\|gb\|CP002876.1\| Nitrosomonas sp. Is79A3, complete genome | 4.00818e-20 | M01232:27:000000000-ARHCV:1:1115:27399:16093 |
| mesorhizobium australicum | gi\|433663430\|gb\|CP003358.1\| Mesorhizobium australicum WSM2073, complete genome | 1.75972e-43 | M01232:27:000000000-ARHCV:1:2104:24556:19114 |
| rubrobacter xylanophilus | gi\|108764099\|gb\|CP000386.1\| Rubrobacter xylanophilus DSM 9941, complete genome | 6.07446e-30 | M01232:27:000000000-ARHCV:1:1118:7252:12159 |
| pseudomonas mendocina | gi\|145573243\|gb\|CP000680.1\| Pseudomonas mendocina ymp, complete genome | 2.32093e-34 | M01232:27:000000000-ARHCV:1:2112:22790:22795 |
| vibrio cholerae | gi\|327482915\|gb\|CP002555.1\| Vibrio cholerae LMA3894-4 chromosome I, complete sequence | 6.30182e-21 | M01232:27:000000000-ARHCV:1:2105:17130:6521 |
| nocardioides sp. | gi\|119534933\|gb\|CP000509.1\| Nocardioides sp. JS614, complete genome | 3.62229e-24 | M01232:27:000000000-ARHCV:1:1116:23748:19150 |
| pelobacter propionicus | gi\|118501159\|gb\|CP000482.1\| Pelobacter propionicus DSM 2379, complete genome | 5.3782e-44 | M01232:27:000000000-ARHCV:1:1118:10624:14435 |
| azospirillum lipoferum | gi\|357422594\|emb\|FQ311868.1\| Azospirillum lipoferum 4B main chromosome, complete genome | 3.74477e-19 | M01232:27:000000000-ARHCV:1:1118:15926:2324 |
| bacillus cereus | gi\|753559736\|gb\|CP009590.1\| Bacillus cereus G9241, complete genome | 1.41306e-84 | M01232:27:000000000-ARHCV:1:1111:4949:5288 |

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|---|---|---|---|
| brevibacillus brevis | gi\|226092535\|dbj\|AP008955.1\| Brevibacillus brevis NBRC 100599 DNA, complete genome | 9.45653e-07 | M01232:27:000000000-ARHCV:1:2105:20822:1845 |
| marinobacter salarius | gi\|582027004\|gb\|CP007152.1\| Marinobacter salarius strain R9SW1, complete genome | 6.8719e-14 | M01232:27:000000000-ARHCV:1:1112:10280:22749 |
| sphingomonas wittichii | gi\|148498119\|gb\|CP000699.1\| Sphingomonas wittichii RW1, complete genome | 1.04421e-35 | M01232:27:000000000-ARHCV:1:1104:12806:24921 |
| uncultured rumen | gi\|113927692\|dbj\|AB270373.1\| Uncultured rumen bacterium gene for 16S rRNA, partial sequence, clone: T28H60SE01 | 4.70993e-139 | M01232:27:000000000-ARHCV:1:1115:7931:2043 |
| natrinema pellirubrum | gi\|433304139\|gb\|CP003372.1\| Natrinema pellirubrum DSM 15624, complete genome | 7.91041e-11 | M01232:27:000000000-ARHCV:1:1115:27312:7525 |
| uncultured verrucomicrobiales | gi\|192792224\|gb\|EU794173.1\| Uncultured Verrucomicrobiales subdivision 5 bacterium clone EMP_D8 16S ribosomal RNA gene, partial sequence | 2.16053e-147 | M01232:27:000000000-ARHCV:1:1114:28759:14048 |
| sphingobium sp. | gi\|764452294\|gb\|CP010954.1\| Sphingobium sp. YBL2, complete genome | 2.11693e-20 | M01232:27:000000000-ARHCV:1:2103:22399:10752 |
| [polyangium] brachysporum | gi\|826168461\|gb\|CP011371.1\| [Polyangium] brachysporum strain DSM 7029, complete genome | 2.99817e-44 | M01232:27:000000000-ARHCV:1:2105:11014:5178 |
| cyprinus carpio | gi\|685042180\|emb\|LN590705.1\| Cyprinus carpio genome assembly common carp genome ,scaffold LG11 | 1.76128e-22 | M01232:27:000000000-ARHCV:1:1114:9763:24979 |
| predicted: clupea | gi\|831297920\|ref\|XR_001162309.1\| PREDICTED: Clupea harengus uncharacterized LOC105898325 (LOC105898325), transcript variant X2, ncRNA | 3.66641e-05 | M01232:27:000000000-ARHCV:1:1109:13460:19828 |
| candidatus chloracidobacterium | gi\|347586250\|gb\|CP002514.1\| Candidatus Chloracidobacterium thermophilum B chromosome 1, complete sequence | 1.6724e-43 | M01232:27:000000000-ARHCV:1:2110:22215:13900 |

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|---|---|---|---|
| deinococcus gobiensis | gi\|379998737\|gb\|CP002191.1\| Deinococcus gobiensis I-0, complete genome | 6.30953e-06 | M01232:27:000000000-ARHCV:1:1112:25030:17024 |
| sorangium cellulosum | gi\|520999024\|gb\|CP003969.1\| Sorangium cellulosum So0157-2, complete genome | 3.20926e-10 | M01232:27:000000000-ARHCV:1:1113:21301:12336 |
| massilia sp. | gi\|909841042\|gb\|CP012201.1\| Massilia sp. NR 4-1, complete genome | 3.55204e-49 | M01232:27:000000000-ARHCV:1:1114:25681:8055 |
| solibacter usitatus | gi\|116222307\|gb\|CP000473.1\| Solibacter usitatus Ellin6076, complete genome | 9.20863e-27 | M01232:27:000000000-ARHCV:1:1113:29059:12957 |
| intrasporangium calvum | gi\|315587265\|gb\|CP002343.1\| Intrasporangium calvum DSM 43043, complete genome | 0.000547629 | M01232:27:000000000-ARHCV:1:2118:18169:16123 |
| mycobacterium vanbaalenii | gi\|119953846\|gb\|CP000511.1\| Mycobacterium vanbaalenii PYR-1, complete genome | 1.90335e-46 | M01232:27:000000000-ARHCV:1:2118:25232:21137 |
| gluconacetobacter xylinus | gi\|582020631\|gb\|CP004360.1\| Gluconacetobacter xylinus E25, complete genome | 5.88464e-06 | M01232:27:000000000-ARHCV:1:2105:16055:15877 |
| burkholderia sp. | gi\|506944542\|dbj\|AP013059.1\| Burkholderia sp. RPE64 DNA, chromosome 2, complete genome | 1.39643e-10 | M01232:27:000000000-ARHCV:1:2110:16258:18577 |
| bradyrhizobium diazoefficiens | gi\|806922190\|dbj\|AP014685.1\| Bradyrhizobium diazoefficiens DNA, complete genome, strain: NK6 | 1.86039e-73 | M01232:27:000000000-ARHCV:1:2112:16016:10689 |
| sandaracinus amylolyticus | gi\|816953346\|gb\|CP011125.1\| Sandaracinus amylolyticus strain DSM 53668, complete genome | 7.65224e-72 | M01232:27:000000000-ARHCV:1:2117:13448:3581 |
| stigmatella aurantiaca | gi\|309390350\|gb\|CP002271.1\| Stigmatella aurantiaca DW4/3-1, complete genome | 1.08843e-90 | M01232:27:000000000-ARHCV:1:2113:16105:16951 |

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|---|---|---|---|
| azoarcus aromaticum | gi\|56311475\|emb\|CR555306.1\| Azoarcus aromaticum EbN1 complete genome | 6.2408e-14 | M01232:27:000000000-ARHCV:1:1107:12351:21858 |
| serratia sp. | gi\|640856369\|gb\|CP005927.1\| Serratia sp. FS14, complete genome | 2.85001e-05 | M01232:27:000000000-ARHCV:1:1102:27117:6874 |
| uncultured prokaryote | gi\|717490281\|gb\|KM410669.1\| Uncultured prokaryote clone GS10-10-58 16S ribosomal RNA gene, partial sequence | 6.36787e-108 | M01232:27:000000000-ARHCV:1:2107:22081:16917 |
| chain 5, | gi\|485601478\|pdb\|3J3F\|5 Chain 5, Structure Of The H. Sapiens 60s Rrna | 5.29414e-35 | M01232:27:000000000-ARHCV:1:1101:13206:20834 |
| rhodospirillum photometricum | gi\|378401447\|emb\|HE663493.1\| Rhodospirillum photometricum DSM 122 draft genome sequence | 1.39588e-19 | M01232:27:000000000-ARHCV:1:2106:17100:19800 |
| staphylococcus haemolyticus | gi\|807232188\|gb\|CP011116.1\| Staphylococcus haemolyticus strain Sh29/312/L2, complete genome | 4.16482e-45 | M01232:27:000000000-ARHCV:1:1105:20470:17900 |
| achromobacter xylosoxidans | gi\|874219584\|gb\|CP012046.1\| Achromobacter xylosoxidans strain MN001, complete genome | 0.000149725 | M01232:27:000000000-ARHCV:1:1114:23247:11402 |

## APPENDIX 1 (B): Metagenomic BLAST results for the Compost inoculum source: amoA gene

| Organism/HIT | Accession | e-value | Fastq header |
|---|---|---|---|
| No hits | None | 0 | M01232:27:000000000-ARHCV:1:1116:6310:12312 |

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|---|---|---|---|
| uncultured ammonia-oxidizing | gi\|310644898\|gb\|HM481189.1\| Uncultured ammonia-oxidizing bacterium clone C-3 ammonia monooxygenase subunit A (amoA) gene, partial cds | 2.19511e-137 | M01232:27:000000000-ARHCV:1:1106:23664:10168 |
| uncultured bacterium | gi\|727358850\|gb\|KM404662.1\| Uncultured bacterium clone BI4 ammonia monooxygenase gene, partial cds | 3.25011e-43 | M01232:27:000000000-ARHCV:1:2112:18746:4112 |
| nitrosospira sp. | gi\|81251149\|gb\|DQ228464.1\| Nitrosospira sp. NIJS18 ammonia monooxygenase subunit A (amoA) gene, partial cds | 2.89043e-121 | M01232:27:000000000-ARHCV:1:1101:17803:9943 |
| pseudomonas aeruginosa | gi\|610413304\|gb\|CP007224.1\| Pseudomonas aeruginosa PA96 genome | 1.28426e-154 | M01232:27:000000000-ARHCV:1:1108:21007:8526 |
| uncultured intrasporangium | gi\|643015984\|gb\|KF117320.1\| Uncultured Intrasporangium sp. clone GPMKYIG01DO1W4 genomic sequence | 5.35691e-09 | M01232:27:000000000-ARHCV:1:1106:12187:21118 |
| propionibacterium acnes | gi\|657118275\|gb\|CP006032.1\| Propionibacterium acnes hdn-1, complete genome | 4.25218e-20 | M01232:27:000000000-ARHCV:1:2113:18573:11781 |
| pseudomonas chlororaphis | gi\|787852299\|gb\|CP011110.1\| Pseudomonas chlororaphis strain PCL1606, complete genome | 9.63456e-08 | M01232:27:000000000-ARHCV:1:2113:10417:20722 |
| dechlorosoma suillum | gi\|359353254\|gb\|CP003153.1\| Dechlorosoma suillum PS, complete genome | 1.60071e-06 | M01232:27:000000000-ARHCV:1:1115:6145:18684 |

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|---|---|---|---|
| uncultured beta | gi\|387538963\|gb\|JQ638763.1\| Uncultured beta proteobacterium clone WWTP-G9 ammonia monooxygenase subunit A (amoA) gene, partial cds | 1.29136e-51 | M01232:27:000000000-ARHCV:1:2112:21650:15302 |
| polymorphum gilvum | gi\|326411376\|gb\|CP002568.1\| Polymorphum gilvum SL003B-26A1, complete genome | 9.27401e-22 | M01232:27:000000000-ARHCV:1:2109:16028:7531 |
| conus episcopatus | gi\|920148880\|dbj\|AK443485.1\| Conus episcopatus mRNA, sequence id | 1.69783e-18 | M01232:27:000000000-ARHCV:1:1105:25504:3567 |
| rhodospirillum centenum | gi\|288926859\|gb\|CP000613.2\| Rhodospirillum centenum SW, complete genome | 8.17902e-06 | M01232:27:000000000-ARHCV:1:2105:13352:11949 |
| predicted: microplitis | gi\|665784875\|ref\|XM_008545999.1\| PREDICTED: Microplitis demolitor ribosome biogenesis protein NSA2 homolog (LOC103568951), mRNA | 3.87007e-13 | M01232:27:000000000-ARHCV:1:1112:20390:1497 |
| african horse | gi\|768677459\|gb\|KP009697.1\| African horse sickness virus isolate HS 29/00 segment 7, complete sequence | 1.87279e-07 | M01232:27:000000000-ARHCV:1:1116:21193:18714 |
| cronobacter turicensis | gi\|323575285\|emb\|FN543093.2\| Cronobacter turicensis z3032 complete genome | 7.02587e-25 | M01232:27:000000000-ARHCV:1:1111:12650:13426 |
| homo sapiens | gi\|563580301\|ref\|NG_033851.1\| Homo sapiens transcription factor 12 (TCF12), RefSeqGene on chromosome 15 | 5.2984e-08 | M01232:27:000000000-ARHCV:1:2119:8621:8861 |

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|---|---|---|---|
| pseudomonas pseudoalcaligenes | gi\|652789639\|emb\|LK391695.1\| Pseudomonas pseudoalcaligenes genome assembly Ppseudo_Pac ,chromosome : I | 1.63391e-24 | M01232:27:000000000-ARHCV:1:1107:20783:18463 |
| nitrosomonas sp. | gi\|62945208\|dbj\|AB212172.1\| Nitrosomonas sp. NS20 gene for ammonia monooxygenase subunit A, partial cds | 2.7953e-48 | M01232:27:000000000-ARHCV:1:1118:25215:14130 |
| alcanivorax pacificus | gi\|745803723\|gb\|CP004387.1\| Alcanivorax pacificus W11-5, complete genome | 1.82132e-88 | M01232:27:000000000-ARHCV:1:2116:14959:22159 |
| dactylococcopsis salina | gi\|428692674\|gb\|CP003944.1\| Dactylococcopsis salina PCC 8305, complete genome | 3.49128e-08 | M01232:27:000000000-ARHCV:1:1118:27099:11920 |
| cyprinus carpio | gi\|685042601\|emb\|LN591126.1\| Cyprinus carpio genome assembly common carp genome ,scaffold 000028826 | 4.96933e-18 | M01232:27:000000000-ARHCV:1:1116:18188:4115 |
| frankia sp. | gi\|311225233\|gb\|CP002299.1\| Frankia sp. EuI1c, complete genome | 3.34404e-34 | M01232:27:000000000-ARHCV:1:2119:25256:20580 |
| pseudomonas resinovorans | gi\|512374267\|dbj\|AP013068.1\| Pseudomonas resinovorans NBRC 106553 DNA, complete geonome | 6.84575e-72 | M01232:27:000000000-ARHCV:1:2117:9258:12803 |
| gordonia sp. | gi\|403643428\|gb\|CP002907.1\| Gordonia sp. KTR9, complete genome | 2.47919e-18 | M01232:27:000000000-ARHCV:1:2115:12897:5845 |

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|---|---|---|---|
| influenza a | gi\|484848971\|gb\|KC871517.1\| Influenza A virus (A/ruddy shelduck/Mongolia/974/2010(H10N7)) segment 8 nuclear export protein (NEP) and nonstructural protein 1 (NS1) genes, complete cds | 2.10643e-122 | M01232:27:000000000-ARHCV:1:2118:11807:8129 |
| hyphomicrobium denitrificans | gi\|299523359\|gb\|CP002083.1\| Hyphomicrobium denitrificans ATCC 51888, complete genome | 1.0625e-10 | M01232:27:000000000-ARHCV:1:1115:14937:24655 |
| myxococcus fulvus | gi\|337255776\|gb\|CP002830.1\| Myxococcus fulvus HW-1, complete genome | 0.00100604 | M01232:27:000000000-ARHCV:1:1118:14014:11847 |
| mesorhizobium huakuii | gi\|657121522\|gb\|CP006581.1\| Mesorhizobium huakuii 7653R genome | 1.49794e-44 | M01232:27:000000000-ARHCV:1:2118:5552:7818 |
| enterobacter cloacae | gi\|723224579\|gb\|CP009854.1\| Enterobacter cloacae strain ECNIH5, complete genome | 5.21677e-35 | M01232:27:000000000-ARHCV:1:2118:13770:22522 |
| burkholderia gladioli | gi\|772900580\|gb\|CP009322.1\| Burkholderia gladioli strain ATCC 10248 chromosome 2, complete sequence | 6.43699e-06 | M01232:27:000000000-ARHCV:1:1118:19683:2193 |
| predicted: camelus | gi\|560893763\|ref\|XM_006173320.1\| PREDICTED: Camelus ferus uncharacterized LOC102511175 (LOC102511175), partial mRNA | 0.00448779 | M01232:27:000000000-ARHCV:1:1110:17639:6370 |
| rhodomicrobium vannielii | gi\|311217923\|gb\|CP002292.1\| Rhodomicrobium vannielii ATCC 17100, complete genome | 2.33256e-07 | M01232:27:000000000-ARHCV:1:2111:22345:9948 |

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|---|---|---|---|
| sinorhizobium meliloti | gi\|675826159\|gb\|CP009144.1\| Sinorhizobium meliloti strain RMO17, complete genome | 1.12567e-65 | M01232:27:000000000-ARHCV:1:2107:13027:23153 |
| bradyrhizobium diazoefficiens | gi\|806922190\|dbj\|AP014685.1\| Bradyrhizobium diazoefficiens DNA, complete genome, strain: NK6 | 1.0226e-39 | M01232:27:000000000-ARHCV:1:2105:18639:9176 |
| ramlibacter tataouinensis | gi\|334728683\|gb\|CP000245.1\| Ramlibacter tataouinensis TTB310, complete genome | 7.20736e-26 | M01232:27:000000000-ARHCV:1:2116:13052:24115 |
| nocardiopsis dassonvillei | gi\|296843433\|gb\|CP002040.1\| Nocardiopsis dassonvillei subsp. dassonvillei DSM 43111 chromosome 1, complete sequence | 6.17068e-14 | M01232:27:000000000-ARHCV:1:2102:12079:6323 |
| uncultured prokaryote | gi\|874565226\|emb\|LN853019.1\| Uncultured prokaryote from Rat gut metagenome metamobilome, plasmid pRGRH0356 | 1.54297e-68 | M01232:27:000000000-ARHCV:1:1114:26242:17778 |
| sphingobium chlorophenolicum | gi\|334100279\|gb\|CP002798.1\| Sphingobium chlorophenolicum L-1 chromosome 1, complete sequence | 1.27516e-53 | M01232:27:000000000-ARHCV:1:1115:10647:21883 |
| geobacter bemidjiensis | gi\|197085762\|gb\|CP001124.1\| Geobacter bemidjiensis Bem, complete genome | 2.00339e-18 | M01232:27:000000000-ARHCV:1:2103:22091:9327 |
| bradyrhizobium sp. | gi\|146189981\|emb\|CU234118.1\| Bradyrhizobium sp. ORS278,complete sequence | 7.06142e-76 | M01232:27:000000000-ARHCV:1:1110:21094:12888 |

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|---|---|---|---|
| piscirickettsia salmonis | gi\|923110819\|gb\|CP012508.1\| Piscirickettsia salmonis strain PM32597B1, complete genome | 0.000142374 | M01232:27:000000000-ARHCV:1:2114:27034:12696 |
| salpingoeca rosetta | gi\|514700273\|ref\|XM_004997648.1\| Salpingoeca rosetta methionine synthase mRNA | 9.09612e-05 | M01232:27:000000000-ARHCV:1:1103:6289:17565 |
| bradyrhizobium japonicum | gi\|627779227\|gb\|CP007569.1\| Bradyrhizobium japonicum SEMIA 5079 genome | 1.8736e-68 | M01232:27:000000000-ARHCV:1:1104:13089:13936 |
| sphingopyxis fribergensis | gi\|734569737\|gb\|CP009122.1\| Sphingopyxis fribergensis strain Kp5.2, complete genome | 7.22628e-16 | M01232:27:000000000-ARHCV:1:1101:27604:20335 |
| pseudomonas stutzeri | gi\|848254014\|gb\|CP011854.1\| Pseudomonas stutzeri strain SLG510A3-8, complete genome | 4.80211e-09 | M01232:27:000000000-ARHCV:1:1104:11756:6728 |
| mesorhizobium opportunistum | gi\|336024847\|gb\|CP002279.1\| Mesorhizobium opportunistum WSM2075, complete genome | 5.24629e-64 | M01232:27:000000000-ARHCV:1:2108:19795:14268 |
| arthrobacter chlorophenolicus | gi\|219857661\|gb\|CP001341.1\| Arthrobacter chlorophenolicus A6, complete genome | 7.60731e-72 | M01232:27:000000000-ARHCV:1:1102:25720:18317 |
| neorhizobium galegae, | gi\|659653153\|emb\|HG938353.1\| Neorhizobium galegae, complete genome | 5.35883e-49 | M01232:27:000000000-ARHCV:1:2116:22874:6272 |

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|---|---|---|---|
| streptomyces hygroscopicus | gi\|451789617\|gb\|CP003720.1\| Streptomyces hygroscopicus subsp. jinggangensis TL01, complete genome | 3.62659e-50 | M01232:27:000000000-ARHCV:1:2108:22407:2316 |
| pandoraea thiooxydans | gi\|827097060\|gb\|CP011568.1\| Pandoraea thiooxydans strain DSM 25325, complete genome | 4.32226e-20 | M01232:27:000000000-ARHCV:1:2110:27322:10604 |
| geobacter sp. | gi\|320123932\|gb\|CP002479.1\| Geobacter sp. M18, complete genome | 2.29991e-07 | M01232:27:000000000-ARHCV:1:2108:27933:7693 |
| apteryx australis | gi\|840012387\|emb\|LK064788.1\| Apteryx australis mantelli genome assembly AptMant0, scaffold scaffold215 | 0.00437271 | M01232:27:000000000-ARHCV:1:1104:8220:17422 |
| cochliobolus sativus | gi\|628086263\|ref\|XM_007706792.1\| Cochliobolus sativus ND90Pr hypothetical protein partial mRNA | 0.000158791 | M01232:27:000000000-ARHCV:1:1114:18201:16432 |
| geobacter pickeringii | gi\|747129828\|gb\|CP009788.1\| Geobacter pickeringii strain G13, complete genome | 7.03408e-08 | M01232:27:000000000-ARHCV:1:1114:18166:3390 |
| pseudomonas parafulva | gi\|730587126\|gb\|CP009747.1\| Pseudomonas parafulva strain CRS01-1, complete genome | 7.32303e-08 | M01232:27:000000000-ARHCV:1:2112:15712:2825 |
| rhizobium leguminosarum | gi\|574588616\|gb\|CP007067.1\| Rhizobium leguminosarum bv. trifolii CB782, complete genome | 2.03608e-08 | M01232:27:000000000-ARHCV:1:1114:8391:9243 |

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|---|---|---|---|
| acidithiobacillus ferrivorans | gi\|343773687\|gb\|CP002985.1\| Acidithiobacillus ferrivorans SS3, complete genome | 2.60133e-17 | M01232:27:000000000-ARHCV:1:1117:28094:12270 |
| owenweeksia hongkongensis | gi\|359346608\|gb\|CP003156.1\| Owenweeksia hongkongensis DSM 17368, complete genome | 3.96157e-28 | M01232:27:000000000-ARHCV:1:2117:7485:9826 |
| pseudomonas mendocina | gi\|145573243\|gb\|CP000680.1\| Pseudomonas mendocina ymp, complete genome | 0.000234 25 | M01232:27:000000000-ARHCV:1:2117:19191:13515 |
| pseudomonas putida | gi\|169757190\|gb\|CP000949.1\| Pseudomonas putida W619, complete genome | 2.15654e-117 | M01232:27:000000000-ARHCV:1:1108:23055:10421 |
| mesorhizobium australicum | gi\|433663430\|gb\|CP003358.1\| Mesorhizobium australicum WSM2073, complete genome | 3.03717e-86 | M01232:27:000000000-ARHCV:1:2102:13579:18028 |
| sinorhizobium fredii | gi\|227339586\|gb\|CP001389.1\| Sinorhizobium fredii NGR234, complete genome | 8.96721e-47 | M01232:27:000000000-ARHCV:1:2106:28907:17397 |
| isoptericola variabilis | gi\|334105928\|gb\|CP002810.1\| Isoptericola variabilis 225, complete genome | 7.01838e-77 | M01232:27:000000000-ARHCV:1:1113:17600:14187 |
| wenzhouxiangella marina | gi\|906394962\|gb\|CP012154.1\| Wenzhouxiangella marina strain KCTC 42284, complete genome | 5.69087e-32 | M01232:27:000000000-ARHCV:1:1107:9049:23087 |

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|---|---|---|---|
| chelativorans sp. | gi\|110283346\|gb\|CP000390.1\| Chelativorans sp. BNC1, complete genome | 1.20609e-15 | M01232:27:000000000-ARHCV:1:1112:17857:10980 |
| sinorhizobium medicae | gi\|150026743\|gb\|CP000738.1\| Sinorhizobium medicae WSM419, complete genome | 1.55666e-08 | M01232:27:000000000-ARHCV:1:2111:24844:23585 |
| bordetella petrii | gi\|163258032\|emb\|AM902716.1\| Bordetella petrii strain DSM 12804, complete genome | 1.03863e-38 | M01232:27:000000000-ARHCV:1:1108:9612:22875 |
| pseudomonas fluorescens | gi\|253992019\|gb\|CP000094.2\| Pseudomonas fluorescens Pf0-1, complete genome | 8.56481e-77 | M01232:27:000000000-ARHCV:1:1119:14539:20596 |
| scopulariopsis brevicaulis | gi\|670734190\|gb\|KJ443166.1\| Scopulariopsis brevicaulis strain G415 RNA polymerase II subunit 2 (RPB2) gene, partial cds | 3.2485e-36 | M01232:27:000000000-ARHCV:1:1109:5370:14996 |
| paracoccus aminophilus | gi\|529579793\|gb\|CP006650.1\| Paracoccus aminophilus JCM 7686, complete genome | 6.57314e-06 | M01232:27:000000000-ARHCV:1:2119:12803:11498 |
| rhodopseudomonas palustris | gi\|192282182\|gb\|CP001096.1\| Rhodopseudomonas palustris TIE-1, complete genome | 1.17448e-35 | M01232:27:000000000-ARHCV:1:1102:8393:16836 |
| azoarcus sp. | gi\|119668705\|emb\|AM406670.1\| Azoarcus sp. BH72, complete genome | 0.00194404 | M01232:27:000000000-ARHCV:1:1119:12139:7653 |

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|---|---|---|---|
| burkholderia cepacia | gi\|685656837\|gb\|CP007786.1\| Burkholderia cepacia strain DDS 7H-2 chromosome 2, complete sequence | 2.12223e-21 | M01232:27:000000000-ARHCV:1:1101:24263:7250 |
| brevundimonas subvibrioides | gi\|302191744\|gb\|CP002102.1\| Brevundimonas subvibrioides ATCC 15264, complete genome | 2.92096e-10 | M01232:27:000000000-ARHCV:1:1115:11881:16457 |
| mycobacterium haemophilum | gi\|846146862\|gb\|CP011883.1\| Mycobacterium haemophilum DSM 44634 strain ATCC 29548, complete genome | 4.28436e-25 | M01232:27:000000000-ARHCV:1:2113:14647:19536 |
| burkholderia glumae | gi\|779723397\|gb\|CP009435.1\| Burkholderia glumae LMG 2196 = ATCC 33617 chromosome I, complete sequence | 1.95484e-38 | M01232:27:000000000-ARHCV:1:1113:24303:5852 |
| rhizobium etli | gi\|190694918\|gb\|CP001074.1\| Rhizobium etli CIAT 652, complete genome | 1.01367e-35 | M01232:27:000000000-ARHCV:1:1111:14217:8151 |
| neurospora crassa | gi\|758992422\|ref\|XM_952236.2\| Neurospora crassa OR74A DNA repair helicase RAD25 mRNA | 2.25027e-42 | M01232:27:000000000-ARHCV:1:1113:20811:20760 |
| aspergillus clavatus | gi\|121698395\|ref\|XM_001267807.1\| Aspergillus clavatus NRRL 1 conserved hypothetical protein (ACLA_080660), partial mRNA | 4.22763e-08 | M01232:27:000000000-ARHCV:1:1108:28410:12022 |
| lysobacter enzymogenes | gi\|672710209\|emb\|LM994052.1\| Lysobacter enzymogenes partial groEL gene for chaperonin, strain LMG 8762 | 1.06882e-60 | M01232:27:000000000-ARHCV:1:1104:23336:1940 |

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|---|---|---|---|
| deinococcus peraridilitoris | gi\|429128598\|gb\|CP003382.1\| Deinococcus peraridilitoris DSM 19664, complete genome | 0.000571113 | M01232:27:000000000-ARHCV:1:2111:13866:7327 |
| serratia marcescens | gi\|828966150\|gb\|CP011642.1\| Serratia marcescens strain CAV1492, complete genome | 2.62799e-08 | M01232:27:000000000-ARHCV:1:2105:10675:16121 |
| saccharomonospora viridis | gi\|256583961\|gb\|CP001683.1\| Saccharomonospora viridis DSM 43017, complete genome | 1.72147e-118 | M01232:27:000000000-ARHCV:1:1113:25844:11839 |
| saprolegnia diclina | gi\|669145243\|ref\|XM_008609962.1\| Saprolegnia diclina VS20 50S ribosomal protein LP2 mRNA | 0.000764305 | M01232:27:000000000-ARHCV:1:1114:12377:23588 |
| gemmatirosa kalamazoonesis | gi\|575456455\|gb\|CP007128.1\| Gemmatirosa kalamazoonesis strain KBS708, complete genome | 9.35604e-17 | M01232:27:000000000-ARHCV:1:1108:19270:9695 |
| acidovorax sp. | gi\|407894523\|gb\|CP003872.1\| Acidovorax sp. KKS102, complete genome | 2.71856e-09 | M01232:27:000000000-ARHCV:1:1116:11988:3874 |
| nitrobacter hamburgensis | gi\|91798527\|gb\|CP000319.1\| Nitrobacter hamburgensis X14, complete genome | 7.01286e-10 | M01232:27:000000000-ARHCV:1:2113:11859:19396 |
| uncultured soil | gi\|667756517\|gb\|KJ021786.1\| Uncultured soil bacterium clone HN-min40 ammonia monooxygenase subunit A (amoA) gene, partial cds >gi\|667756525\|gb\|KJ021791.1\| Uncultured soil bacterium clone HN-min48 ammonia monooxygenase subunit A (amoA) gene, partial cds | 2.9811e-14 | M01232:27:000000000-ARHCV:1:1103:22228:18255 |

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|---|---|---|---|
| actinobacillus pleuropneumoniae | gi\|189914400\|gb\|CP001091.1\| Actinobacillus pleuropneumoniae serovar 7 str. AP76, complete genome | 7.71016e-13 | M01232:27:000000000-ARHCV:1:2117:8896:9503 |
| blastococcus saxobsidens | gi\|378781357\|emb\|FO117623.1\| Blastococcus saxobsidens DD2 complete genome | 3.41298e-06 | M01232:27:000000000-ARHCV:1:2111:26306:13619 |
| uncultured bacteroidales | gi\|310752645\|gb\|HQ108062.1\| Uncultured Bacteroidales bacterium clone CD5 16S ribosomal RNA gene, partial sequence | 2.77512e-151 | M01232:27:000000000-ARHCV:1:2113:15103:11112 |
| pseudomonas entomophila | gi\|95101722\|emb\|CT573326.1\| Pseudomonas entomophila str. L48 chromosome,complete sequence | 6.44036e-48 | M01232:27:000000000-ARHCV:1:1107:5865:10756 |
| triticum aestivum | gi\|669026884\|emb\|HG670306.1\| Triticum aestivum chromosome 3B, genomic scaffold, cultivar Chinese Spring | 1.42235e-08 | M01232:27:000000000-ARHCV:1:2107:25334:10641 |
| limnochorda pilosa | gi\|921142775\|dbj\|AP014924.1\| Limnochorda pilosa DNA, complete genome, strain: HC45 | 1.77913e-23 | M01232:27:000000000-ARHCV:1:2109:19132:9793 |
| rhodococcus opacus | gi\|226237899\|dbj\|AP011115.1\| Rhodococcus opacus B4 DNA, complete genome | 4.15096e-35 | M01232:27:000000000-ARHCV:1:1118:10877:4647 |
| bacillus cereus | gi\|753605351\|gb\|CP009628.1\| Bacillus cereus ATCC 4342, complete genome | 4.73653e-61 | M01232:27:000000000-ARHCV:1:2111:22736:11434 |

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|---|---|---|---|
| brevibacillus brevis | gi\|226092535\|dbj\|AP008955.1\| Brevibacillus brevis NBRC 100599 DNA, complete genome | 4.15631e-10 | M01232:27:000000000-ARHCV:1:2103:19264:14697 |
| geodermatophilus obscurus | gi\|284061874\|gb\|CP001867.1\| Geodermatophilus obscurus DSM 43160, complete genome | 1.50851e-09 | M01232:27:000000000-ARHCV:1:1119:14971:22504 |
| azospirillum sp. | gi\|288909149\|dbj\|AP010946.1\| Azospirillum sp. B510 DNA, complete genome | 1.0625e-10 | M01232:27:000000000-ARHCV:1:1101:15441:18164 |
| thiobacillus denitrificans | gi\|74055513\|gb\|CP000116.1\| Thiobacillus denitrificans ATCC 25259, complete genome | 2.27194e-62 | M01232:27:000000000-ARHCV:1:2102:26178:22393 |
| predicted: pantholops | gi\|556756331\|ref\|XM_005973454.1\| PREDICTED: Pantholops hodgsonii peroxisomal bifunctional enzyme-like (LOC102339283), mRNA | 1.93135e-17 | M01232:27:000000000-ARHCV:1:1103:15608:22535 |
| corynascella inaequalis | gi\|756809221\|gb\|KP204016.1\| Corynascella inaequalis strain CBS 331.75 RNA polymerase subunit II (RPB2) gene, partial cds | 1.19551e-20 | M01232:27:000000000-ARHCV:1:2105:12417:17187 |
| sphingomonas wittichii | gi\|148498119\|gb\|CP000699.1\| Sphingomonas wittichii RW1, complete genome | 5.31102e-31 | M01232:27:000000000-ARHCV:1:1110:8757:10919 |
| ilumatobacter coccineus | gi\|464097432\|dbj\|AP012057.1\| Ilumatobacter coccineus YM16-304 DNA, complete genome | 1.20609e-15 | M01232:27:000000000-ARHCV:1:1119:23124:10852 |

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|---|---|---|---|
| rhizobium sp. | gi\|584450787\|emb\|HG916852.1\| Rhizobium sp. LPU83 main chrosome complete genome | 1.19328e-25 | M01232:27:000000000-ARHCV:1:2116:11699:10924 |
| sphingobium sp. | gi\|764452294\|gb\|CP010954.1\| Sphingobium sp. YBL2, complete genome | 1.02496e-55 | M01232:27:000000000-ARHCV:1:1118:13732:6618 |
| kocuria palustris | gi\|923033569\|gb\|CP012507.1\| Kocuria palustris strain MU14/1, complete genome | 3.29437e-31 | M01232:27:000000000-ARHCV:1:1105:19924:20183 |
| novosphingobium pentaromativorans | gi\|698178797\|gb\|CP009291.1\| Novosphingobium pentaromativorans US6-1, complete genome | 5.0183e-26 | M01232:27:000000000-ARHCV:1:1114:24035:10572 |
| corallococcus coralloides | gi\|89142843\|emb\|AM183453.1\| Corallococcus coralloides partial rpoB gene for RNA polymerase, beta subunit, strain DSM 52499 | 6.39634e-62 | M01232:27:000000000-ARHCV:1:1104:15250:4414 |
| human chromosome | gi\|14571655\|emb\|AL109769.5\| Human chromosome 14 DNA sequence BAC R-501E21 of library RPCI-11 from chromosome 14 of Homo sapiens (Human), complete sequence | 2.17964e-142 | M01232:27:000000000-ARHCV:1:1109:5260:15068 |
| dinoroseobacter shibae | gi\|157910316\|gb\|CP000830.1\| Dinoroseobacter shibae DFL 12, complete genome | 3.00144e-12 | M01232:27:000000000-ARHCV:1:1104:28668:13026 |
| agrobacterium radiobacter | gi\|221721649\|gb\|CP000628.1\| Agrobacterium radiobacter K84 chromosome 1, complete sequence | 4.33657e-14 | M01232:27:000000000-ARHCV:1:1102:22563:11762 |

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|---|---|---|---|
| hordeum vulgare | gi\|798386805\|gb\|AC256286.1\| Hordeum vulgare clone HV_Mba442-A16, complete sequence | 5.04382e-18 | M01232:27:000000000-ARHCV:1:1102:12061:6892 |
| mycobacterium sinense | gi\|333484608\|gb\|CP002329.1\| Mycobacterium sinense strain JDM601, complete genome | 1.52191e-34 | M01232:27:000000000-ARHCV:1:2107:26629:13977 |
| rhodobacter sphaeroides | gi\|145554299\|gb\|CP000661.1\| Rhodobacter sphaeroides ATCC 17025, complete genome | 1.5358e-05 | M01232:27:000000000-ARHCV:1:2105:26447:15725 |
| phenylobacterium zucineum | gi\|196476886\|gb\|CP000747.1\| Phenylobacterium zucineum HLK1, complete genome | 1.3863e-44 | M01232:27:000000000-ARHCV:1:2102:24454:8719 |
| thermomicrobium roseum | gi\|221155340\|gb\|CP001275.1\| Thermomicrobium roseum DSM 5159, complete genome | 1.78697e-13 | M01232:27:000000000-ARHCV:1:1113:13847:6854 |
| dickeya dadantii | gi\|270342133\|gb\|CP001836.1\| Dickeya dadantii Ech586, complete genome | 3.28559e-07 | M01232:27:000000000-ARHCV:1:1107:8989:9856 |
| stenotrophomonas rhizophila | gi\|627787876\|gb\|CP007597.1\| Stenotrophomonas rhizophila strain DSM14405 genome | 2.03961e-08 | M01232:27:000000000-ARHCV:1:2118:7816:16266 |
| deinococcus gobiensis | gi\|379998737\|gb\|CP002191.1\| Deinococcus gobiensis I-0, complete genome | 4.45351e-14 | M01232:27:000000000-ARHCV:1:1114:14878:16135 |

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|---|---|---|---|
| arthrobacter sp. | gi\|403227528\|gb\|CP003203.1\| Arthrobacter sp. Rue61a, complete genome | 4.52267e-26 | M01232:27:000000000-ARHCV:1:2103:25501:13422 |
| halomonas sp. | gi\|802125597\|emb\|LN813019.1\| Halomonas sp. R57-5 genome assembly HalomonasR57-5, chromosome : I | 1.3279e-94 | M01232:27:000000000-ARHCV:1:1108:18630:24688 |
| pseudoalteromonas sp. | gi\|315013624\|gb\|CP001796.1\| Pseudoalteromonas sp. SM9913 chromosome I, complete sequence | 3.04831e-28 | M01232:27:000000000-ARHCV:1:1118:13384:3871 |
| polaromonas naphthalenivorans | gi\|120591888\|gb\|CP000529.1\| Polaromonas naphthalenivorans CJ2, complete genome | 8.65181e-11 | M01232:27:000000000-ARHCV:1:1112:3049:12349 |
| pseudomonas sp. | gi\|426265132\|gb\|CP003880.1\| Pseudomonas sp. UW4, complete genome | 3.88442e-79 | M01232:27:000000000-ARHCV:1:1111:23245:5571 |
| chromohalobacter salexigens | gi\|91795226\|gb\|CP000285.1\| Chromohalobacter salexigens DSM 3043, complete genome | 4.30847e-09 | M01232:27:000000000-ARHCV:1:1108:22014:15974 |
| nitrobacter vulgaris | gi\|90102361\|gb\|DQ421377.1\| Nitrobacter vulgaris clone 2 nitrite oxidoreductase alpha subunit (norA) gene, partial cds | 2.19511e-137 | M01232:27:000000000-ARHCV:1:2104:13264:15205 |
| streptomyces rapamycinicus | gi\|521353217\|gb\|CP006567.1\| Streptomyces rapamycinicus NRRL 5491 genome | 1.20819e-15 | M01232:27:000000000-ARHCV:1:2105:22915:23439 |

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|---|---|---|---|
| clavibacter michiganensis | gi\|472820487\|emb\|HE614873.1\| Clavibacter michiganensis subsp. nebraskensis NCPPB 2581 complete genome | 4.49953e-19 | M01232:27:000000000-ARHCV:1:1116:22337:20582 |
| uncultured nitrosospira | gi\|283444729\|gb\|GU136442.1\| Uncultured Nitrosospira sp. clone 1-24 ammonia monooxygenase subunit A-like (amoA) gene, partial sequence | 8.97755e-10 | M01232:27:000000000-ARHCV:1:1104:20117:5078 |
| rasamsonia emersonii | gi\|915175950\|ref\|XM_013477024.1\| Rasamsonia emersonii CBS 393.64 hypothetical protein mRNA | 1.54918e-19 | M01232:27:000000000-ARHCV:1:2101:17353:22444 |
| halobacterium salinarum | gi\|167726115\|emb\|AM774415.1\| Halobacterium salinarum R1 complete genome | 5.59119e-19 | M01232:27:000000000-ARHCV:1:2103:20471:17362 |
| pseudomonas syringae | gi\|574595116\|gb\|CP007014.1\| Pseudomonas syringae CC1557, complete sequence | 7.92055e-36 | M01232:27:000000000-ARHCV:1:1105:25084:19959 |

## APPENDIX 1 (C): Metagenomic BLAST results for the Compost inoculum source: nxrA gene

| Organism/HIT | Accession | e-value | Fastq header |
|---|---|---|---|
| No hits | None | 0 | M01232:27:000000000-ARHCV:1:1107:15446:15413 |
| uncultured bacterium | gi\|90102335\|gb\|DQ421364.1\| Uncultured bacterium clone B6 nitrite oxidoreductase alpha subunit (norA) gene, partial cds | 1.14345e-47 | M01232:27:000000000-ARHCV:1:1113:5261:10982 |
| nitrobacter winogradskyi | gi\|661349927\|gb\|KJ023581.1\| Nitrobacter winogradskyi isolate DGGE gel band nit-5 NorA (norA) gene, partial cds | 5.01956e-94 | M01232:27:000000000-ARHCV:1:1108:12520:14138 |

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|---|---|---|---|
| uncultured nitrite-oxidizing | gi\|724471095\|gb\|KM408671.1\| Uncultured nitrite-oxidizing bacterium clone IB2-2 nitrite oxidoreductase alpha subunit (nxrA) gene, partial cds | 6.55065e-88 | M01232:27:000000000-ARHCV:1:1111:15494:23953 |
| nitrobacter vulgaris | gi\|90102361\|gb\|DQ421377.1\| Nitrobacter vulgaris clone 2 nitrite oxidoreductase alpha subunit (norA) gene, partial cds | 1.32112e-134 | M01232:27:000000000-ARHCV:1:2119:14373:11213 |
| uncultured nitrobacter | gi\|395486130\|gb\|JX020944.1\| Uncultured Nitrobacter sp. isolate DGGE gel band 12 nitrite oxidoreductase alpha subunit (norA) gene, partial cds | 1.34946e-119 | M01232:27:000000000-ARHCV:1:2112:21862:18418 |
| nitrobacter hamburgensis | gi\|661349921\|gb\|KJ023578.1\| Nitrobacter hamburgensis isolate DGGE gel band nit-2 NorA (norA) gene, partial cds | 3.77863e-115 | M01232:27:000000000-ARHCV:1:2118:23326:1876 |
| nitrobacter alkalicus | gi\|661349929\|gb\|KJ023582.1\| Nitrobacter alkalicus isolate DGGE gel band nit-6 NorA (norA) gene, partial cds | 7.81225e-142 | M01232:27:000000000-ARHCV:1:2102:24019:8264 |
| cellulomonas flavigena | gi\|296019684\|gb\|CP001964.1\| Cellulomonas flavigena DSM 20109, complete genome | 5.24629e-64 | M01232:27:000000000-ARHCV:1:2117:22661:18027 |
| sphingopyxis fribergensis | gi\|734569737\|gb\|CP009122.1\| Sphingopyxis fribergensis strain Kp5.2, complete genome | 4.34773e-09 | M01232:27:000000000-ARHCV:1:1114:16521:5695 |
| castellaniella defragrans | gi\|589264544\|emb\|HG916765.1\| Castellaniella defragrans 65Phen complete genome | 1.8736e-68 | M01232:27:000000000-ARHCV:1:1110:14504:4795 |
| unidentified bacterium | gi\|106733792\|gb\|DQ514276.1\| Unidentified bacterium clone IGe2 nitrite oxidoreductase alpha subunit (nxrA) gene, partial cds | 2.35196e-87 | M01232:27:000000000-ARHCV:1:1107:15524:13781 |
| mycobacterium sp. | gi\|119692146\|gb\|CP000518.1\| Mycobacterium sp. KMS, complete genome | 2.43242e-62 | M01232:27:000000000-ARHCV:1:2119:16956:17091 |
| pseudomonas aeruginosa | gi\|926451317\|emb\|LN871187.1\| Pseudomonas aeruginosa genome assembly PAO1OR, chromosome : I | 8.37354e-48 | M01232:27:000000000-ARHCV:1:1117:21338:20503 |
| intrasporangium calvum | gi\|315587265\|gb\|CP002343.1\| Intrasporangium calvum DSM 43043, complete genome | 6.50447e-93 | M01232:27:000000000-ARHCV:1:1118:21590:9661 |

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| hoeflea sp. | gi\|822663067\|gb\|CP011479.1\| Hoeflea sp. IMCC20628, complete genome | 1.9827e-28 | M01232:27:000000000-ARHCV:1:2111:13930:24109 |
| streptomyces albus | gi\|749174630\|gb\|CP010519.1\| Streptomyces albus strain DSM 41398, complete genome | 2.58299e-22 | M01232:27:000000000-ARHCV:1:2117:15395:12616 |
| methylobacterium aquaticum | gi\|760865928\|dbj\|AP014704.1\| Methylobacterium aquaticum DNA, complete genome, strain: MA-22A | 8.8682e-12 | M01232:27:000000000-ARHCV:1:2106:15898:16461 |
| cyprinus carpio | gi\|685042180\|emb\|LN590705.1\| Cyprinus carpio genome assembly common carp genome ,scaffold LG11 | 1.90884e-17 | M01232:27:000000000-ARHCV:1:2117:23497:15288 |
| methylibium petroleiphilum | gi\|124257968\|gb\|CP000555.1\| Methylibium petroleiphilum PM1, complete genome | 1.86763e-57 | M01232:27:000000000-ARHCV:1:1103:4451:10482 |
| rhodopseudomonas palustris | gi\|86570155\|gb\|CP000250.1\| Rhodopseudomonas palustris HaA2, complete genome | 3.30511e-50 | M01232:27:000000000-ARHCV:1:1102:26416:11236 |
| infectious bronchitis | gi\|386370460\|gb\|JQ088078.1\| Infectious bronchitis virus strain CK/SWE/0658946/10, complete genome | 7.75717e-147 | M01232:27:000000000-ARHCV:1:1109:16426:11213 |
| azospirillum lipoferum | gi\|357422594\|emb\|FQ311868.1\| Azospirillum lipoferum 4B main chromosome, complete genome | 5.71616e-53 | M01232:27:000000000-ARHCV:1:1111:11507:19514 |
| streptomyces davawensis | gi\|408526205\|emb\|HE971709.1\| Streptomyces davawensis strain JCM 4913 complete genome | 8.65556e-72 | M01232:27:000000000-ARHCV:1:1106:4440:14560 |
| anaeromyxobacter sp. | gi\|196170270\|gb\|CP001131.1\| Anaeromyxobacter sp. K, complete genome | 4.80904e-17 | M01232:27:000000000-ARHCV:1:2102:9197:15364 |
| novosphingobium pentaromativorans | gi\|698178797\|gb\|CP009291.1\| Novosphingobium pentaromativorans US6-1, complete genome | 8.17409e-67 | M01232:27:000000000-ARHCV:1:1119:25823:4640 |
| microlunatus phosphovorus | gi\|334683429\|dbj\|AP012204.1\| Microlunatus phosphovorus NM-1 DNA, complete genome | 1.97927e-28 | M01232:27:000000000-ARHCV:1:1105:20999:22583 |
| streptomyces sp. | gi\|822591927\|gb\|CP011492.1\| Streptomyces sp. CNQ-509, complete genome | 5.24629e-64 | M01232:27:000000000-ARHCV:1:1106:26468:9420 |
| azorhizobium caulinodans | gi\|158328513\|dbj\|AP009384.1\| Azorhizobium caulinodans ORS 571 DNA, complete genome | 8.34029e-97 | M01232:27:000000000-ARHCV:1:1106:13081:14267 |
| anaeromyxobacter dehalogenans | gi\|219952977\|gb\|CP001359.1\| Anaeromyxobacter dehalogenans 2CP-1, complete genome | 7.02244e-32 | M01232:27:000000000-ARHCV:1:2105:25247:7109 |

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|---|---|---|---|
| pseudonocardia dioxanivorans | gi\|326948588\|gb\|CP002593.1\| Pseudonocardia dioxanivorans CB1190, complete genome | 1.8869e-63 | M01232:27:000000000-ARHCV:1:1104:15225:9267 |
| kocuria palustris | gi\|923033569\|gb\|CP012507.1\| Kocuria palustris strain MU14/1, complete genome | 4.35294e-15 | M01232:27:000000000-ARHCV:1:2116:25429:11441 |
| apteryx australis | gi\|841904503\|emb\|LK064812.1\| Apteryx australis mantelli genome assembly AptMant0, scaffold scaffold95 | 1.08102e-80 | M01232:27:000000000-ARHCV:1:1113:15899:15449 |
| nippostrongylus brasiliensis | gi\|687008631\|emb\|LM437148.1\| Nippostrongylus brasiliensis genome assembly N_brasiliensis_RM07_v1_5_4 ,scaffold NBR_scaffold0003529 | 9.64487e-06 | M01232:27:000000000-ARHCV:1:1116:26625:6160 |
| arthrobacter phenanthrenivorans | gi\|323467537\|gb\|CP002379.1\| Arthrobacter phenanthrenivorans Sphe3, complete genome | 1.05271e-100 | M01232:27:000000000-ARHCV:1:1109:11195:8825 |
| micromonospora sp. | gi\|315407437\|gb\|CP002399.1\| Micromonospora sp. L5, complete genome | 1.92738e-48 | M01232:27:000000000-ARHCV:1:2102:26237:17434 |
| nitrospira moscoviensis | gi\|922305621\|gb\|CP011801.1\| Nitrospira moscoviensis strain NSP M-1, complete genome | 7.23539e-11 | M01232:27:000000000-ARHCV:1:1118:8290:10096 |
| streptomyces cyaneogriseus | gi\|758855033\|gb\|CP010849.1\| Streptomyces cyaneogriseus subsp. noncyanogenus strain NMWT 1, complete genome | 6.88321e-53 | M01232:27:000000000-ARHCV:1:2110:17456:3092 |
| limnochorda pilosa | gi\|921142775\|dbj\|AP014924.1\| Limnochorda pilosa DNA, complete genome, strain: HC45 | 1.74192e-11 | M01232:27:000000000-ARHCV:1:1106:12046:3455 |
| blastochloris viridis | gi\|891154826\|dbj\|AP014854.1\| Blastochloris viridis DNA, complete genome, strain: DSM 133 | 1.15532e-05 | M01232:27:000000000-ARHCV:1:2102:11135:11944 |
| isoptericola variabilis | gi\|334105928\|gb\|CP002810.1\| Isoptericola variabilis 225, complete genome | 6.52789e-13 | M01232:27:000000000-ARHCV:1:2110:12924:15547 |
| methylobacterium populi | gi\|179342784\|gb\|CP001029.1\| Methylobacterium populi BJ001, complete genome | 4.73746e-06 | M01232:27:000000000-ARHCV:1:2108:17153:18600 |
| azoarcus aromaticum | gi\|56311475\|emb\|CR555306.1\| Azoarcus aromaticum EbN1 complete genome | 7.20843e-07 | M01232:27:000000000-ARHCV:1:2105:12843:13035 |
| variovorax paradoxus | gi\|239799596\|gb\|CP001635.1\| Variovorax paradoxus S110 chromosome 1, complete sequence | 1.21676e-10 | M01232:27:000000000-ARHCV:1:2113:18505:6122 |

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| mycobacterium gilvum | gi\|315265130\|gb\|CP002386.1\| Mycobacterium gilvum Spyr1 plasmid pMSPYR101, complete sequence | 6.13593e-133 | M01232:27:000000000-ARHCV:1:1112:18370:6781 |
| polymorphum gilvum | gi\|326411376\|gb\|CP002568.1\| Polymorphum gilvum SL003B-26A1, complete genome | 9.15865e-41 | M01232:27:000000000-ARHCV:1:1101:11035:11662 |
| cyanobacterium aponinum | gi\|428682694\|gb\|CP003947.1\| Cyanobacterium aponinum PCC 10605, complete genome | 3.36433e-08 | M01232:27:000000000-ARHCV:1:1118:16571:9915 |
| ruegeria pomeroyi | gi\|564474190\|gb\|CP000031.2\| Ruegeria pomeroyi DSS-3, complete genome | 6.63102e-43 | M01232:27:000000000-ARHCV:1:2119:16678:21860 |
| haloterrigena turkmenica | gi\|284012950\|gb\|CP001860.1\| Haloterrigena turkmenica DSM 5511, complete genome | 0.00201144 | M01232:27:000000000-ARHCV:1:1116:15009:24747 |
| xylanimonas cellulosilytica | gi\|269303491\|gb\|CP001821.1\| Xylanimonas cellulosilytica DSM 15894, complete genome | 1.18486e-30 | M01232:27:000000000-ARHCV:1:2105:12010:22868 |
| uncultured ammonia-oxidizing | gi\|924899145\|gb\|KP987174.1\| Uncultured ammonia-oxidizing bacterium clone AOB-16-1-S27 ammonia monooxygenase subunit A (amoA) gene, partial cds | 1.29337e-149 | M01232:27:000000000-ARHCV:1:2114:10936:6877 |
| planctomyces brasiliensis | gi\|324966854\|gb\|CP002546.1\| Planctomyces brasiliensis DSM 5305, complete genome | 2.62847e-06 | M01232:27:000000000-ARHCV:1:1118:24904:15624 |
| solibacter usitatus | gi\|116222307\|gb\|CP000473.1\| Solibacter usitatus Ellin6076, complete genome | 1.10768e-15 | M01232:27:000000000-ARHCV:1:1101:15585:24305 |
| rhodococcus aetherivorans | gi\|816214082\|gb\|CP011341.1\| Rhodococcus aetherivorans strain IcdP1, complete genome | 5.65124e-09 | M01232:27:000000000-ARHCV:1:1110:20424:19814 |
| influenza a | gi\|78097833\|gb\|CY005740.1\| Influenza A virus (A/duck/NZL/164/1976(H11N3)) segment 7, complete sequence | 4.66051e-144 | M01232:27:000000000-ARHCV:1:1111:26124:4772 |
| dermacoccus nishinomiyaensis | gi\|664687059\|gb\|CP008889.1\| Dermacoccus nishinomiyaensis strain M25, complete genome | 3.00817e-25 | M01232:27:000000000-ARHCV:1:2102:21399:7056 |
| methylobacterium nodulans | gi\|219944660\|gb\|CP001349.1\| Methylobacterium nodulans ORS 2060, complete genome | 2.33848e-27 | M01232:27:000000000-ARHCV:1:2111:20209:8974 |
| halomicrobium mukohataei | gi\|257168392\|gb\|CP001688.1\| Halomicrobium mukohataei DSM 12286, complete genome | 1.20175e-20 | M01232:27:000000000-ARHCV:1:1117:23477:13428 |

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| streptomyces avermitilis | gi\|148878541\|dbj\|BA000030.3\| Streptomyces avermitilis MA-4680 = NBRC 14893 DNA, complete genome | 2.6198e-12 | M01232:27:000000000-ARHCV:1:2114:16896:8042 |
| methylobacterium radiotolerans | gi\|170652972\|gb\|CP001001.1\| Methylobacterium radiotolerans JCM 2831, complete genome | 3.16517e-11 | M01232:27:000000000-ARHCV:1:1117:24466:10310 |
| opitutus terrae | gi\|177839040\|gb\|CP001032.1\| Opitutus terrae PB90-1, complete genome | 1.97927e-28 | M01232:27:000000000-ARHCV:1:1111:18095:24194 |
| azotobacter vinelandii | gi\|482534342\|gb\|CP005095.1\| Azotobacter vinelandii CA6, complete genome | 4.11342e-55 | M01232:27:000000000-ARHCV:1:2111:25089:23554 |
| hymenobacter sp. | gi\|926465561\|gb\|CP012623.1\| Hymenobacter sp. DG25A, complete genome | 0.000542458 | M01232:27:000000000-ARHCV:1:1104:15347:16967 |
| rubrobacter xylanophilus | gi\|108764099\|gb\|CP000386.1\| Rubrobacter xylanophilus DSM 9941, complete genome | 1.5436e-24 | M01232:27:000000000-ARHCV:1:1105:12719:24051 |
| burkholderia vietnamiensis | gi\|773006514\|gb\|CP009632.1\| Burkholderia vietnamiensis LMG 10929 chromosome III, complete sequence | 2.02172e-13 | M01232:27:000000000-ARHCV:1:2111:23019:14802 |
| rhizobium etli | gi\|647790312\|gb\|CP006986.1\| Rhizobium etli bv. mimosae str. IE4771, complete genome | 1.35659e-54 | M01232:27:000000000-ARHCV:1:2111:28702:16645 |
| streptomyces lividans | gi\|672367150\|gb\|CP009124.1\| Streptomyces lividans TK24, complete genome | 1.14171e-55 | M01232:27:000000000-ARHCV:1:1112:13230:24994 |
| thioflavicoccus mobilis | gi\|431827765\|gb\|CP003051.1\| Thioflavicoccus mobilis 8321, complete genome | 4.82178e-24 | M01232:27:000000000-ARHCV:1:1101:24372:8043 |
| singulisphaera acidiphila | gi\|430012750\|gb\|CP003364.1\| Singulisphaera acidiphila DSM 18658, complete genome | 2.28593e-08 | M01232:27:000000000-ARHCV:1:1106:13963:4439 |
| marichromatium purpuratum | gi\|570731247\|gb\|CP007031.1\| Marichromatium purpuratum 984, complete genome | 8.1303e-08 | M01232:27:000000000-ARHCV:1:2115:8961:17576 |
| nitrosospira sp. | gi\|81251149\|gb\|DQ228464.1\| Nitrosospira sp. NIJS18 ammonia monooxygenase subunit A (amoA) gene, partial cds | 7.97982e-127 | M01232:27:000000000-ARHCV:1:2110:13140:12187 |
| terriglobus roseus | gi\|390410848\|gb\|CP003379.1\| Terriglobus roseus DSM 18391, complete genome | 1.9827e-28 | M01232:27:000000000-ARHCV:1:1109:13017:21755 |
| rhodopirellula baltica | gi\|32448346\|emb\|BX294156.1\| Rhodopirellula baltica SH 1 complete genome | 1.41547e-08 | M01232:27:000000000-ARHCV:1:2102:16153:9172 |

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| oligotropha carboxidovorans | gi\|336096911\|gb\|CP002826.1\| Oligotropha carboxidovorans OM5, complete genome | 1.41306e-84 | M01232:27:000000000-ARHCV:1:2109:13108:9667 |
| sinorhizobium fredii | gi\|365177649\|emb\|HE616890.1\| Sinorhizobium fredii HH103 main chromosome, complete sequence | 3.36503e-16 | M01232:27:000000000-ARHCV:1:2106:24596:22085 |
| chondromyces crocatus | gi\|908353314\|gb\|CP012159.1\| Chondromyces crocatus strain Cm c5, complete genome | 2.67736e-21 | M01232:27:000000000-ARHCV:1:1114:13298:12881 |
| sphingomonas wittichii | gi\|148498119\|gb\|CP000699.1\| Sphingomonas wittichii RW1, complete genome | 6.99388e-22 | M01232:27:000000000-ARHCV:1:1106:17452:21329 |
| nocardiopsis alba | gi\|402798256\|gb\|CP003788.1\| Nocardiopsis alba ATCC BAA-2165, complete genome | 1.95865e-13 | M01232:27:000000000-ARHCV:1:1105:26618:14077 |
| predicted: tupaia | gi\|562849428\|ref\|XM_006153441.1\| PREDICTED: Tupaia chinensis pyruvate kinase, liver and RBC (PKLR), partial mRNA | 1.20204e-05 | M01232:27:000000000-ARHCV:1:2105:20052:22264 |
| guinea fowl | gi\|807045770\|emb\|LN610099.1\| Guinea fowl coronavirus GfCoV/FR/2011 complete genome | 9.45176e-116 | M01232:27:000000000-ARHCV:1:2117:25858:16086 |
| burkholderia fungorum | gi\|780551828\|gb\|CP010025.1\| Burkholderia fungorum strain ATCC BAA-463 chromosome 3, complete sequence | 2.02172e-13 | M01232:27:000000000-ARHCV:1:1105:16430:9192 |
| agrobacterium radiobacter | gi\|221725460\|gb\|CP000629.1\| Agrobacterium radiobacter K84 chromosome 2, complete sequence | 2.71031e-13 | M01232:27:000000000-ARHCV:1:2112:13215:20706 |
| phycisphaera mikurensis | gi\|381385343\|dbj\|AP012338.1\| Phycisphaera mikurensis NBRC 102666 DNA, complete genome | 1.4976e-09 | M01232:27:000000000-ARHCV:1:1110:19211:24058 |
| pirellula staleyi | gi\|283436255\|gb\|CP001848.1\| Pirellula staleyi DSM 6068, complete genome | 1.99677e-23 | M01232:27:000000000-ARHCV:1:1102:17504:2239 |
| natronomonas moolapensis | gi\|452081962\|emb\|HF582854.1\| Natronomonas moolapensis 8.8.11 complete genome | 0.000424022 | M01232:27:000000000-ARHCV:1:2108:6302:14383 |
| predicted: camelus | gi\|744552351\|ref\|XM_010975897.1\| PREDICTED: Camelus dromedarius seryl-tRNA synthetase (SARS), mRNA | 0.00194747 | M01232:27:000000000-ARHCV:1:1101:23275:18797 |
| dechloromonas aromatica | gi\|71845263\|gb\|CP000089.1\| Dechloromonas aromatica RCB, complete genome | 2.38285e-27 | M01232:27:000000000-ARHCV:1:2112:13224:3973 |

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| rhodovulum sulfidophilum | gi\|770474292\|dbj\|AP014800.1\| Rhodovulum sulfidophilum DNA, complete genome, strain: DSM 2351 | 8.2239e-82 | M01232:27:000000000-ARHCV:1:1111:18577:25000 |
| bradyrhizobium sp. | gi\|146189981\|emb\|CU234118.1\| Bradyrhizobium sp. ORS278,complete sequence | 8.86755e-14 | M01232:27:000000000-ARHCV:1:2104:10256:21096 |
| devosia sp. | gi\|901895891\|gb\|CP011300.1\| Devosia sp. H5989, complete genome | 1.13775e-55 | M01232:27:000000000-ARHCV:1:2118:15292:7555 |
| frankia sp. | gi\|158107272\|gb\|CP000820.1\| Frankia sp. EAN1pec, complete genome | 1.54175e-18 | M01232:27:000000000-ARHCV:1:1104:18511:11536 |
| gloeobacter violaceus | gi\|37508091\|dbj\|BA000045.2\| Gloeobacter violaceus PCC 7421 DNA, complete genome | 7.99479e-11 | M01232:27:000000000-ARHCV:1:1104:27625:16023 |
| marinobacter adhaerens | gi\|311692891\|gb\|CP001978.1\| Marinobacter adhaerens HP15, complete genome | 0.000571113 | M01232:27:000000000-ARHCV:1:2103:27358:12330 |
| pseudoxanthomonas suwonensis | gi\|317464132\|gb\|CP002446.1\| Pseudoxanthomonas suwonensis 11-1, complete genome | 5.69934e-27 | M01232:27:000000000-ARHCV:1:2113:23301:19874 |
| alistipes finegoldii | gi\|390421916\|gb\|CP003274.1\| Alistipes finegoldii DSM 17242, complete genome | 5.67086e-09 | M01232:27:000000000-ARHCV:1:2109:25781:12021 |
| propionibacterium acnes | gi\|657118275\|gb\|CP006032.1\| Propionibacterium acnes hdn-1, complete genome | 4.306e-20 | M01232:27:000000000-ARHCV:1:2109:18900:14844 |
| pseudomonas protegens | gi\|500239649\|gb\|CP003190.1\| Pseudomonas protegens CHA0, complete genome | 1.20175e-20 | M01232:27:000000000-ARHCV:1:2107:18729:12446 |
| acidiphilium cryptum | gi\|146400702\|gb\|CP000697.1\| Acidiphilium cryptum JF-5, complete genome | 1.42784e-44 | M01232:27:000000000-ARHCV:1:1107:19128:23794 |
| methylobacterium extorquens | gi\|240006747\|gb\|CP001510.1\| Methylobacterium extorquens AM1, complete genome | 9.42247e-12 | M01232:27:000000000-ARHCV:1:1110:11059:15446 |
| mycobacterium haemophilum | gi\|846146862\|gb\|CP011883.1\| Mycobacterium haemophilum DSM 44634 strain ATCC 29548, complete genome | 2.02172e-13 | M01232:27:000000000-ARHCV:1:2115:22892:7688 |
| sphingomonas sp. | gi\|761896199\|gb\|CP010836.1\| Sphingomonas sp. WHSC-8, complete genome | 8.904e-52 | M01232:27:000000000-ARHCV:1:1108:10213:3630 |
| streptomyces lydicus | gi\|768311912\|gb\|CP007699.1\| Streptomyces lydicus A02, complete genome | 5.63089e-14 | M01232:27:000000000-ARHCV:1:1105:25363:4240 |

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| hyphomicrobium sp. | gi\|337757426\|emb\|FQ859181.1\| Hyphomicrobium sp. MC1 chromosome, complete genome | 8.16514e-112 | M01232:27:000000000-ARHCV:1:1117:9289:14863 |
| rhodococcus erythropolis | gi\|532219856\|gb\|CP003761.1\| Rhodococcus erythropolis CCM2595, complete genome | 1.02854e-130 | M01232:27:000000000-ARHCV:1:2105:22181:20471 |
| ramlibacter tataouinensis | gi\|334728683\|gb\|CP000245.1\| Ramlibacter tataouinensis TTB310, complete genome | 0.00297907 | M01232:27:000000000-ARHCV:1:2117:8965:14542 |
| kineococcus radiotolerans | gi\|196121877\|gb\|CP000750.2\| Kineococcus radiotolerans SRS30216, complete genome | 1.29813e-33 | M01232:27:000000000-ARHCV:1:1105:25805:17906 |
| burkholderia multivorans | gi\|189332915\|dbj\|AP009385.1\| Burkholderia multivorans ATCC 17616 DNA, complete genome, chromosome 1 | 2.60133e-17 | M01232:27:000000000-ARHCV:1:2101:16056:11207 |
| streptomyces cattleya | gi\|365804155\|gb\|CP003219.1\| Streptomyces cattleya DSM 46488, complete genome | 8.58639e-07 | M01232:27:000000000-ARHCV:1:1104:11875:9675 |
| deinococcus peraridilitoris | gi\|429128598\|gb\|CP003382.1\| Deinococcus peraridilitoris DSM 19664, complete genome | 5.55178e-24 | M01232:27:000000000-ARHCV:1:2119:6825:11045 |
| acetobacter liquefaciens | gi\|893352\|dbj\|D28511.1\|ABCLSD Acetobacter liquefaciens gene for membrane-bound L-sorbosone dehydrogenase (SNDH), complete cds | 5.67086e-09 | M01232:27:000000000-ARHCV:1:2105:19285:20998 |
| nocardiopsis dassonvillei | gi\|296848233\|gb\|CP002041.1\| Nocardiopsis dassonvillei subsp. dassonvillei DSM 43111 chromosome 2, complete sequence | 9.2431e-07 | M01232:27:000000000-ARHCV:1:1102:5583:16553 |
| pseudomonas brassicacearum | gi\|591390487\|gb\|CP007410.1\| Pseudomonas brassicacearum strain DF41, complete genome | 1.33752e-12 | M01232:27:000000000-ARHCV:1:1118:17447:4754 |
| amycolatopsis mediterranei | gi\|532225686\|gb\|CP003777.1\| Amycolatopsis mediterranei RB, complete genome | 1.93874e-37 | M01232:27:000000000-ARHCV:1:1103:10522:19528 |
| mycobacterium tuberculosis | gi\|923105817\|gb\|CP012506.1\| Mycobacterium tuberculosis strain SCAID 187.0, complete genome | 4.35294e-15 | M01232:27:000000000-ARHCV:1:1113:8035:7160 |
| endosymbiont of | gi\|530669580\|dbj\|AP012978.1\| Endosymbiont of unidentified scaly snail isolate Monju DNA, complete genome | 1.22753e-05 | M01232:27:000000000-ARHCV:1:2110:23149:19933 |
| g.gallus b-creatine | gi\|211231\|gb\|M33713.1\|CHKBABA3 G.gallus B-creatine kinase (B-CK) gene, exons 6 and 7 | 3.19369e-94 | M01232:27:000000000-ARHCV:1:1103:24573:17109 |

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| nocardioides sp. | gi\|119534933\|gb\|CP000509.1\| Nocardioides sp. JS614, complete genome | 0.00466679 | M01232:27:000000000-ARHCV:1:1118:26158:7135 |
| thioalkalivibrio paradoxus | gi\|570725612\|gb\|CP007029.1\| Thioalkalivibrio paradoxus ARh 1, complete genome | 1.9377e-43 | M01232:27:000000000-ARHCV:1:2106:5767:13724 |
| hyphomicrobium denitrificans | gi\|299523359\|gb\|CP002083.1\| Hyphomicrobium denitrificans ATCC 51888, complete genome | 7.08078e-33 | M01232:27:000000000-ARHCV:1:1114:28334:18780 |
| candidatus symbiobacter | gi\|550800168\|gb\|CP004885.1\| Candidatus Symbiobacter mobilis CR, complete genome | 3.36503e-16 | M01232:27:000000000-ARHCV:1:1113:14018:23631 |
| triticum aestivum | gi\|669026884\|emb\|HG670306.1\| Triticum aestivum chromosome 3B, genomic scaffold, cultivar Chinese Spring | 2.98754e-05 | M01232:27:000000000-ARHCV:1:1103:17932:7105 |
| croceicoccus naphthovorans | gi\|831206920\|gb\|CP011770.1\| Croceicoccus naphthovorans strain PQ-2, complete genome | 1.5656e-14 | M01232:27:000000000-ARHCV:1:1108:10138:15282 |
| haliangium ochraceum | gi\|262076673\|gb\|CP001804.1\| Haliangium ochraceum DSM 14365, complete genome | 3.40117e-06 | M01232:27:000000000-ARHCV:1:2105:14184:7975 |
| mesorhizobium loti | gi\|474421396\|dbj\|AP012557.1\| Mesorhizobium loti DNA, symbiosis island, strain: NZP2037 | 2.15675e-72 | M01232:27:000000000-ARHCV:1:1101:11552:11748 |
| methylocystis sp. | gi\|401772585\|emb\|HE956757.1\| Methylocystis sp. SC2 complete genome | 9.29009e-22 | M01232:27:000000000-ARHCV:1:1107:11534:22180 |
| edwardsiella ictaluri | gi\|409033099\|gb\|CP001600.2\| Edwardsiella ictaluri 93-146, complete genome | 4.83063e-11 | M01232:27:000000000-ARHCV:1:1117:5847:17507 |
| tsukamurella paurometabola | gi\|296025884\|gb\|CP001966.1\| Tsukamurella paurometabola DSM 20162, complete genome | 4.33859e-05 | M01232:27:000000000-ARHCV:1:1105:19434:2932 |
| starkeya novella | gi\|296926528\|gb\|CP002026.1\| Starkeya novella DSM 506, complete genome | 4.1722e-15 | M01232:27:000000000-ARHCV:1:1114:21790:1270 |
| heligmosomoides polygyrus | gi\|688443769\|emb\|LL194751.1\| Heligmosomoides polygyrus genome assembly H_bakeri_Edinburgh ,scaffold HPBE_scaffold0006271 | 6.73334e-07 | M01232:27:000000000-ARHCV:1:2113:2509:16128 |
| sphingobium chlorophenolicum | gi\|334100279\|gb\|CP002798.1\| Sphingobium chlorophenolicum L-1 chromosome 1, complete sequence | 4.9761e-07 | M01232:27:000000000-ARHCV:1:2101:9119:9297 |
| ralstonia pickettii | gi\|546336137\|gb\|CP006667.1\| Ralstonia pickettii DTP0602 chromosome 1, complete sequence | 6.3492e-23 | M01232:27:000000000-ARHCV:1:2113:25024:17584 |

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|---|---|---|---|
| ricinus communis | gi\|255589234\|ref\|XM_002534842.1\| Ricinus communis conserved hypothetical protein, mRNA | 4.12597e-10 | M01232:27:000000000-ARHCV:1:1102:10943:16512 |
| african horse | gi\|871044370\|gb\|KT030336.1\| African horse sickness virus strain Labstr/ZAF/1998/OBP-116 serotype 1 VP7 gene, complete cds | 9.67168e-42 | M01232:27:000000000-ARHCV:1:1119:27143:21133 |
| bacteroides salanitronis | gi\|324316725\|gb\|CP002530.1\| Bacteroides salanitronis DSM 18170, complete genome | 9.48936e-07 | M01232:27:000000000-ARHCV:1:2112:18091:4716 |
| stenotrophomonas maltophilia | gi\|922679809\|gb\|CP011010.1\| Stenotrophomonas maltophilia strain ISMMS3, complete genome | 1.99677e-23 | M01232:27:000000000-ARHCV:1:1112:17044:4518 |
| uncultured rumen | gi\|896685151\|gb\|KR068291.1\| Uncultured rumen bacterium clone YAK-F101 16S ribosomal RNA gene, partial sequence | 6.06021e-143 | M01232:27:000000000-ARHCV:1:2118:15911:21196 |
| ilumatobacter coccineus | gi\|464097432\|dbj\|AP012057.1\| Ilumatobacter coccineus YM16-304 DNA, complete genome | 2.71957e-15 | M01232:27:000000000-ARHCV:1:2117:16585:17652 |
| rhizobium sp. | gi\|430001947\|emb\|FO082820.1\| Rhizobium sp. str. NT-26 chromosome, complete genome | 2.78502e-40 | M01232:27:000000000-ARHCV:1:1117:22631:10345 |
| pseudomonas cichorii | gi\|572998810\|gb\|CP007039.1\| Pseudomonas cichorii JBC1, complete genome | 1.05602e-09 | M01232:27:000000000-ARHCV:1:1107:26384:11997 |
| amycolatopsis japonica | gi\|667678325\|gb\|CP008953.1\| Amycolatopsis japonica strain MG417-CF17, complete genome | 1.44032e-09 | M01232:27:000000000-ARHCV:1:1107:17800:21666 |
| bifidobacterium longum | gi\|665999851\|gb\|CP008885.1\| Bifidobacterium longum strain BXY01, complete genome | 6.18067e-11 | M01232:27:000000000-ARHCV:1:1114:9083:23774 |
| corynebacterium humireducens | gi\|748228406\|gb\|CP005286.1\| Corynebacterium humireducens NBRC 106098 = DSM 45392, complete genome | 2.58299e-22 | M01232:27:000000000-ARHCV:1:2104:9704:16136 |
| corallococcus coralloides | gi\|380727201\|gb\|CP003389.1\| Corallococcus coralloides DSM 2259, complete genome | 3.1352e-66 | M01232:27:000000000-ARHCV:1:1115:13679:22994 |
| agrobacterium vitis | gi\|221734005\|gb\|CP000633.1\| Agrobacterium vitis S4 chromosome 1, complete sequence | 1.21028e-15 | M01232:27:000000000-ARHCV:1:1118:6789:13038 |
| nitrococcus mobilis | gi\|106733740\|gb\|DQ514250.1\| Nitrococcus mobilis clone 1 nitrite oxidoreductase alpha subunit (nxrA) gene, partial cds | 1.09615e-85 | M01232:27:000000000-ARHCV:1:2114:20353:6199 |

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|---|---|---|---|
| acidovorax citrulli | gi\|120587178\|gb\|CP000512.1\| Acidovorax citrulli AAC00-1, complete genome | 0.00186218 | M01232:27:000000000-ARHCV:1:1114:14540:9230 |
| escherichia coli | gi\|742672810\|gb\|CP005930.1\| Escherichia coli APEC IMT5155, complete genome | 1.22753e-05 | M01232:27:000000000-ARHCV:1:2101:17497:22937 |
| pseudomonas rhizosphaerae | gi\|692342259\|gb\|CP009533.1\| Pseudomonas rhizosphaerae strain DSM 16299, complete genome | 0.000523684 | M01232:27:000000000-ARHCV:1:1106:10966:14503 |
| microbacterium testaceum | gi\|323272819\|dbj\|AP012052.1\| Microbacterium testaceum StLB037 DNA, complete genome | 8.4367e-05 | M01232:27:000000000-ARHCV:1:1119:18977:4254 |
| rhodococcus pyridinivorans | gi\|568237083\|gb\|CP006997.1\| Rhodococcus pyridinivorans SB3094 plasmid, complete sequence | 6.45806e-05 | M01232:27:000000000-ARHCV:1:1105:24928:16568 |
| postia placenta | gi\|242209322\|ref\|XM_002470464.1\| Postia placenta Mad-698-R hypothetical histidine kinase, mRNA | 0.000571113 | M01232:27:000000000-ARHCV:1:2108:21175:6597 |
| [polyangium] brachysporum | gi\|826168461\|gb\|CP011371.1\| [Polyangium] brachysporum strain DSM 7029, complete genome | 8.84123e-57 | M01232:27:000000000-ARHCV:1:1112:25212:19966 |
| alicyclobacillus acidocaldarius | gi\|339287872\|gb\|CP002902.1\| Alicyclobacillus acidocaldarius subsp. acidocaldarius Tc-4-1, complete genome | 1.2382e-05 | M01232:27:000000000-ARHCV:1:1119:4294:11359 |
| bradyrhizobium oligotrophicum | gi\|456351576\|dbj\|AP012603.1\| Bradyrhizobium oligotrophicum S58 DNA, complete genome | 1.49209e-15 | M01232:27:000000000-ARHCV:1:2111:11108:4645 |
| propionibacterium avidum | gi\|480313929\|gb\|CP005287.1\| Propionibacterium avidum 44067, complete genome | 4.38385e-10 | M01232:27:000000000-ARHCV:1:2117:18608:5992 |
| phenylobacterium zucineum | gi\|196476886\|gb\|CP000747.1\| Phenylobacterium zucineum HLK1, complete genome | 2.19262e-10 | M01232:27:000000000-ARHCV:1:1108:7376:11190 |
| wenzhouxiangella marina | gi\|906394962\|gb\|CP012154.1\| Wenzhouxiangella marina strain KCTC 42284, complete genome | 4.36868e-10 | M01232:27:000000000-ARHCV:1:1104:17172:2933 |
| drosophila melanogaster | gi\|667676433\|gb\|AE013599.5\| Drosophila melanogaster chromosome 2R | 3.33404e-24 | M01232:27:000000000-ARHCV:1:1110:5394:19839 |
| mesorhizobium opportunistum | gi\|336024847\|gb\|CP002279.1\| Mesorhizobium opportunistum WSM2075, complete genome | 4.99281e-99 | M01232:27:000000000-ARHCV:1:1102:24190:20568 |
| altererythrobacter marensis | gi\|831204259\|gb\|CP011805.1\| Altererythrobacter marensis strain KCTC 22370, complete genome | 5.19194e-49 | M01232:27:000000000-ARHCV:1:2110:5671:6507 |

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|---|---|---|---|
| beauveria bassiana | gi\|667647920\|ref\|XM_008597570.1\| Beauveria bassiana ARSEF 2860 ribosomal protein S19 partial mRNA | 3.38892e-11 | M01232:27:000000000-ARHCV:1:2118:19333:1573 |
| streptomyces pristinaespiralis | gi\|924532124\|gb\|CP011340.1\| Streptomyces pristinaespiralis strain HCCB 10218, complete genome | 9.85074e-10 | M01232:27:000000000-ARHCV:1:1101:9506:24227 |
| pseudomonas stutzeri | gi\|395806679\|gb\|CP003725.1\| Pseudomonas stutzeri DSM 10701, complete genome | 9.03088e-42 | M01232:27:000000000-ARHCV:1:1117:26420:16665 |
| thioalkalivibrio sulfidophilus | gi\|219994503\|gb\|CP001339.1\| Thioalkalivibrio sulfidophilus HL-EbGr7, complete genome | 4.38385e-10 | M01232:27:000000000-ARHCV:1:2110:28224:8658 |
| altererythrobacter atlanticus | gi\|918027538\|gb\|CP011452.2\| Altererythrobacter atlanticus strain 26DY36, complete genome | 5.67086e-09 | M01232:27:000000000-ARHCV:1:2103:18251:6834 |
| azotobacter chroococcum | gi\|747125374\|gb\|CP010415.1\| Azotobacter chroococcum NCIMB 8003, complete genome | 5.67086e-09 | M01232:27:000000000-ARHCV:1:2108:19998:18322 |
| paenibacillus durus | gi\|686566537\|gb\|CP009288.1\| Paenibacillus durus strain DSM 1735, complete genome | 0.00175557 | M01232:27:000000000-ARHCV:1:1110:6170:11794 |
| arthrobacter sp. | gi\|674643865\|emb\|LN483070.1\| Arthrobacter sp. 11W110_air genome assembly PRJEB5507_assembly_1, scaffold CONTIG000001 | 7.28401e-13 | M01232:27:000000000-ARHCV:1:1109:25675:17233 |
| pseudomonas denitrificans | gi\|472247168\|gb\|CP004143.1\| Pseudomonas denitrificans ATCC 13867, complete genome | 1.18486e-30 | M01232:27:000000000-ARHCV:1:2105:17256:23698 |
| azoarcus sp. | gi\|119668705\|emb\|AM406670.1\| Azoarcus sp. BH72, complete genome | 2.02523e-13 | M01232:27:000000000-ARHCV:1:1116:23182:24383 |
| bordetella pertussis | gi\|821319309\|emb\|LN849008.1\| Bordetella pertussis genome assembly BPD420, chromosome : 1 | 2.48912e-08 | M01232:27:000000000-ARHCV:1:2115:12835:3331 |
| burkholderia sp. | gi\|307582611\|gb\|CP002217.1\| Burkholderia sp. CCGE1003 chromosome 1, complete sequence | 0.000257971 | M01232:27:000000000-ARHCV:1:1116:20948:14479 |
| chelatococcus sp. | gi\|919432499\|gb\|CP012398.1\| Chelatococcus sp. CO-6, complete genome | 2.52041e-27 | M01232:27:000000000-ARHCV:1:2117:16657:21412 |
| stigmatella aurantiaca | gi\|309390350\|gb\|CP002271.1\| Stigmatella aurantiaca DW4/3-1, complete genome | 4.16482e-45 | M01232:27:000000000-ARHCV:1:1101:16827:1215 |
| neisseria meningitidis | gi\|837359514\|gb\|CP007667.1\| Neisseria meningitidis strain B6116/77, complete genome | 4.31529e-15 | M01232:27:000000000-ARHCV:1:1109:5806:9778 |

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|---|---|---|---|
| myxococcus fulvus | gi\|337255776\|gb\|CP002830.1\| Myxococcus fulvus HW-1, complete genome | 0.00205409 | M01232:27:000000000-ARHCV:1:2108:19607:15295 |
| mesorhizobium huakuii | gi\|657121522\|gb\|CP006581.1\| Mesorhizobium huakuii 7653R genome | 1.18486e-30 | M01232:27:000000000-ARHCV:1:1113:25598:21201 |
| gordonia sp. | gi\|403643428\|gb\|CP002907.1\| Gordonia sp. KTR9, complete genome | 1.04319e-120 | M01232:27:000000000-ARHCV:1:1118:22881:8488 |
| uncultured bacteroidales | gi\|190402440\|gb\|EU573802.1\| Uncultured Bacteroidales bacterium clone CE5 16S ribosomal RNA gene, partial sequence | 6.01749e-148 | M01232:27:000000000-ARHCV:1:2103:7750:16256 |
| rhodospirillum photometricum | gi\|378401447\|emb\|HE663493.1\| Rhodospirillum photometricum DSM 122 draft genome sequence | 1.87728e-08 | M01232:27:000000000-ARHCV:1:1119:18419:17718 |
| roseibacterium elongatum | gi\|594547454\|gb\|CP004372.1\| Roseibacterium elongatum DSM 19469, complete genome | 9.20863e-27 | M01232:27:000000000-ARHCV:1:1108:28976:16113 |
| nocardia cyriacigeorgica | gi\|374843763\|emb\|FO082843.1\| Nocardia cyriacigeorgica GUH-2 chromosome complete genome | 0.000158791 | M01232:27:000000000-ARHCV:1:2101:12127:18126 |
| granulicella mallensis | gi\|358750971\|gb\|CP003130.1\| Granulicella mallensis MP5ACTX8, complete genome | 4.25416e-30 | M01232:27:000000000-ARHCV:1:1119:10147:7945 |
| azospirillum brasilense | gi\|646258717\|gb\|CP007794.1\| Azospirillum brasilense strain Az39 plasmid AbAZ39_p1, complete sequence | 3.35921e-16 | M01232:27:000000000-ARHCV:1:1103:7109:20892 |
| conexibacter woesei | gi\|283945692\|gb\|CP001854.1\| Conexibacter woesei DSM 14684, complete genome | 5.14171e-08 | M01232:27:000000000-ARHCV:1:1113:26276:8684 |
| gallus gallus | gi\|407948025\|gb\|JX847203.1\| Gallus gallus cytochrome P450 2J2-like protein (LOC772391) mRNA, partial cds | 1.39362e-21 | M01232:27:000000000-ARHCV:1:2119:8178:17709 |
| amycolatopsis orientalis | gi\|505812666\|gb\|CP003410.1\| Amycolatopsis orientalis HCCB10007, complete genome | 1.11591e-05 | M01232:27:000000000-ARHCV:1:2104:17304:19996 |
| martelella endophytica | gi\|779727330\|gb\|CP010803.1\| Martelella endophytica strain YC6887, complete genome | 2.52e-37 | M01232:27:000000000-ARHCV:1:1115:28225:10966 |
| paenibacillus sp. | gi\|686539396\|gb\|CP009284.1\| Paenibacillus sp. FSL R7-0331, complete genome | 3.52424e-30 | M01232:27:000000000-ARHCV:1:1107:23457:6385 |

## APPENDIX 1 (D) Metagenomic BLAST results for the Garden inoculum source: nxrA gene

| Organism/HIT | Accession | e-value | Fastq header |
|---|---|---|---|
| No hits | None | 0 | M01232:27:000000000-ARHCV:1:1118:12778:6679 |
| uncultured nitrite-oxidizing | gi\|445068180\|gb\|KC152738.1\| Uncultured nitrite-oxidizing bacterium clone NT2c28 nitrite oxidoreductase alpha subunit (nxrA) gene, partial cds | 8.34029e-97 | M01232:27:000000000-ARHCV:1:1114:9481:1859 |
| uncultured bacterium | gi\|380772399\|gb\|JN969912.1\| Uncultured bacterium clone 63 nitrite oxidoreductase gene, partial cds | 1.58445e-50 | M01232:27:000000000-ARHCV:1:2117:19126:5855 |
| nitrobacter vulgaris | gi\|90102361\|gb\|DQ421377.1\| Nitrobacter vulgaris clone 2 nitrite oxidoreductase alpha subunit (norA) gene, partial cds | 1.76132e-108 | M01232:27:000000000-ARHCV:1:2106:5427:4927 |
| nitrobacter winogradskyi | gi\|661349933\|gb\|KJ023584.1\| Nitrobacter winogradskyi isolate DGGE gel band nit-8 NorA (norA) gene, partial cds | 1.09131e-07 | M01232:27:000000000-ARHCV:1:2116:28144:14678 |
| nitrobacter alkalicus | gi\|90102349\|gb\|DQ421371.1\| Nitrobacter alkalicus clone 2b nitrite oxidoreductase alpha subunit (norA) gene, partial cds >gi\|90102351\|gb\|DQ421372.1\| Nitrobacter alkalicus clone 2a nitrite oxidoreductase alpha subunit (norA) gene, partial cds | 3.69928e-130 | M01232:27:000000000-ARHCV:1:1109:26589:11640 |
| unidentified bacterium | gi\|106733782\|gb\|DQ514271.1\| Unidentified bacterium clone LGj nitrite oxidoreductase alpha subunit (nxrA) gene, partial cds | 3.79366e-07 | M01232:27:000000000-ARHCV:1:1102:6359:22956 |
| altererythrobacter marensis | gi\|831204259\|gb\|CP011805.1\| Altererythrobacter marensis strain KCTC 22370, complete genome | 1.35933e-24 | M01232:27:000000000-ARHCV:1:2106:21843:21194 |
| serratia marcescens | gi\|440050501\|gb\|CP003942.1\| Serratia marcescens FGI94, complete genome | 0.0043855 | M01232:27:000000000-ARHCV:1:1102:15800:15466 |
| cyprinus carpio | gi\|685042153\|emb\|LN590678.1\| Cyprinus carpio genome assembly common carp genome ,scaffold LG28 | 0.00136508 | M01232:27:000000000-ARHCV:1:2106:14937:11230 |

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|---|---|---|---|
| streptomyces sp. | gi\|829477974\|gb\|CP011664.1\| Streptomyces sp. Mg1, complete genome | 4.12268e-06 | M01232:27:000000000-ARHCV:1:2110:26260:6822 |
| hoeflea sp. | gi\|822663067\|gb\|CP011479.1\| Hoeflea sp. IMCC20628, complete genome | 4.06361e-05 | M01232:27:000000000-ARHCV:1:2111:11691:9154 |
| pseudomonas aeruginosa | gi\|926451317\|emb\|LN871187.1\| Pseudomonas aeruginosa genome assembly PAO1OR, chromosome : I | 2.08913e-142 | M01232:27:000000000-ARHCV:1:1114:21815:3910 |
| sphingomonas sp. | gi\|469477505\|gb\|CP004036.1\| Sphingomonas sp. MM-1, complete genome | 9.42247e-12 | M01232:27:000000000-ARHCV:1:2108:21710:14176 |
| streptomyces albus | gi\|749174630\|gb\|CP010519.1\| Streptomyces albus strain DSM 41398, complete genome | 2.84951e-11 | M01232:27:000000000-ARHCV:1:1103:9473:4584 |
| pseudomonas denitrificans | gi\|472247168\|gb\|CP004143.1\| Pseudomonas denitrificans ATCC 13867, complete genome | 1.02487e-10 | M01232:27:000000000-ARHCV:1:1114:18917:11835 |
| myxococcus fulvus | gi\|817709775\|gb\|CP006003.1\| Myxococcus fulvus 124B02, complete genome | 5.59119e-19 | M01232:27:000000000-ARHCV:1:2101:9257:16378 |
| mesorhizobium huakuii | gi\|657121522\|gb\|CP006581.1\| Mesorhizobium huakuii 7653R genome | 7.41387e-26 | M01232:27:000000000-ARHCV:1:1116:22992:9303 |
| hyphomicrobium nitrativorans | gi\|563343116\|gb\|CP006912.1\| Hyphomicrobium nitrativorans NL23, complete genome | 1.40819e-64 | M01232:27:000000000-ARHCV:1:1102:24829:14234 |
| isoptericola variabilis | gi\|470488827\|ref\|NR_076890.1\| Isoptericola variabilis strain 225 23S ribosomal RNA gene, complete sequence | 1.33192e-20 | M01232:27:000000000-ARHCV:1:2113:24614:11263 |
| propionibacterium avidum | gi\|480313929\|gb\|CP005287.1\| Propionibacterium avidum 44067, complete genome | 4.38385e-10 | M01232:27:000000000-ARHCV:1:1106:25399:4943 |
| thermoanaerobacter kivui | gi\|694165048\|gb\|CP009170.1\| Thermoanaerobacter kivui strain DSM 2030, complete genome | 0.000447744 | M01232:27:000000000-ARHCV:1:1118:20644:14245 |
| singulisphaera acidiphila | gi\|430012750\|gb\|CP003364.1\| Singulisphaera acidiphila DSM 18658, complete genome | 1.53304e-19 | M01232:27:000000000-ARHCV:1:1112:28824:12626 |
| tistrella mobilis | gi\|388531416\|gb\|CP003239.1\| Tistrella mobilis KA081020-065 plasmid pTM3, complete sequence | 0.000571113 | M01232:27:000000000-ARHCV:1:2110:21102:15473 |
| pseudogulbenkiania sp. | gi\|345641016\|dbj\|AP012224.1\| Pseudogulbenkiania sp. NH8B DNA, complete genome | 3.31775e-26 | M01232:27:000000000-ARHCV:1:2105:13290:23547 |

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|---|---|---|---|
| saccharomonospora viridis | gi\|256583961\|gb\|CP001683.1\| Saccharomonospora viridis DSM 43017, complete genome | 1.18928e-14 | M01232:27:000000000-ARHCV:1:1113:9360:24370 |
| mycobacterium rhodesiae | gi\|359817839\|gb\|CP003169.1\| Mycobacterium rhodesiae NBB3, complete genome | 1.84728e-78 | M01232:27:000000000-ARHCV:1:2119:7624:20217 |
| dyella japonica | gi\|664782987\|gb\|CP008884.1\| Dyella japonica A8, complete genome | 2.5467e-32 | M01232:27:000000000-ARHCV:1:2118:15272:4172 |
| rhodopseudomonas palustris | gi\|90103542\|gb\|CP000301.1\| Rhodopseudomonas palustris BisB18, complete genome | 9.35604e-17 | M01232:27:000000000-ARHCV:1:2111:18597:6297 |
| starkeya novella | gi\|296926528\|gb\|CP002026.1\| Starkeya novella DSM 506, complete genome | 5.12819e-69 | M01232:27:000000000-ARHCV:1:2111:4358:12784 |
| bradyrhizobium japonicum | gi\|736032532\|gb\|CP010313.1\| Bradyrhizobium japonicum strain E109, complete genome | 4.0517e-15 | M01232:27:000000000-ARHCV:1:2117:23156:18410 |
| sinorhizobium fredii | gi\|227337257\|gb\|CP000874.1\| Sinorhizobium fredii NGR234 plasmid pNGR234b, complete sequence | 3.31819e-21 | M01232:27:000000000-ARHCV:1:2103:26851:17133 |
| nitrococcus mobilis | gi\|106733740\|gb\|DQ514250.1\| Nitrococcus mobilis clone 1 nitrite oxidoreductase alpha subunit (nxrA) gene, partial cds | 2.02571e-11 | M01232:27:000000000-ARHCV:1:1114:19085:8155 |
| conexibacter woesei | gi\|283945692\|gb\|CP001854.1\| Conexibacter woesei DSM 14684, complete genome | 0.00310693 | M01232:27:000000000-ARHCV:1:2119:16942:7703 |
| uncultured nitrobacter | gi\|395486128\|gb\|JX020943.1\| Uncultured Nitrobacter sp. isolate DGGE gel band 11 nitrite oxidoreductase alpha subunit (norA) gene, partial cds | 1.3093e-40 | M01232:27:000000000-ARHCV:1:1109:16979:9468 |
| rhodothermus marinus | gi\|345111121\|gb\|CP003029.1\| Rhodothermus marinus SG0.5JP17-172, complete genome | 2.63902e-05 | M01232:27:000000000-ARHCV:1:1112:16531:11598 |
| ensifer adhaerens | gi\|589085422\|gb\|CP007236.1\| Ensifer adhaerens OV14 chromosome 1 sequence | 1.11394e-50 | M01232:27:000000000-ARHCV:1:2110:11141:21127 |
| uncultured organism | gi\|315937114\|gb\|HM486076.1\| Uncultured organism CA915 glycopeptide biosynthetic gene cluster, complete sequence | 1.02598e-28 | M01232:27:000000000-ARHCV:1:1109:25755:19396 |
| mycobacterium sp. | gi\|126232413\|gb\|CP000580.1\| Mycobacterium sp. JLS, complete genome | 8.34029e-97 | M01232:27:000000000-ARHCV:1:1106:14769:14256 |

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| marinovum algicola | gi\|868874654\|gb\|CP010855.1\| Marinovum algicola DG 898, complete genome | 9.17183e-06 | M01232:27:000000000-ARHCV:1:1113:14930:4354 |
| thauera sp. | gi\|237624339\|gb\|CP001281.2\| Thauera sp. MZ1T, complete genome | 4.07298e-39 | M01232:27:000000000-ARHCV:1:1110:14042:14276 |
| filomicrobium sp. | gi\|781851931\|emb\|LN829119.1\| Filomicrobium sp. Y genome assembly Y1, chromosome : 1 | 6.9813e-43 | M01232:27:000000000-ARHCV:1:2115:20569:12079 |
| methylobacterium extorquens | gi\|218525559\|gb\|CP001299.1\| Methylobacterium extorquens CM4 plasmid pCMU01, complete sequence | 9.38703e-12 | M01232:27:000000000-ARHCV:1:1102:8123:17761 |
| akkermansia muciniphila | gi\|187424568\|gb\|CP001071.1\| Akkermansia muciniphila ATCC BAA-835, complete genome | 0.00150919 | M01232:27:000000000-ARHCV:1:2110:10144:15642 |
| sanguibacter keddieii | gi\|269095543\|gb\|CP001819.1\| Sanguibacter keddieii DSM 10542, complete genome | 1.2254e-05 | M01232:27:000000000-ARHCV:1:2107:11335:14552 |
| methyloceanibacter caenitepidi | gi\|743966687\|dbj\|AP014648.1\| Methyloceanibacter caenitepidi DNA, complete genome, strain: Gela4 | 1.11967e-70 | M01232:27:000000000-ARHCV:1:1107:17252:21787 |
| rhodococcus sp. | gi\|769526933\|gb\|CP010797.1\| Rhodococcus sp. B7740, complete genome | 5.98398e-88 | M01232:27:000000000-ARHCV:1:1117:28953:15357 |
| ralstonia mannitolilytica | gi\|770603614\|gb\|CP010799.2\| Ralstonia mannitolilytica strain SN82F48 chromosome 1, complete sequence | 1.22753e-05 | M01232:27:000000000-ARHCV:1:1101:16260:7723 |
| thalassospira xiamenensis | gi\|745807430\|gb\|CP004388.1\| Thalassospira xiamenensis M-5 = DSM 17429, complete genome | 1.36857e-16 | M01232:27:000000000-ARHCV:1:2118:10439:11250 |
| oryza sativa | gi\|51491515\|gb\|AC119288.4\| Oryza sativa Japonica Group chromosome 5 clone OSJNBa0017J22, complete sequence | 1.98627e-07 | M01232:27:000000000-ARHCV:1:2117:20994:22138 |
| opitutaceae bacterium | gi\|573471959\|gb\|CP007053.1\| Opitutaceae bacterium TAV5, complete genome | 1.81461e-28 | M01232:27:000000000-ARHCV:1:2119:10547:23457 |
| mycobacterium canettii | gi\|432160663\|emb\|FO203509.1\| Mycobacterium canettii CIPT 140070010 complete genome | 2.35603e-87 | M01232:27:000000000-ARHCV:1:2118:8577:17292 |
| gemmatirosa kalamazoonesis | gi\|575456455\|gb\|CP007128.1\| Gemmatirosa kalamazoonesis strain KBS708, complete genome | 1.03651e-07 | M01232:27:000000000-ARHCV:1:2114:17388:9038 |
| opitutus terrae | gi\|177839040\|gb\|CP001032.1\| Opitutus terrae PB90-1, complete genome | 1.90717e-53 | M01232:27:000000000-ARHCV:1:1102:26398:9977 |

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|---|---|---|---|
| mycobacterium gilvum | gi\|315265130\|gb\|CP002386.1\| Mycobacterium gilvum Spyr1 plasmid pMSPYR101, complete sequence | 2.87504e-126 | M01232:27:000000000-ARHCV:1:1111:8796:7532 |
| methylocella silvestris | gi\|217501576\|gb\|CP001280.1\| Methylocella silvestris BL2, complete genome | 1.76502e-05 | M01232:27:000000000-ARHCV:1:1112:17864:9725 |
| streptomyces lydicus | gi\|768311912\|gb\|CP007699.1\| Streptomyces lydicus A02, complete genome | 1.15998e-45 | M01232:27:000000000-ARHCV:1:2112:9098:6517 |
| rhodobacter sphaeroides | gi\|145554299\|gb\|CP000661.1\| Rhodobacter sphaeroides ATCC 17025, complete genome | 5.16757e-24 | M01232:27:000000000-ARHCV:1:1114:21764:16851 |
| deinococcus swuensis | gi\|730601855\|gb\|CP010028.1\| Deinococcus swuensis strain DY59, complete genome | 3.36958e-05 | M01232:27:000000000-ARHCV:1:2108:13122:21248 |
| azospirillum lipoferum | gi\|357422594\|emb\|FQ311868.1\| Azospirillum lipoferum 4B main chromosome, complete genome | 2.89929e-56 | M01232:27:000000000-ARHCV:1:2114:13325:23727 |
| alcanivorax pacificus | gi\|745803723\|gb\|CP004387.1\| Alcanivorax pacificus W11-5, complete genome | 2.29029e-107 | M01232:27:000000000-ARHCV:1:2114:4819:12800 |
| influenza a | gi\|479284922\|gb\|KC815855.1\| Influenza A virus (A/mallard/Italy/3401/2005(H5N1)) segment 8 nuclear export protein (NEP) and nonstructural protein 1 (NS1) genes, complete cds | 2.17587e-142 | M01232:27:000000000-ARHCV:1:1103:27401:18282 |
| predicted: pantholops | gi\|556746637\|ref\|XM_005968721.1\| PREDICTED: Pantholops hodgsonii 3-oxoacyl-[acyl-carrier-protein] reductase, chloroplastic-like (LOC102344020), mRNA | 1.19967e-20 | M01232:27:000000000-ARHCV:1:1105:13437:23413 |
| hyphomonas neptunium | gi\|114737225\|gb\|CP000158.1\| Hyphomonas neptunium ATCC 15444, complete genome | 9.22459e-27 | M01232:27:000000000-ARHCV:1:2118:15444:10368 |
| ilumatobacter coccineus | gi\|464097432\|dbj\|AP012057.1\| Ilumatobacter coccineus YM16-304 DNA, complete genome | 8.51919e-82 | M01232:27:000000000-ARHCV:1:2106:19187:14726 |
| burkholderia ambifaria | gi\|773032576\|gb\|CP009800.1\| Burkholderia ambifaria AMMD chromosome 3, complete sequence | 6.27819e-12 | M01232:27:000000000-ARHCV:1:2111:27509:14743 |
| infectious bronchitis | gi\|827048795\|gb\|KP343691.1\| Infectious bronchitis virus strain ck/CH/LGX/130530, complete genome | 6.65351e-44 | M01232:27:000000000-ARHCV:1:2118:20310:8888 |

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| devosia sp. | gi\|901895891\|gb\|CP011300.1\| Devosia sp. H5989, complete genome | 1.08293e-80 | M01232:27:000000000-ARHCV:1:2111:22115:16513 |
| phenylobacterium zucineum | gi\|196476886\|gb\|CP000747.1\| Phenylobacterium zucineum HLK1, complete genome | 1.23489e-19 | M01232:27:000000000-ARHCV:1:1114:23738:20095 |
| xanthobacter autotrophicus | gi\|154158043\|gb\|CP000781.1\| Xanthobacter autotrophicus Py2, complete genome | 4.40045e-54 | M01232:27:000000000-ARHCV:1:2115:26732:16661 |
| celeribacter indicus | gi\|748262133\|gb\|CP004393.1\| Celeribacter indicus strain P73, complete genome | 1.9827e-28 | M01232:27:000000000-ARHCV:1:2112:23735:2230 |
| mycobacterium smegmatis | gi\|433294648\|gb\|CP003078.1\| Mycobacterium smegmatis JS623, complete genome | 0.00205409 | M01232:27:000000000-ARHCV:1:1105:25266:21953 |
| myxococcus xanthus | gi\|108460647\|gb\|CP000113.1\| Myxococcus xanthus DK 1622, complete genome | 5.13125e-09 | M01232:27:000000000-ARHCV:1:1110:3941:10855 |
| hyphomicrobium sp. | gi\|337757426\|emb\|FQ859181.1\| Hyphomicrobium sp. MC1 chromosome, complete genome | 1.54918e-19 | M01232:27:000000000-ARHCV:1:1113:15030:20828 |
| bradyrhizobium sp. | gi\|381356398\|dbj\|AP012279.1\| Bradyrhizobium sp. S23321 DNA, complete genome | 1.86458e-24 | M01232:27:000000000-ARHCV:1:2105:12951:24777 |
| klebsiella pneumoniae | gi\|820949075\|gb\|CP011421.1\| Klebsiella pneumoniae strain yzusk-4 genome | 4.39969e-05 | M01232:27:000000000-ARHCV:1:1119:10086:1304 |
| nocardia nova | gi\|576082433\|gb\|CP006850.1\| Nocardia nova SH22a, complete genome | 9.32367e-17 | M01232:27:000000000-ARHCV:1:2104:11250:20613 |
| nitrospira moscoviensis | gi\|922305621\|gb\|CP011801.1\| Nitrospira moscoviensis strain NSP M-1, complete genome | 2.40316e-15 | M01232:27:000000000-ARHCV:1:1103:14994:7027 |
| azotobacter chroococcum | gi\|747125374\|gb\|CP010415.1\| Azotobacter chroococcum NCIMB 8003, complete genome | 5.67086e-09 | M01232:27:000000000-ARHCV:1:2117:18039:23306 |
| polymorphum gilvum | gi\|326411376\|gb\|CP002568.1\| Polymorphum gilvum SL003B-26A1, complete genome | 9.57688e-56 | M01232:27:000000000-ARHCV:1:1103:8345:11035 |
| burkholderia glumae | gi\|755895103\|gb\|CP002580.1\| Burkholderia glumae PG1 chromosome 1, complete sequence | 5.67086e-09 | M01232:27:000000000-ARHCV:1:2109:8121:6032 |
| pseudomonas chlororaphis | gi\|787852299\|gb\|CP011110.1\| Pseudomonas chlororaphis strain PCL1606, complete genome | 4.30544e-59 | M01232:27:000000000-ARHCV:1:2111:12794:3677 |
| paracoccus aminophilus | gi\|529579793\|gb\|CP006650.1\| Paracoccus aminophilus JCM 7686, complete genome | 3.05904e-16 | M01232:27:000000000-ARHCV:1:1117:19610:17124 |

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| pandoraea oxalativorans | gi\|828143611\|gb\|CP011253.2\| Pandoraea oxalativorans strain DSM 23570, complete genome | 6.18206e-06 | M01232:27:000000000-ARHCV:1:1104:21359:18609 |
| desulfovibrio aespoeensis | gi\|316941978\|gb\|CP002431.1\| Desulfovibrio aespoeensis Aspo-2, complete genome | 1.39267e-07 | M01232:27:000000000-ARHCV:1:1115:6259:11974 |
| modestobacter marinus | gi\|388483940\|emb\|FO203431.1\| Modestobacter marinus str. BC501 chromosome, complete genome | 4.84472e-07 | M01232:27:000000000-ARHCV:1:2104:15534:17140 |
| enterococcus malodoratus | gi\|315259499\|gb\|HQ611251.1\| Enterococcus malodoratus strain ATCC 43197 DNA-directed RNA polymerase beta subunit (rpoB) gene, partial cds | 3.96886e-15 | M01232:27:000000000-ARHCV:1:2118:9332:5506 |
| amycolatopsis mediterranei | gi\|532225686\|gb\|CP003777.1\| Amycolatopsis mediterranei RB, complete genome | 1.93874e-37 | M01232:27:000000000-ARHCV:1:2107:25040:11306 |
| synechococcus sp. | gi\|572996165\|gb\|CP006882.1\| Synechococcus sp. WH 8109, complete genome | 5.21549e-16 | M01232:27:000000000-ARHCV:1:1103:24345:12711 |
| endosymbiont of | gi\|530669580\|dbj\|AP012978.1\| Endosymbiont of unidentified scaly snail isolate Monju DNA, complete genome | 1.2254e-05 | M01232:27:000000000-ARHCV:1:2119:14028:23697 |
| paracoccus denitrificans | gi\|119376152\|gb\|CP000490.1\| Paracoccus denitrificans PD1222 chromosome 2, complete sequence | 5.43519e-39 | M01232:27:000000000-ARHCV:1:2103:22622:9371 |
| thioalkalivibrio paradoxus | gi\|570725612\|gb\|CP007029.1\| Thioalkalivibrio paradoxus ARh 1, complete genome | 4.52576e-19 | M01232:27:000000000-ARHCV:1:1107:6396:13270 |
| neorhizobium galegae, | gi\|659653153\|emb\|HG938353.1\| Neorhizobium galegae, complete genome | 8.39212e-61 | M01232:27:000000000-ARHCV:1:2104:16327:23921 |
| sorangium cellulosum | gi\|161158851\|emb\|AM746676.1\| Sorangium cellulosum 'So ce 56' complete genome | 4.91319e-08 | M01232:27:000000000-ARHCV:1:1109:23914:2629 |
| beutenbergia cavernae | gi\|229564415\|gb\|CP001618.1\| Beutenbergia cavernae DSM 12333, complete genome | 1.48116e-17 | M01232:27:000000000-ARHCV:1:2101:12105:19430 |
| streptomyces ambofaciens | gi\|917646380\|gb\|CP012382.1\| Streptomyces ambofaciens ATCC 23877, complete genome | 1.89694e-28 | M01232:27:000000000-ARHCV:1:2115:22396:20715 |
| delftia acidovorans | gi\|160361034\|gb\|CP000884.1\| Delftia acidovorans SPH-1, complete genome | 5.12144e-09 | M01232:27:000000000-ARHCV:1:1118:19746:24092 |

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| rubrivivax gelatinosus | gi\|381376528\|dbj\|AP012320.1\| Rubrivivax gelatinosus IL144 DNA, complete genome | 4.18712e-40 | M01232:27:000000000-ARHCV:1:1112:5690:19793 |
| delftia sp. | gi\|333741867\|gb\|CP002735.1\| Delftia sp. Cs1-4, complete genome | 0.000158791 | M01232:27:000000000-ARHCV:1:2109:24682:10527 |
| arsenicicoccus sp. | gi\|908724324\|gb\|CP012070.1\| Arsenicicoccus sp. oral taxon 190, complete genome | 4.32226e-20 | M01232:27:000000000-ARHCV:1:2105:17600:16762 |
| rhizobium sp. | gi\|549127343\|emb\|HG518323.1\| Rhizobium sp. IRBG74 linear chromosome, complete genome | 2.44086e-62 | M01232:27:000000000-ARHCV:1:1111:12810:24810 |
| mycobacterium neoaurum | gi\|674790876\|gb\|CP006936.2\| Mycobacterium neoaurum VKM Ac-1815D, complete genome | 7.15011e-52 | M01232:27:000000000-ARHCV:1:1118:26005:20649 |
| burkholderia fungorum | gi\|780551828\|gb\|CP010025.1\| Burkholderia fungorum strain ATCC BAA-463 chromosome 3, complete sequence | 3.35339e-16 | M01232:27:000000000-ARHCV:1:2101:19347:22944 |
| pseudomonas putida | gi\|169757190\|gb\|CP000949.1\| Pseudomonas putida W619, complete genome | 8.95247e-08 | M01232:27:000000000-ARHCV:1:1114:9517:24232 |
| anaerolinea thermophila | gi\|319993263\|dbj\|AP012029.1\| Anaerolinea thermophila UNI-1 DNA, complete genome | 3.76692e-12 | M01232:27:000000000-ARHCV:1:1101:8803:14421 |
| mesorhizobium australicum | gi\|433663430\|gb\|CP003358.1\| Mesorhizobium australicum WSM2073, complete genome | 8.904e-52 | M01232:27:000000000-ARHCV:1:1106:16232:4261 |
| ralstonia solanacearum | gi\|334194119\|gb\|CP002819.1\| Ralstonia solanacearum Po82, complete genome | 7.93843e-16 | M01232:27:000000000-ARHCV:1:1114:29101:16242 |
| chelativorans sp. | gi\|110283346\|gb\|CP000390.1\| Chelativorans sp. BNC1, complete genome | 2.26101e-21 | M01232:27:000000000-ARHCV:1:1114:16259:12619 |
| streptosporangium roseum | gi\|270504784\|gb\|CP001814.1\| Streptosporangium roseum DSM 43021, complete genome | 3.93996e-40 | M01232:27:000000000-ARHCV:1:1112:18076:7915 |
| streptomyces leeuwenhoekii | gi\|822875672\|emb\|LN831790.1\| Streptomyces leeuwenhoekii genome assembly sleC34, chromosome : chromosome | 2.01822e-13 | M01232:27:000000000-ARHCV:1:1111:17304:8782 |
| arthrobacter sp. | gi\|403227528\|gb\|CP003203.1\| Arthrobacter sp. Rue61a, complete genome | 8.29585e-102 | M01232:27:000000000-ARHCV:1:2104:15781:21386 |
| aeromonas hydrophila | gi\|827370414\|gb\|CP006870.1\| Aeromonas hydrophila NJ-35, complete genome | 1.13697e-07 | M01232:27:000000000-ARHCV:1:2101:14340:4009 |

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| sinorhizobium sp. | gi\|754297128\|gb\|KM405900.1\| Sinorhizobium sp. JZ3_1_nodule RNA polymerase beta (rpoB) gene, partial cds | 4.16653e-09 | M01232:27:000000000-ARHCV:1:1104:12422:20779 |
| mycobacterium vanbaalenii | gi\|119953846\|gb\|CP000511.1\| Mycobacterium vanbaalenii PYR-1, complete genome | 5.67733e-72 | M01232:27:000000000-ARHCV:1:2110:21787:23190 |
| microbacterium sp. | gi\|914697494\|gb\|CP012299.1\| Microbacterium sp. CGR1, complete genome | 2.74172e-80 | M01232:27:000000000-ARHCV:1:1115:15667:24581 |
| rhodomicrobium vannielii | gi\|311217923\|gb\|CP002292.1\| Rhodomicrobium vannielii ATCC 17100, complete genome | 1.19328e-25 | M01232:27:000000000-ARHCV:1:2110:4860:15383 |
| salinispora tropica | gi\|145301903\|gb\|CP000667.1\| Salinispora tropica CNB-440, complete genome | 4.23149e-35 | M01232:27:000000000-ARHCV:1:1109:20423:1216 |
| nocardiopsis alba | gi\|402798256\|gb\|CP003788.1\| Nocardiopsis alba ATCC BAA-2165, complete genome | 7.67586e-14 | M01232:27:000000000-ARHCV:1:2102:15498:15644 |
| uncultured ammonia-oxidizing | gi\|591400927\|gb\|KJ081645.1\| Uncultured ammonia-oxidizing bacterium clone lj-aob1 ammonia monooxygenase (amoA) gene, partial cds | 2.85791e-69 | M01232:27:000000000-ARHCV:1:2117:23916:14956 |
| pelagibacterium halotolerans | gi\|351591711\|gb\|CP003075.1\| Pelagibacterium halotolerans B2, complete genome | 7.25881e-13 | M01232:27:000000000-ARHCV:1:2112:10520:7799 |
| streptomyces halstedii | gi\|112419465\|dbj\|AB241068.1\| Streptomyces halstedii halstoctacosanolide biosynthetic gene cluster (hlsA, hlsB, hlsC, hlsD, hlsE, hlsF, hlsG, hlsH, hlsI), complete cds | 3.39446e-33 | M01232:27:000000000-ARHCV:1:1101:25124:18334 |
| azoarcus sp. | gi\|358635055\|dbj\|AP012304.1\| Azoarcus sp. KH32C DNA, complete genome | 9.92636e-07 | M01232:27:000000000-ARHCV:1:1105:10033:7452 |
| brucella suis | gi\|756787364\|gb\|CP010850.1\| Brucella suis strain Human/AR/US/1981 chromosome I, complete sequence | 1.29847e-09 | M01232:27:000000000-ARHCV:1:2104:28049:15861 |
| cellulomonas flavigena | gi\|296019684\|gb\|CP001964.1\| Cellulomonas flavigena DSM 20109, complete genome | 2.37276e-82 | M01232:27:000000000-ARHCV:1:1101:6887:15219 |
| pseudoxanthomonas suwonensis | gi\|317464132\|gb\|CP002446.1\| Pseudoxanthomonas suwonensis 11-1, complete genome | 1.12644e-11 | M01232:27:000000000-ARHCV:1:2105:16150:10204 |

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| pseudomonas entomophila | gi\|95101722\|emb\|CT573326.1\| Pseudomonas entomophila str. L48 chromosome,complete sequence | 3.30684e-11 | M01232:27:000000000-ARHCV:1:1118:9513:21565 |
| verticillium albo-atrum | gi\|558701596\|gb\|KC350985.1\| Verticillium albo-atrum strain CBS 130340 28S ribosomal RNA gene, partial sequence | 2.96591e-35 | M01232:27:000000000-ARHCV:1:2105:15116:2550 |
| gluconacetobacter diazotrophicus | gi\|209529865\|gb\|CP001189.1\| Gluconacetobacter diazotrophicus PAl 5, complete genome | 0.00195458 | M01232:27:000000000-ARHCV:1:2112:12835:20282 |
| bradyrhizobium oligotrophicum | gi\|456351576\|dbj\|AP012603.1\| Bradyrhizobium oligotrophicum S58 DNA, complete genome | 2.44566e-52 | M01232:27:000000000-ARHCV:1:2115:10326:11340 |
| streptomyces cyaneogriseus | gi\|758855033\|gb\|CP010849.1\| Streptomyces cyaneogriseus subsp. noncyanogenus strain NMWT 1, complete genome | 5.06396e-89 | M01232:27:000000000-ARHCV:1:2110:23064:13087 |
| brevundimonas subvibrioides | gi\|302191744\|gb\|CP002102.1\| Brevundimonas subvibrioides ATCC 15264, complete genome | 4.95449e-42 | M01232:27:000000000-ARHCV:1:1101:28552:13146 |
| agrobacterium sp. | gi\|325059306\|gb\|CP002248.1\| Agrobacterium sp. H13-3 circular chromosome, complete sequence | 1.96872e-33 | M01232:27:000000000-ARHCV:1:1118:25802:4043 |
| haloarcula hispanica | gi\|564120640\|gb\|CP006884.1\| Haloarcula hispanica N601 chromosome 1, complete sequence | 2.23875e-05 | M01232:27:000000000-ARHCV:1:2119:24215:10721 |
| azotobacter vinelandii | gi\|482534342\|gb\|CP005095.1\| Azotobacter vinelandii CA6, complete genome | 4.07736e-60 | M01232:27:000000000-ARHCV:1:1101:20613:20407 |
| rubrobacter xylanophilus | gi\|108764099\|gb\|CP000386.1\| Rubrobacter xylanophilus DSM 9941, complete genome | 2.43242e-62 | M01232:27:000000000-ARHCV:1:2111:10007:20178 |
| ramlibacter tataouinensis | gi\|334728683\|gb\|CP000245.1\| Ramlibacter tataouinensis TTB310, complete genome | 4.80573e-73 | M01232:27:000000000-ARHCV:1:1105:19835:17720 |
| streptomyces lividans | gi\|672367150\|gb\|CP009124.1\| Streptomyces lividans TK24, complete genome | 1.67361e-67 | M01232:27:000000000-ARHCV:1:1106:11986:5548 |
| kineococcus radiotolerans | gi\|196121877\|gb\|CP000750.2\| Kineococcus radiotolerans SRS30216, complete genome | 1.21466e-10 | M01232:27:000000000-ARHCV:1:2119:10944:7294 |
| azoarcus aromaticum | gi\|56311475\|emb\|CR555306.1\| Azoarcus aromaticum EbN1 complete genome | 8.79865e-13 | M01232:27:000000000-ARHCV:1:2104:12221:20056 |
| propionibacterium propionicum | gi\|395140796\|gb\|CP002734.1\| Propionibacterium propionicum F0230a, complete genome | 7.37738e-09 | M01232:27:000000000-ARHCV:1:1106:27676:14677 |

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| marinobacter sp. | gi\|914748683\|gb\|CP011929.1\| Marinobacter sp. CP1, complete genome | 2.6198e-12 | M01232:27:000000000-ARHCV:1:1108:21120:23809 |
| sandaracinus amylolyticus | gi\|816953346\|gb\|CP011125.1\| Sandaracinus amylolyticus strain DSM 53668, complete genome | 1.71867e-71 | M01232:27:000000000-ARHCV:1:2106:22021:24569 |
| streptomyces fulvissimus | gi\|485091803\|gb\|CP005080.1\| Streptomyces fulvissimus DSM 40593, complete genome | 4.71074e-41 | M01232:27:000000000-ARHCV:1:1109:10791:3267 |
| marichromatium purpuratum | gi\|570731247\|gb\|CP007031.1\| Marichromatium purpuratum 984, complete genome | 7.15683e-23 | M01232:27:000000000-ARHCV:1:2112:11945:9069 |
| pseudozyma flocculosa | gi\|630969975\|ref\|XM_007883205.1\| Pseudozyma flocculosa PF-1 hypothetical protein partial mRNA | 0.000543446 | M01232:27:000000000-ARHCV:1:2110:10020:8374 |
| mycobacterium abscessus | gi\|506960221\|gb\|CP004374.1\| Mycobacterium abscessus subsp. bolletii 50594, complete genome | 7.06853e-33 | M01232:27:000000000-ARHCV:1:2111:20939:19460 |
| sphingopyxis alaskensis | gi\|98975575\|gb\|CP000356.1\| Sphingopyxis alaskensis RB2256, complete genome | 8.84035e-66 | M01232:27:000000000-ARHCV:1:2101:25166:6939 |
| leptothrix cholodnii | gi\|170774137\|gb\|CP001013.1\| Leptothrix cholodnii SP-6, complete genome | 3.23695e-16 | M01232:27:000000000-ARHCV:1:2116:27914:6609 |
| sulfuritalea hydrogenivorans | gi\|572099409\|dbj\|AP012547.1\| Sulfuritalea hydrogenivorans sk43H DNA, complete genome | 1.94106e-43 | M01232:27:000000000-ARHCV:1:2105:7803:9041 |
| mycobacterium avium | gi\|756081930\|gb\|CP010114.1\| Mycobacterium avium subsp. paratuberculosis strain E93, complete genome | 8.41406e-92 | M01232:27:000000000-ARHCV:1:2102:20552:6368 |
| erwinia billingiae | gi\|299060424\|emb\|FP236843.1\| Erwinia billingiae strain Eb661 complete chromosome | 2.52783e-11 | M01232:27:000000000-ARHCV:1:2109:18737:20128 |
| [cellvibrio] gilvus | gi\|336102715\|gb\|CP002665.1\| [Cellvibrio] gilvus ATCC 13127, complete genome | 0.00205409 | M01232:27:000000000-ARHCV:1:2104:11586:10716 |
| saccharothrix espanaensis | gi\|407879691\|emb\|HE804045.1\| Saccharothrix espanaensis DSM 44229 complete genome | 1.09512e-15 | M01232:27:000000000-ARHCV:1:2110:6571:18768 |
| atopobium parvulum | gi\|257472321\|gb\|CP001721.1\| Atopobium parvulum DSM 20469, complete genome | 0.000138419 | M01232:27:000000000-ARHCV:1:2105:28946:15532 |
| allochromatium vinosum | gi\|288895136\|gb\|CP001896.1\| Allochromatium vinosum DSM 180, complete genome | 7.89157e-13 | M01232:27:000000000-ARHCV:1:1102:23578:9653 |
| bordetella bronchiseptica | gi\|408445111\|emb\|HE965807.1\| Bordetella bronchiseptica MO149 complete genome | 3.81172e-49 | M01232:27:000000000-ARHCV:1:2107:13934:9318 |

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|---|---|---|---|
| uncultured bacteroidales | gi\|190402431\|gb\|EU573793.1\| Uncultured Bacteroidales bacterium clone CA7 16S ribosomal RNA gene, partial sequence | 1.3305e-129 | M01232:27:000000000-ARHCV:1:1110:22297:15616 |
| kribbella flavida | gi\|283807292\|gb\|CP001736.1\| Kribbella flavida DSM 17836, complete genome | 6.70432e-13 | M01232:27:000000000-ARHCV:1:1117:20290:3234 |
| methanoculleus marisnigri | gi\|125860746\|gb\|CP000562.1\| Methanoculleus marisnigri JR1, complete genome | 4.95335e-41 | M01232:27:000000000-ARHCV:1:2116:25944:17462 |
| rhizobium leguminosarum | gi\|1046228\|gb\|U23471.1\|RLU23471 Rhizobium leguminosarum biovar phaseoli ABC-type permease homolog and sigma-54 factor (rpoN) genes, complete cds and probable sigma-54 modulation protein, partial cds | 1.15181e-50 | M01232:27:000000000-ARHCV:1:1105:11292:1992 |
| bacillus cereus | gi\|753605351\|gb\|CP009628.1\| Bacillus cereus ATCC 4342, complete genome | 2.97594e-70 | M01232:27:000000000-ARHCV:1:2111:6254:22151 |
| streptomyces davawensis | gi\|408526205\|emb\|HE971709.1\| Streptomyces davawensis strain JCM 4913 complete genome | 3.56503e-39 | M01232:27:000000000-ARHCV:1:1113:19103:18000 |
| haliangium ochraceum | gi\|262076673\|gb\|CP001804.1\| Haliangium ochraceum DSM 14365, complete genome | 5.51264e-29 | M01232:27:000000000-ARHCV:1:1104:23605:14910 |
| ralstonia eutropha | gi\|72120635\|gb\|CP000091.1\| Ralstonia eutropha JMP134 chromosome 2, complete sequence | 4.35294e-15 | M01232:27:000000000-ARHCV:1:2114:17674:8865 |
| mesorhizobium loti | gi\|47118328\|dbj\|BA000012.4\| Mesorhizobium loti MAFF303099 DNA, complete genome | 1.45486e-23 | M01232:27:000000000-ARHCV:1:2105:18426:11876 |
| cellulomonas fimi | gi\|332337569\|gb\|CP002666.1\| Cellulomonas fimi ATCC 484, complete genome | 0.00131887 | M01232:27:000000000-ARHCV:1:1117:24990:18977 |
| magnetospirillum magneticum | gi\|82943940\|dbj\|AP007255.1\| Magnetospirillum magneticum AMB-1 DNA, complete genome | 1.13366e-60 | M01232:27:000000000-ARHCV:1:1115:18553:10625 |
| dechlorosoma suillum | gi\|359353254\|gb\|CP003153.1\| Dechlorosoma suillum PS, complete genome | 4.45057e-07 | M01232:27:000000000-ARHCV:1:2107:26955:20459 |
| geobacter sp. | gi\|320123932\|gb\|CP002479.1\| Geobacter sp. M18, complete genome | 2.56478e-27 | M01232:27:000000000-ARHCV:1:2103:9667:14013 |
| octadecabacter arcticus | gi\|476512267\|gb\|CP003742.1\| Octadecabacter arcticus 238, complete genome | 5.36085e-42 | M01232:27:000000000-ARHCV:1:1110:23158:16476 |

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|---|---|---|---|
| pseudomonas trivialis | gi\|902687210\|gb\|CP011507.1\| Pseudomonas trivialis strain IHBB745, complete genome | 1.83699e-18 | M01232:27:000000000-ARHCV:1:1103:2459:10491 |
| nitratireductor aquibiodomus | gi\|332377416\|gb\|JF274966.1\| Nitratireductor aquibiodomus strain NL31 dissimilatory membrane-bound nitrate reductase (narG) gene, partial cds | 1.9003e-58 | M01232:27:000000000-ARHCV:1:1116:6613:15899 |
| xanthomonas citri | gi\|833359322\|gb\|CP011827.1\| Xanthomonas citri pv. citri strain jx-6, complete genome | 1.9107e-48 | M01232:27:000000000-ARHCV:1:1102:14630:7811 |
| streptomyces hygroscopicus | gi\|451789617\|gb\|CP003720.1\| Streptomyces hygroscopicus subsp. jinggangensis TL01, complete genome | 5.96556e-37 | M01232:27:000000000-ARHCV:1:1103:20628:8092 |
| stenotrophomonas maltophilia | gi\|922795094\|gb\|CP011305.1\| Stenotrophomonas maltophilia strain ISMMS2, complete genome | 3.69141e-23 | M01232:27:000000000-ARHCV:1:1112:6437:21677 |
| burkholderia thailandensis | gi\|772989008\|gb\|CP009601.1\| Burkholderia thailandensis 2002721643 chromosome I, complete sequence | 2.03255e-08 | M01232:27:000000000-ARHCV:1:1105:24308:11902 |
| pandoraea thiooxydans | gi\|827097060\|gb\|CP011568.1\| Pandoraea thiooxydans strain DSM 25325, complete genome | 2.03961e-08 | M01232:27:000000000-ARHCV:1:1108:8706:15256 |
| uncultured rumen | gi\|896685010\|gb\|KR068150.1\| Uncultured rumen bacterium clone YAK-E63 16S ribosomal RNA gene, partial sequence | 6.35686e-108 | M01232:27:000000000-ARHCV:1:2103:17111:16393 |
| pimelobacter simplex | gi\|723622094\|gb\|CP009896.1\| Pimelobacter simplex strain VKM Ac-2033D, complete genome | 2.13239e-07 | M01232:27:000000000-ARHCV:1:1105:12914:13879 |
| burkholderia ubonensis | gi\|772924293\|gb\|CP009488.1\| Burkholderia ubonensis MSMB22 chromosome I, complete sequence | 4.38385e-10 | M01232:27:000000000-ARHCV:1:1110:23201:10919 |
| streptomyces albulus | gi\|785755366\|gb\|CP006871.1\| Streptomyces albulus ZPM, complete genome | 5.39687e-44 | M01232:27:000000000-ARHCV:1:2118:24507:20131 |
| cupriavidus taiwanensis | gi\|170937689\|emb\|CU633749.1\| Cupriavidus taiwanensis str. LMG19424 chromosome 1, complete genome | 4.32436e-11 | M01232:27:000000000-ARHCV:1:2114:23427:18439 |
| sphingobium sp. | gi\|764452294\|gb\|CP010954.1\| Sphingobium sp. YBL2, complete genome | 1.17502e-17 | M01232:27:000000000-ARHCV:1:2114:18655:19901 |

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|---|---|---|---|
| methylobacterium sp. | gi\|168192641\|gb\|CP000943.1\| Methylobacterium sp. 4-46, complete genome | 2.63383e-07 | M01232:27:000000000-ARHCV:1:1112:22935:5765 |
| salinispora arenicola | gi\|157914509\|gb\|CP000850.1\| Salinispora arenicola CNS-205, complete genome | 7.81274e-31 | M01232:27:000000000-ARHCV:1:2109:20826:22453 |
| methylobacterium radiotolerans | gi\|170652972\|gb\|CP001001.1\| Methylobacterium radiotolerans JCM 2831, complete genome | 1.93434e-43 | M01232:27:000000000-ARHCV:1:2116:24999:22371 |
| nippostrongylus brasiliensis | gi\|687008631\|emb\|LM437148.1\| Nippostrongylus brasiliensis genome assembly N_brasiliensis_RM07_v1_5_4 ,scaffold NBR_scaffold0003529 | 8.62514e-06 | M01232:27:000000000-ARHCV:1:2105:25956:7326 |
| mycobacterium intracellulare | gi\|698183444\|gb\|CP009499.1\| Mycobacterium intracellulare 1956, complete genome | 7.50532e-15 | M01232:27:000000000-ARHCV:1:1116:14849:10249 |
| pseudomonas knackmussii | gi\|631776901\|emb\|HG322950.1\| Pseudomonas knackmussii B13 complete genome | 1.8736e-68 | M01232:27:000000000-ARHCV:1:1107:21730:5046 |
| sideroxydans lithotrophicus | gi\|291582584\|gb\|CP001965.1\| Sideroxydans lithotrophicus ES-1, complete genome | 1.14782e-50 | M01232:27:000000000-ARHCV:1:1111:24487:17948 |
| rhizobium gallicum | gi\|745836364\|gb\|CP006880.1\| Rhizobium gallicum bv. gallicum R602 plasmid pRgalR602c, complete sequence | 0.00292793 | M01232:27:000000000-ARHCV:1:1110:6812:4599 |
| cupriavidus metallidurans | gi\|288237308\|gb\|CP000353.2\| Cupriavidus metallidurans CH34 megaplasmid, complete sequence | 1.1119e-08 | M01232:27:000000000-ARHCV:1:1115:18706:14368 |
| cupriavidus basilensis | gi\|752314807\|gb\|CP010536.1\| Cupriavidus basilensis strain 4G11 chromosome main, complete sequence | 7.33573e-08 | M01232:27:000000000-ARHCV:1:2102:22457:2529 |
| rhodococcus pyridinivorans | gi\|568232286\|gb\|CP006996.1\| Rhodococcus pyridinivorans SB3094, complete genome | 3.01368e-11 | M01232:27:000000000-ARHCV:1:1111:9939:20042 |
| pseudomonas pseudoalcaligenes | gi\|652789639\|emb\|LK391695.1\| Pseudomonas pseudoalcaligenes genome assembly Ppseudo_Pac ,chromosome : I | 1.16584e-94 | M01232:27:000000000-ARHCV:1:1104:16628:10519 |
| amycolatopsis japonica | gi\|667678325\|gb\|CP008953.1\| Amycolatopsis japonica strain MG417-CF17, complete genome | 9.93833e-21 | M01232:27:000000000-ARHCV:1:2117:19344:11105 |

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|---|---|---|---|
| thermobifida fusca | gi\|71914138\|gb\|CP000088.1\| Thermobifida fusca YX, complete genome | 9.33099e-121 | M01232:27:000000000-ARHCV:1:2113:6640:20406 |
| mesorhizobium opportunistum | gi\|336024847\|gb\|CP002279.1\| Mesorhizobium opportunistum WSM2075, complete genome | 5.57966e-42 | M01232:27:000000000-ARHCV:1:2102:26233:12910 |
| solibacter usitatus | gi\|116222307\|gb\|CP000473.1\| Solibacter usitatus Ellin6076, complete genome | 1.00508e-15 | M01232:27:000000000-ARHCV:1:1119:12416:20333 |
| elaeophora elaphi | gi\|666417843\|emb\|LL713108.1\| Elaeophora elaphi genome assembly E_elaphi ,scaffold EEL_contig0001020 | 1.11414e-33 | M01232:27:000000000-ARHCV:1:2119:15676:11874 |
| arthrobacter aurescens | gi\|119947346\|gb\|CP000474.1\| Arthrobacter aurescens TC1, complete genome | 4.92267e-109 | M01232:27:000000000-ARHCV:1:2105:16190:10344 |
| rubrobacter radiotolerans | gi\|627776062\|gb\|CP007514.1\| Rubrobacter radiotolerans strain RSPS-4, complete genome | 1.45783e-08 | M01232:27:000000000-ARHCV:1:2106:23219:3022 |
| sinorhizobium medicae | gi\|150030273\|gb\|CP000739.1\| Sinorhizobium medicae WSM419 plasmid pSMED01, complete genome | 2.53331e-16 | M01232:27:000000000-ARHCV:1:2117:26095:14752 |
| neofusicoccum parvum | gi\|615420284\|ref\|XM_007586382.1\| Neofusicoccum parvum UCRNP2 putative rieske 2fe-2s family protein mRNA | 0.00175557 | M01232:27:000000000-ARHCV:1:1108:28432:17158 |
| african horsesickness | gi\|390196985\|gb\|JQ742009.1\| African horsesickness virus segment 4, complete sequence | 6.17794e-60 | M01232:27:000000000-ARHCV:1:2106:21210:12183 |
| altererythrobacter atlanticus | gi\|918027538\|gb\|CP011452.2\| Altererythrobacter atlanticus strain 26DY36, complete genome | 4.17985e-40 | M01232:27:000000000-ARHCV:1:2117:16217:6335 |
| halomonas sp. | gi\|802125597\|emb\|LN813019.1\| Halomonas sp. R57-5 genome assembly HalomonasR57-5, chromosome : I | 4.14207e-15 | M01232:27:000000000-ARHCV:1:1104:12137:16363 |
| agrobacterium radiobacter | gi\|221721649\|gb\|CP000628.1\| Agrobacterium radiobacter K84 chromosome 1, complete sequence | 2.83504e-20 | M01232:27:000000000-ARHCV:1:2101:25036:18977 |
| chromobacterium violaceum | gi\|34105712\|gb\|AE016825.1\| Chromobacterium violaceum ATCC 12472, complete genome | 4.4655e-09 | M01232:27:000000000-ARHCV:1:2105:28323:17748 |
| rhodovulum sulfidophilum | gi\|770474292\|dbj\|AP014800.1\| Rhodovulum sulfidophilum DNA, complete genome, strain: DSM 2351 | 2.18973e-22 | M01232:27:000000000-ARHCV:1:1106:10045:23178 |

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|---|---|---|---|
| pseudomonas sp. | gi\|771846103\|dbj\|AP014628.1\| Pseudomonas sp. St29 DNA, complete genome | 4.35294e-15 | M01232:27:000000000-ARHCV:1:1102:5970:21853 |
| pseudomonas fluorescens | gi\|359757955\|gb\|CP003150.1\| Pseudomonas fluorescens F113, complete genome | 4.20166e-40 | M01232:27:000000000-ARHCV:1:2108:9917:12849 |
| leisingera methylohalidivorans | gi\|568223374\|gb\|CP006773.1\| Leisingera methylohalidivorans DSM 14336 strain MB2, DSM 14336, complete genome | 8.53428e-52 | M01232:27:000000000-ARHCV:1:2118:12742:7141 |
| porphyrobacter cryptus | gi\|330414323\|gb\|JF459884.1\| Porphyrobacter cryptus strain T12AT7 ATP synthase beta subunit gene, partial cds >gi\|330414325\|gb\|JF459885.1\| Porphyrobacter cryptus strain T12AT8 ATP synthase beta subunit gene, partial cds | 5.01233e-21 | M01232:27:000000000-ARHCV:1:2114:11168:11627 |
| burkholderia phenoliruptrix | gi\|407233956\|gb\|CP003863.1\| Burkholderia phenoliruptrix BR3459a chromosome 1, complete sequence | 1.92316e-08 | M01232:27:000000000-ARHCV:1:2108:26617:20164 |
| chromohalobacter salexigens | gi\|91795226\|gb\|CP000285.1\| Chromohalobacter salexigens DSM 3043, complete genome | 4.06676e-15 | M01232:27:000000000-ARHCV:1:2103:15254:2728 |
| bradyrhizobium diazoefficiens | gi\|806922190\|dbj\|AP014685.1\| Bradyrhizobium diazoefficiens DNA, complete genome, strain: NK6 | 8.79982e-07 | M01232:27:000000000-ARHCV:1:1104:5506:5005 |
| frankia sp. | gi\|311225233\|gb\|CP002299.1\| Frankia sp. EuI1c, complete genome | 2.1532e-26 | M01232:27:000000000-ARHCV:1:1117:25315:15563 |
| asticcacaulis excentricus | gi\|315417394\|gb\|CP002396.1\| Asticcacaulis excentricus CB 48 chromosome 2, complete sequence | 2.47507e-16 | M01232:27:000000000-ARHCV:1:2110:17317:4828 |
| verrucosispora maris | gi\|328807854\|gb\|CP002638.1\| Verrucosispora maris AB-18-032, complete genome | 4.54965e-12 | M01232:27:000000000-ARHCV:1:1107:18156:2556 |
| methylobacterium aquaticum | gi\|760865928\|dbj\|AP014704.1\| Methylobacterium aquaticum DNA, complete genome, strain: MA-22A | 1.6202e-08 | M01232:27:000000000-ARHCV:1:2105:18753:11449 |
| mycobacterium tuberculosis | gi\|669088828\|gb\|CP008974.1\| Mycobacterium tuberculosis strain 0A115DS genome | 7.14507e-18 | M01232:27:000000000-ARHCV:1:1102:5213:19884 |
| pandoraea sputorum | gi\|743596961\|gb\|CP010431.1\| Pandoraea sputorum strain DSM 21091, complete genome | 1.18302e-10 | M01232:27:000000000-ARHCV:1:1105:16396:4158 |

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|---|---|---|---|
| burkholderia sp. | gi\|295438061\|gb\|CP002014.1\| Burkholderia sp. CCGE1002 chromosome 2, complete sequence | 0.0033115 | M01232:27:000000000-ARHCV:1:1115:14623:13429 |
| variovorax paradoxus | gi\|315593157\|gb\|CP002417.1\| Variovorax paradoxus EPS, complete genome | 2.94218e-106 | M01232:27:000000000-ARHCV:1:2111:22470:6718 |
| bifidobacterium bifidum | gi\|781872459\|dbj\|AP012323.1\| Bifidobacterium bifidum ATCC 29521 = JCM 1255 = DSM 20456 DNA, complete genome | 0.00484579 | M01232:27:000000000-ARHCV:1:1103:15341:6979 |
| mycobacterium kansasii | gi\|690330613\|gb\|CP009483.1\| Mycobacterium kansasii 824, complete genome | 1.51119e-39 | M01232:27:000000000-ARHCV:1:2113:25665:6259 |
| haemonchus placei | gi\|690066868\|emb\|LM585990.1\| Haemonchus placei genome assembly H_placei_MHpl1 ,scaffold HPLM_scaffold0002911 | 4.92589e-08 | M01232:27:000000000-ARHCV:1:1118:26799:21500 |
| corynebacterium casei | gi\|582015241\|gb\|CP004350.1\| Corynebacterium casei LMG S-19264, complete genome | 3.41298e-06 | M01232:27:000000000-ARHCV:1:1103:3360:13038 |
| macaca mulatta | gi\|154240759\|gb\|AC202678.6\| Macaca mulatta BAC CH250-38L21 () complete sequence | 0.000963379 | M01232:27:000000000-ARHCV:1:2115:24577:20135 |
| blastococcus saxobsidens | gi\|378781357\|emb\|FO117623.1\| Blastococcus saxobsidens DD2 complete genome | 5.43519e-39 | M01232:27:000000000-ARHCV:1:1110:9266:12693 |
| microlunatus phosphovorus | gi\|334683429\|dbj\|AP012204.1\| Microlunatus phosphovorus NM-1 DNA, complete genome | 8.70193e-67 | M01232:27:000000000-ARHCV:1:1103:17801:23141 |
| acetobacter pasteurianus | gi\|528530485\|emb\|HF677571.1\| Acetobacter pasteurianus 386B plasmid Apa386Bp1, complete sequence | 6.82345e-19 | M01232:27:000000000-ARHCV:1:2118:22414:22673 |
| mus musculus | gi\|30911140\|gb\|AC131325.8\| Mus musculus chromosome 17, clone RP23-431O10, complete sequence | 2.74304e-05 | M01232:27:000000000-ARHCV:1:1113:14090:12168 |
| achromobacter xylosoxidans | gi\|310757913\|gb\|CP002287.1\| Achromobacter xylosoxidans A8, complete genome | 2.4693e-24 | M01232:27:000000000-ARHCV:1:2102:20775:21595 |
| chelatococcus sp. | gi\|919436391\|gb\|CP012399.1\| Chelatococcus sp. CO-6 plasmid pCO-6, complete sequence | 9.24315e-12 | M01232:27:000000000-ARHCV:1:1103:13193:13935 |
| symbiobacterium thermophilum | gi\|51854827\|dbj\|AP006840.1\| Symbiobacterium thermophilum IAM 14863 DNA, complete genome | 4.29179e-25 | M01232:27:000000000-ARHCV:1:1104:6183:9327 |

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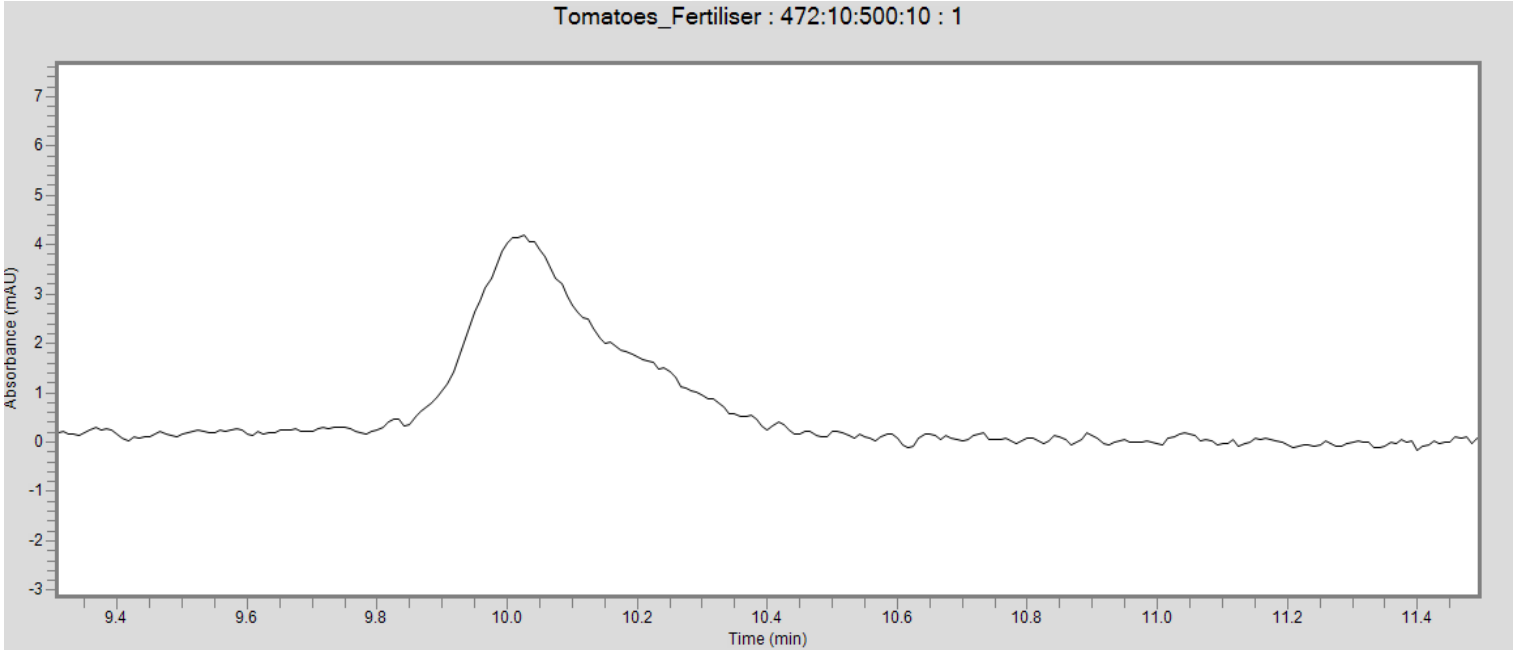
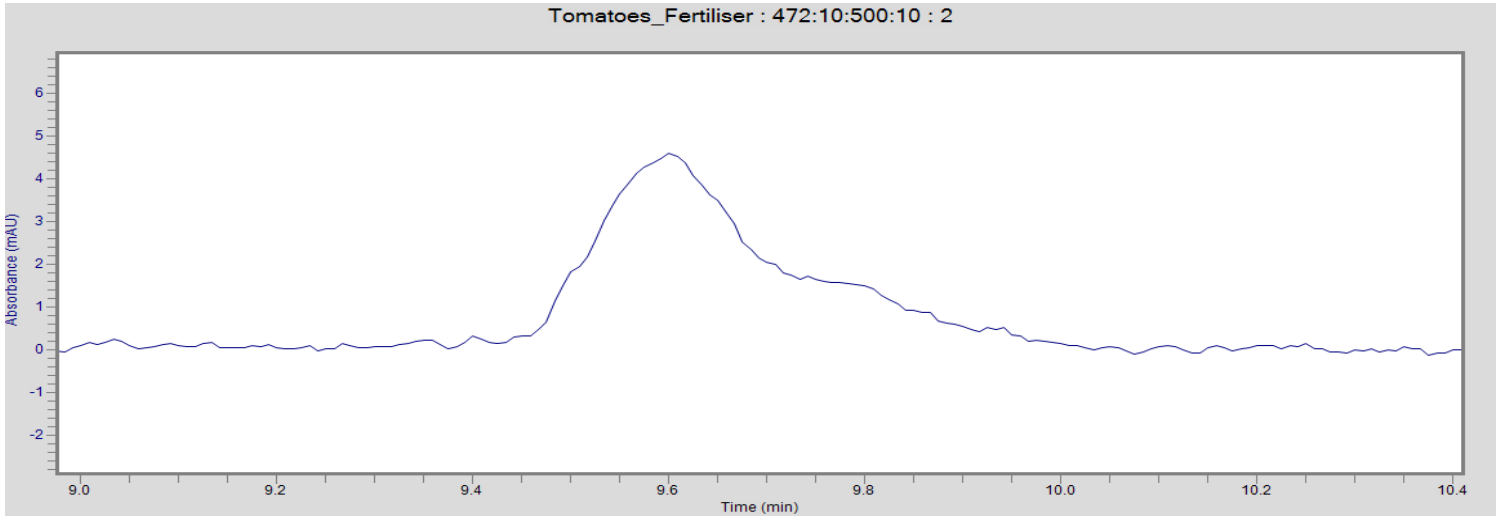
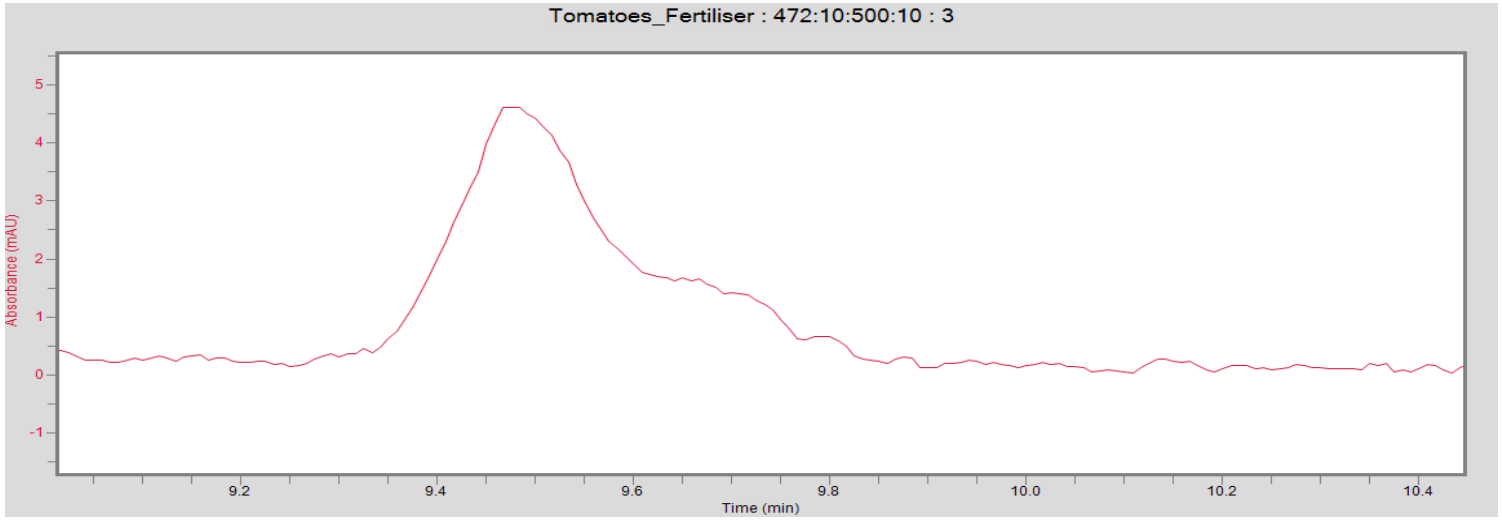
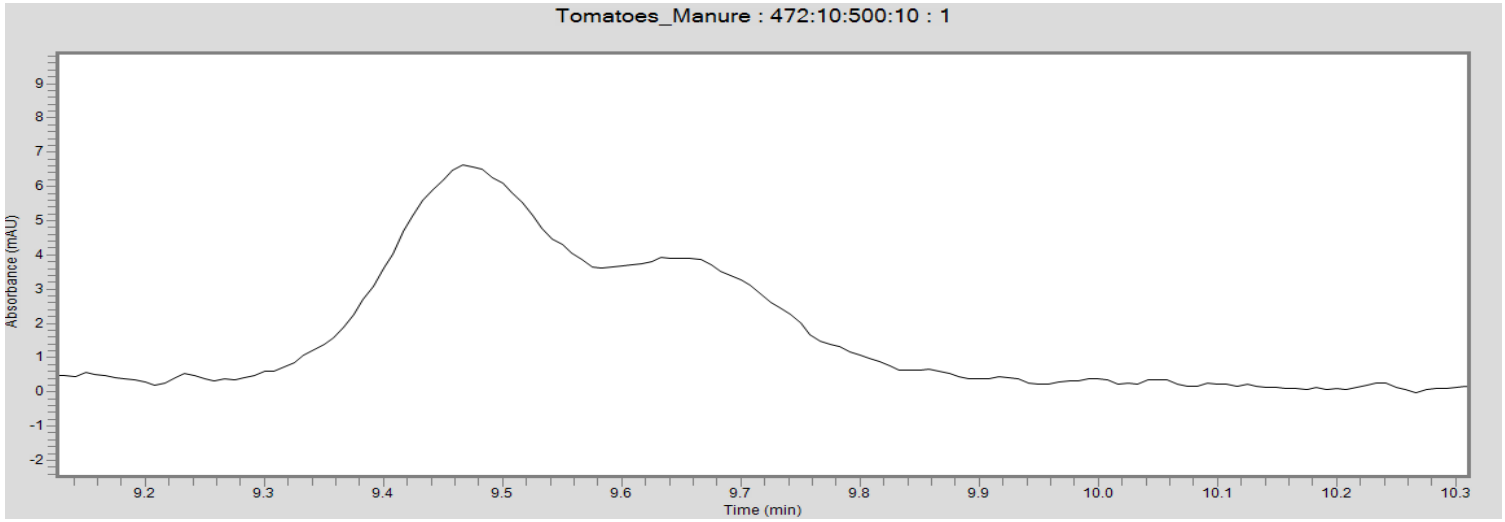
|---|---|---|---|
| pseudomonas syringae | gi\|908345318\|gb\|CP011972.1\| Pseudomonas syringae pv. actinidiae ICMP 18884, complete genome | 9.92936e-13 | M01232:27:000000000-ARHCV:1:2108:28894:16413 |
| colletotrichum graminicola | gi\|827059819\|ref\|XM_008093306.1\| Colletotrichum graminicola M1.001 hypothetical protein partial mRNA | 4.51729e-34 | M01232:27:000000000-ARHCV:1:2101:24779:5792 |
| photobacterium gaetbulicola | gi\|764066760\|gb\|CP005974.1\| Photobacterium gaetbulicola Gung47 chromosome 2, complete sequence | 3.09471e-39 | M01232:27:000000000-ARHCV:1:1105:26315:8733 |

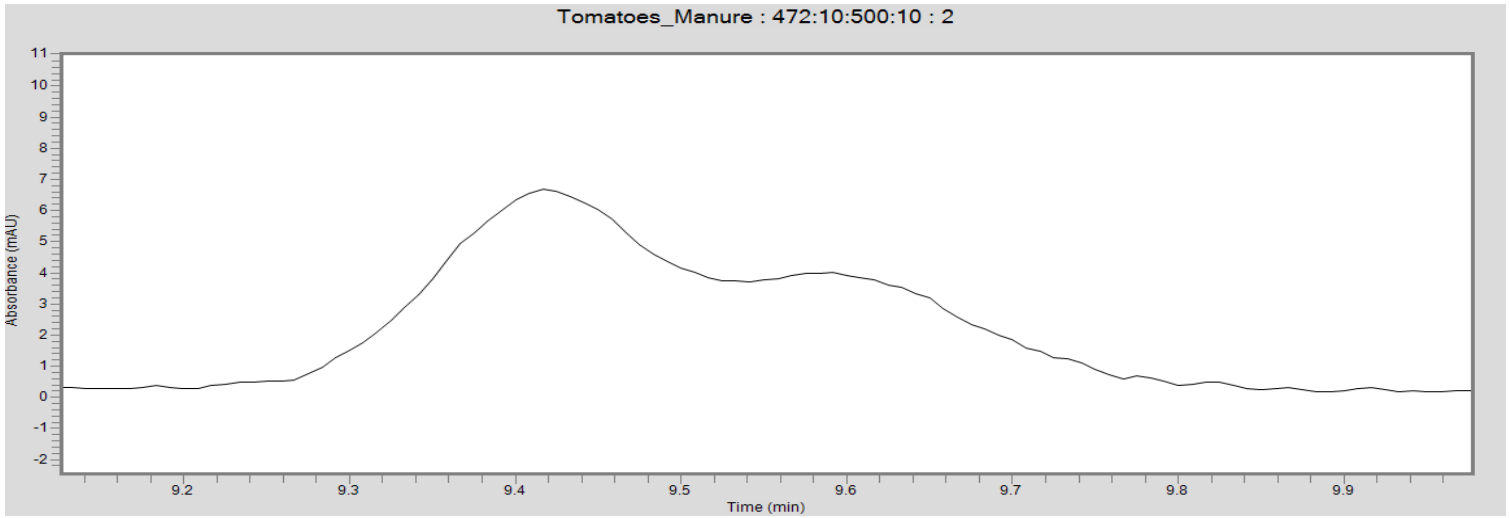
## APPENDIX 2: Chromatograms generated from HPLC analysis of different lycopene standard concentrations and lycopene analysis of tomato fruits from Goat Manure Derived Hydroponic Nutrient Solution (GMDHNS) and Commercial Hydroponic Nutrient Solution (CHNS).

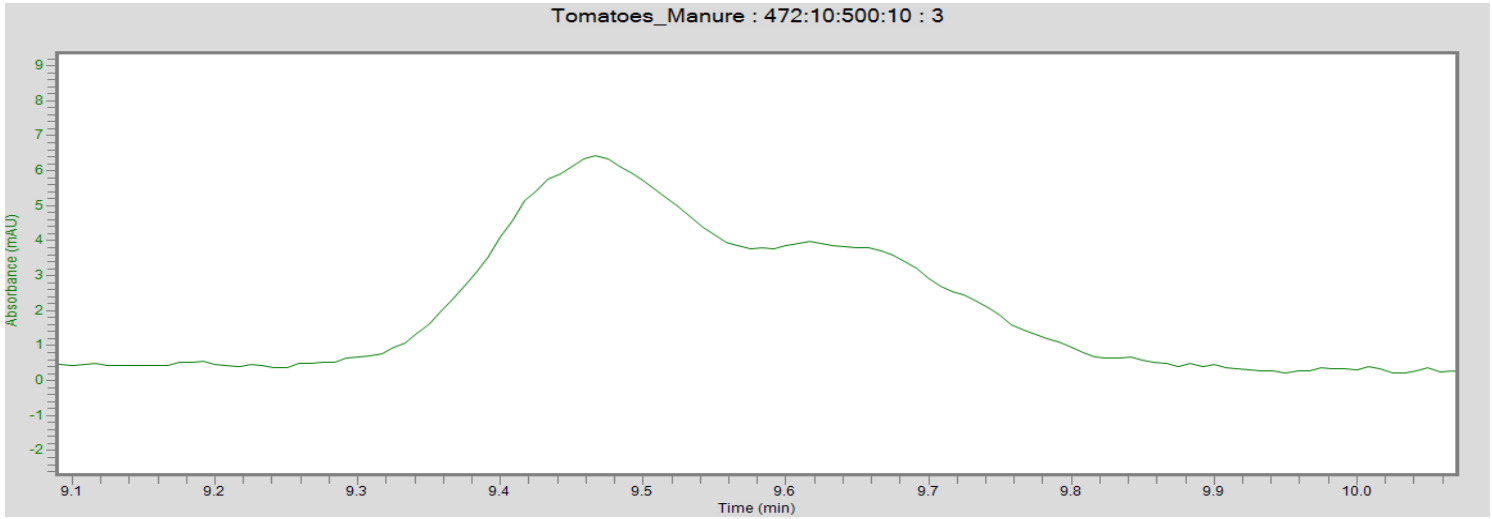


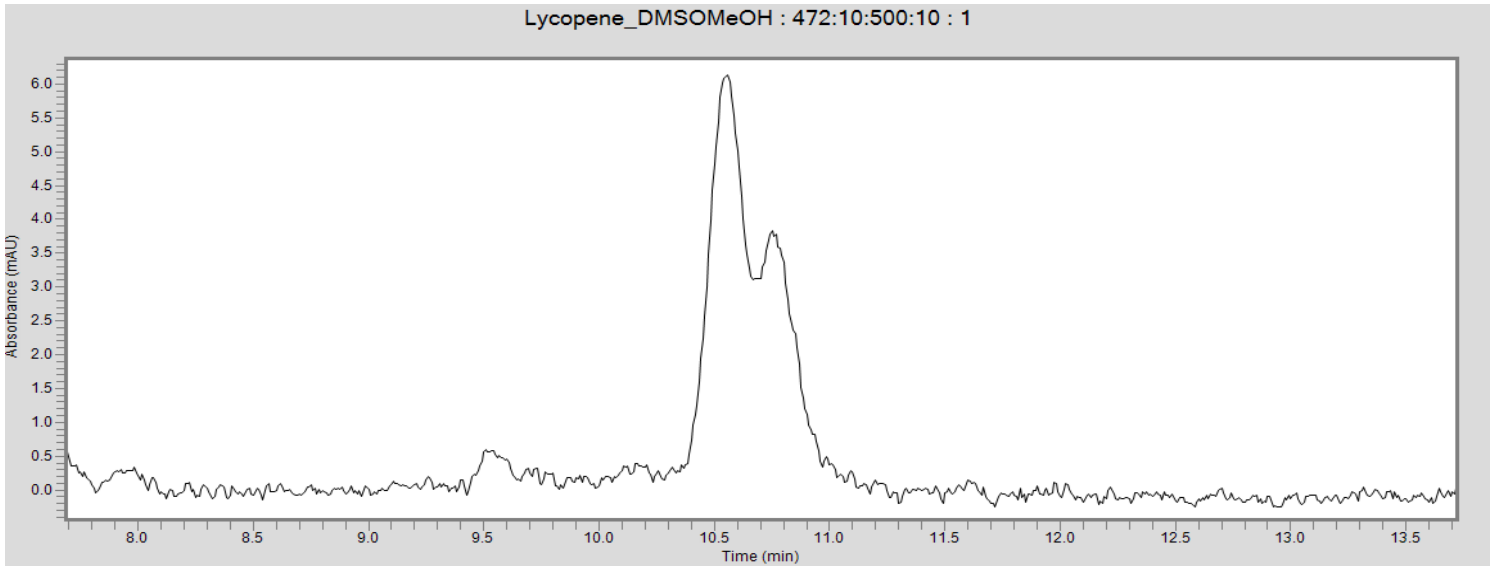

Tomatoes_Fertiliser : 472:10:500:10 : 2
Absorbance (mAU)
6
5
4
3
2
1
0
-1
-2
9.0
9.2
9.4
9.6
9.8
10.0
10.2
10.4
Time (min)

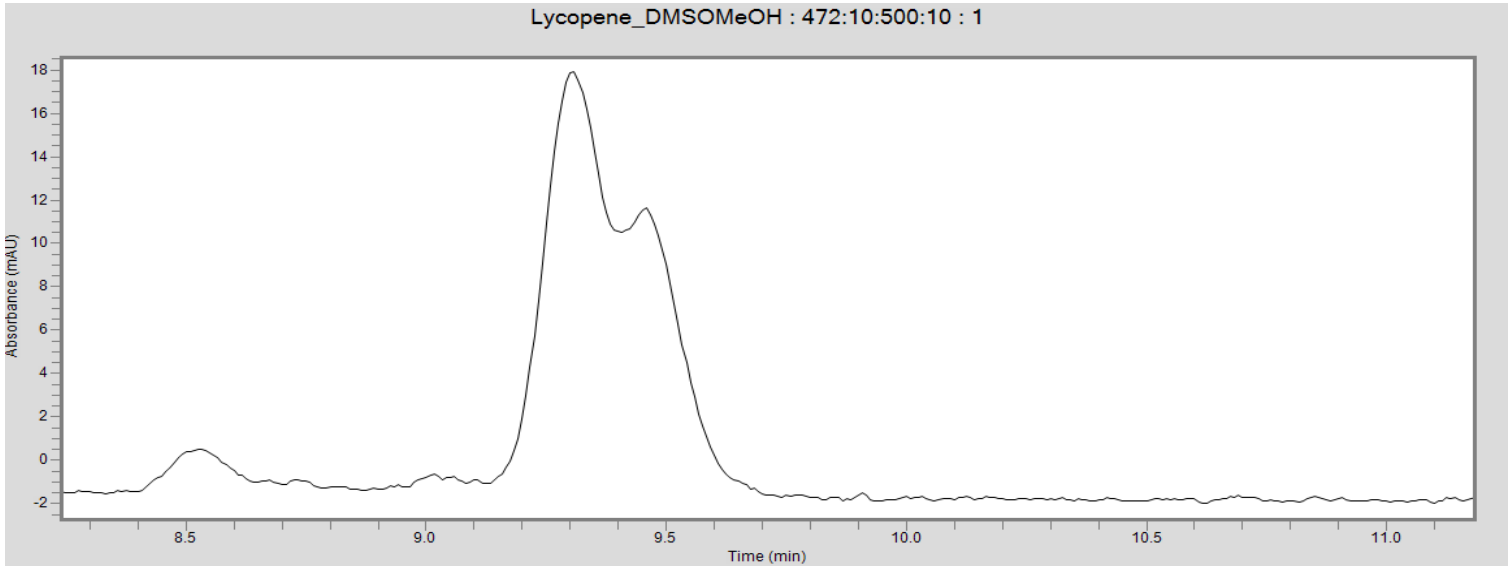

Tomatoes_Fertiliser : 472:10:500:10 : 3
Absorbance (mAU)
5
4
3
2
1
0
-1
9.2
9.4
9.6
9.8
10.0
10.2
10.4
Time (min)

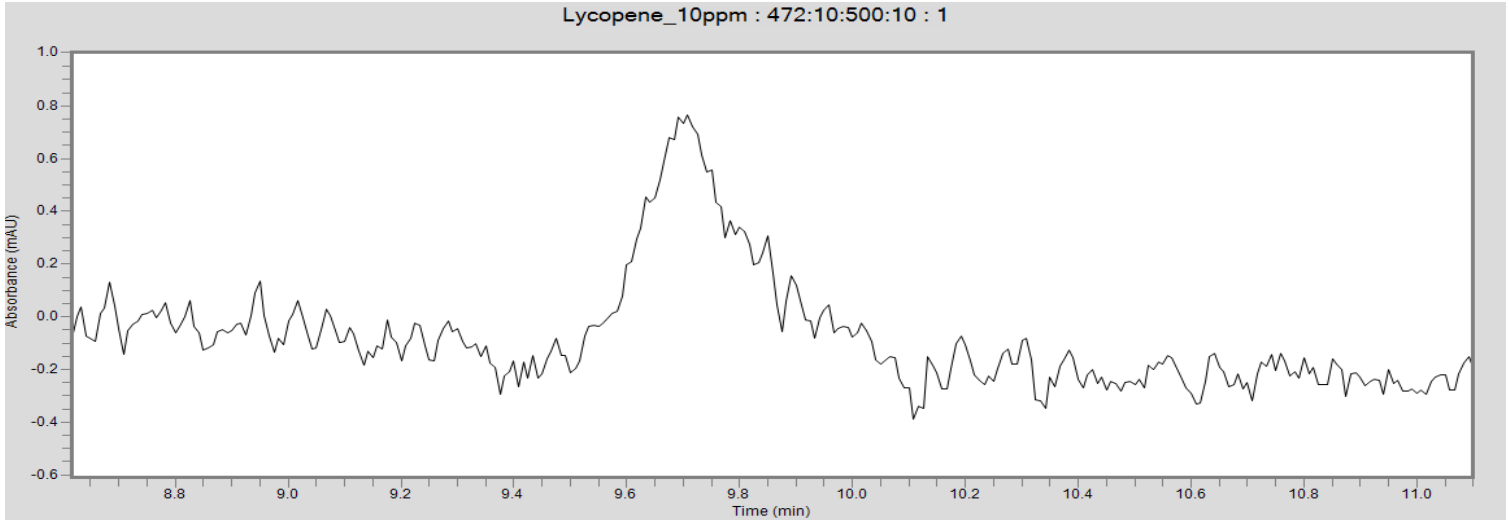

Tomatoes_Manure : 472:10:500:10 : 1
Absorbance (mAU)
Time (min)

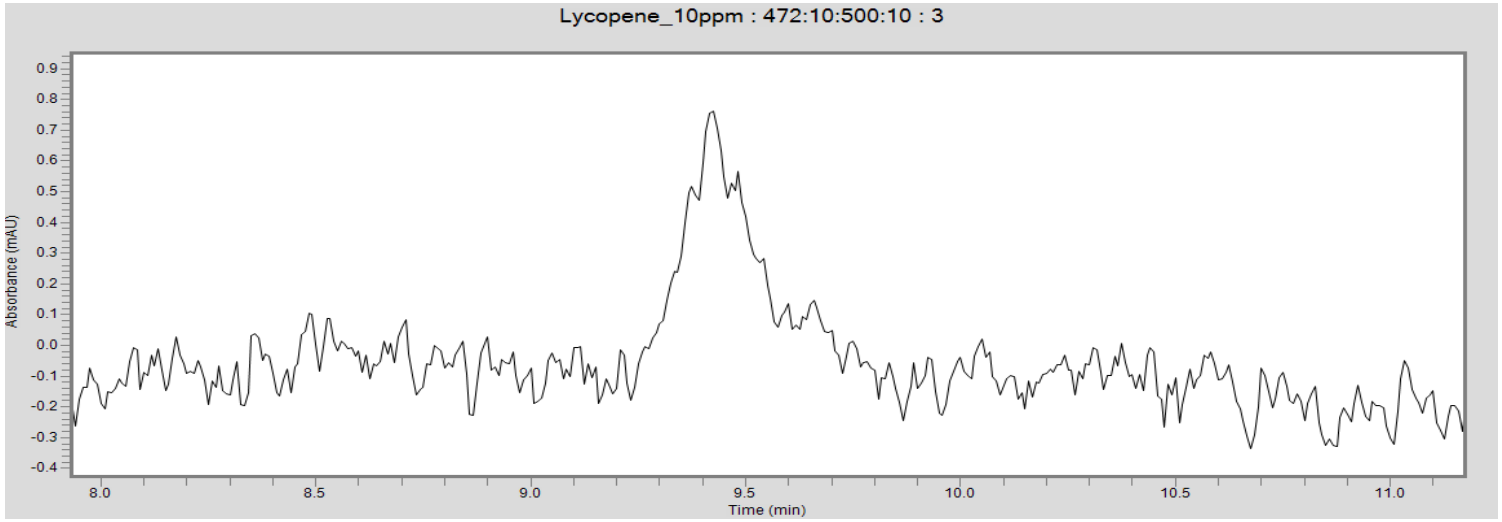

Tomatoes_Manure : 472:10:500:10 : 2
Absorbance (mAU)
Time (min)

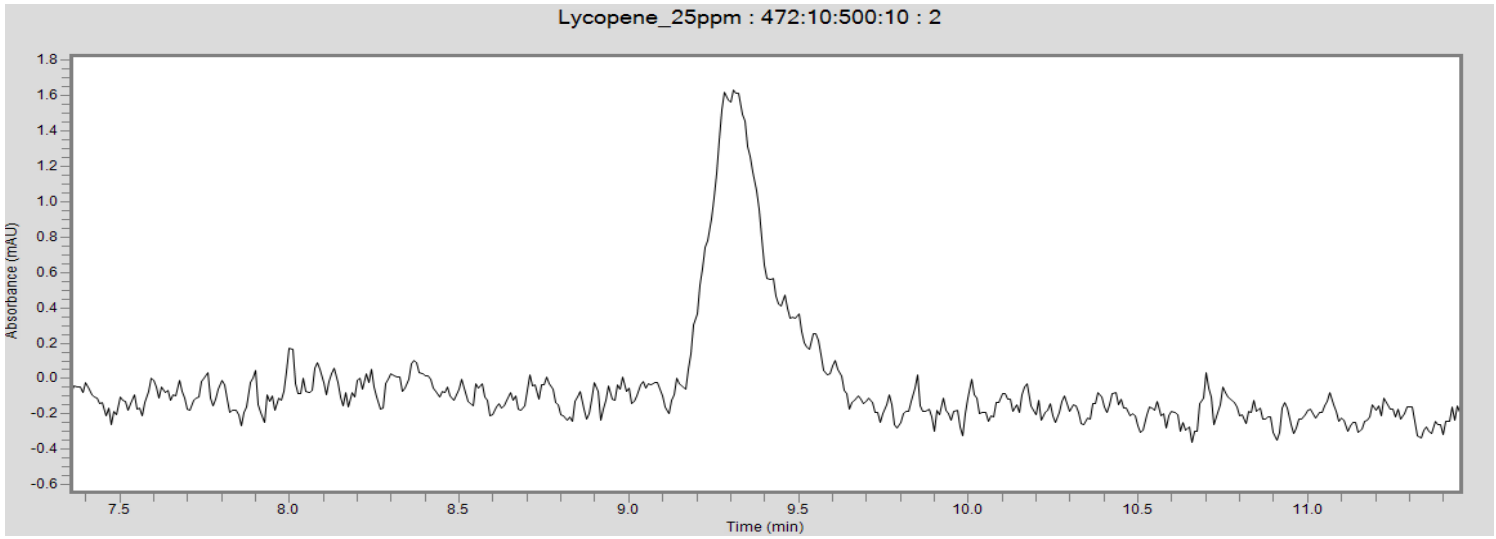

Tomatoes_Manure : 472:10:500:10 : 3
Absorbance (mAU)
Time (min)

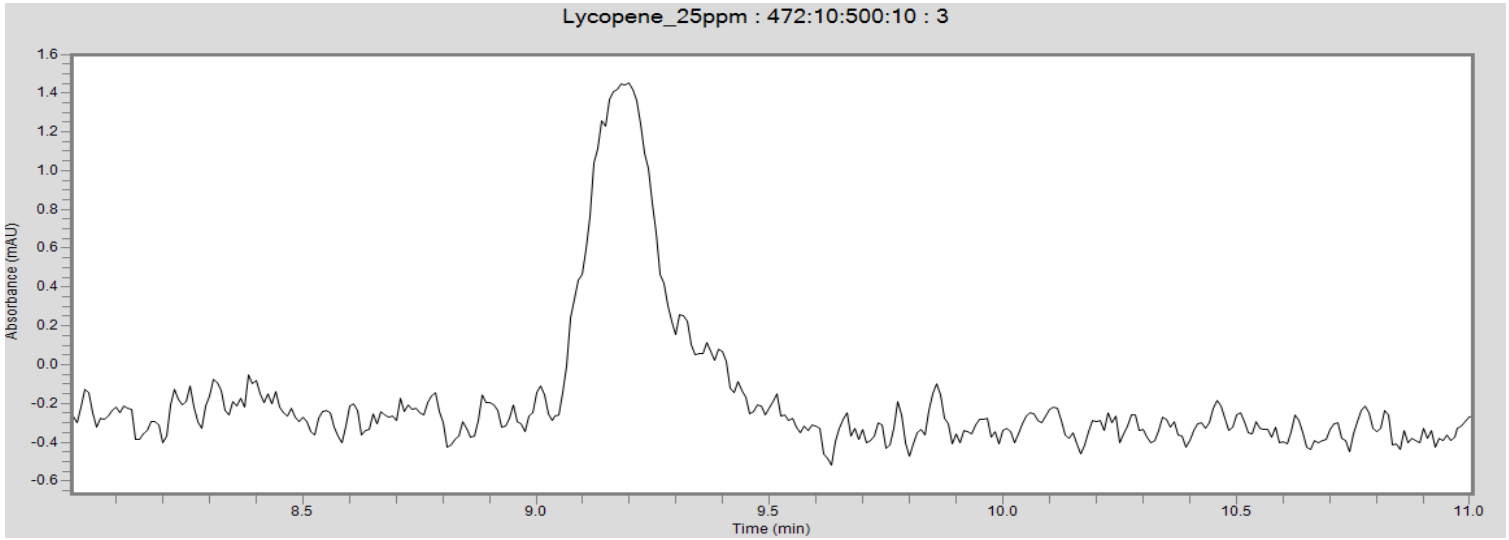

Lycopene_DMSOMeOH : 472:10:500:10 : 1
Absorbance (mAU)
Time (min)

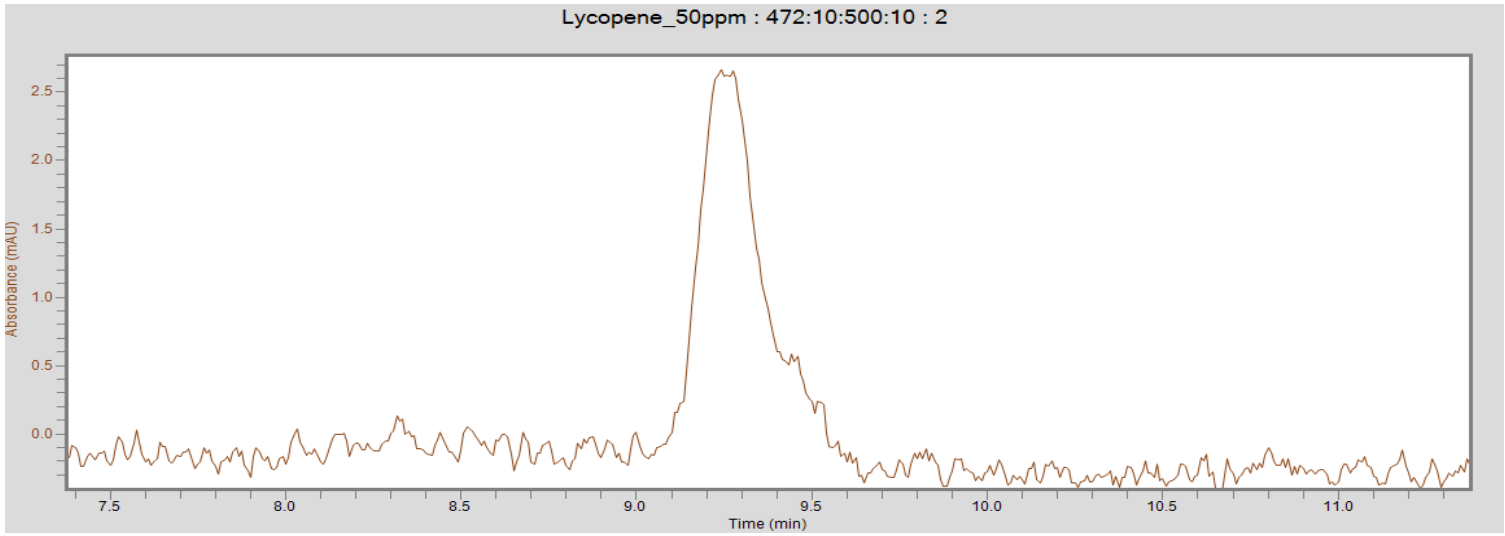

Lycopene_DMSOMeOH : 472:10:500:10 : 1
Absorbance (mAU)
18
16
14
12
10
8
6
4
2
0
-2
8.5
9.0
9.5
10.0
10.5
11.0
Time (min)

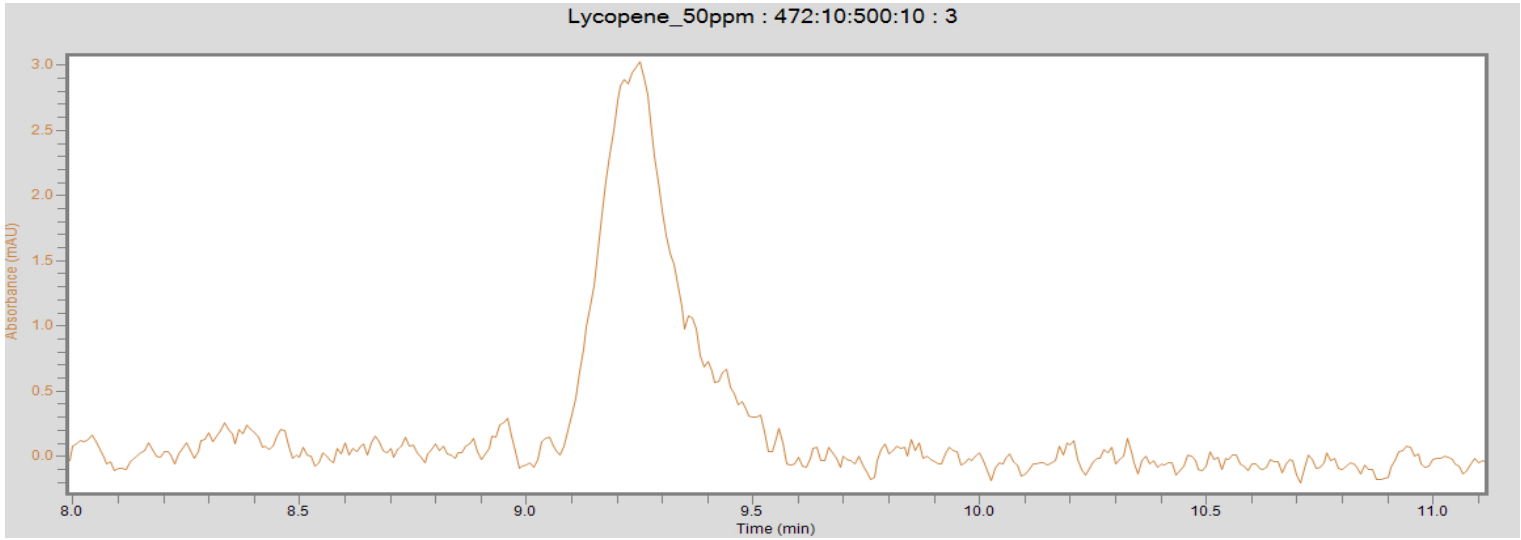

Lycopene_10ppm : 472:10:500:10 : 1
Absorbance (mAU)
1.0
0.8
0.6
0.4
0.2
0.0
-0.2
-0.4
-0.6
8.8
9.0
9.2
9.4
9.6
9.8
10.0
10.2
10.4
10.6
10.8
11.0
Time (min)

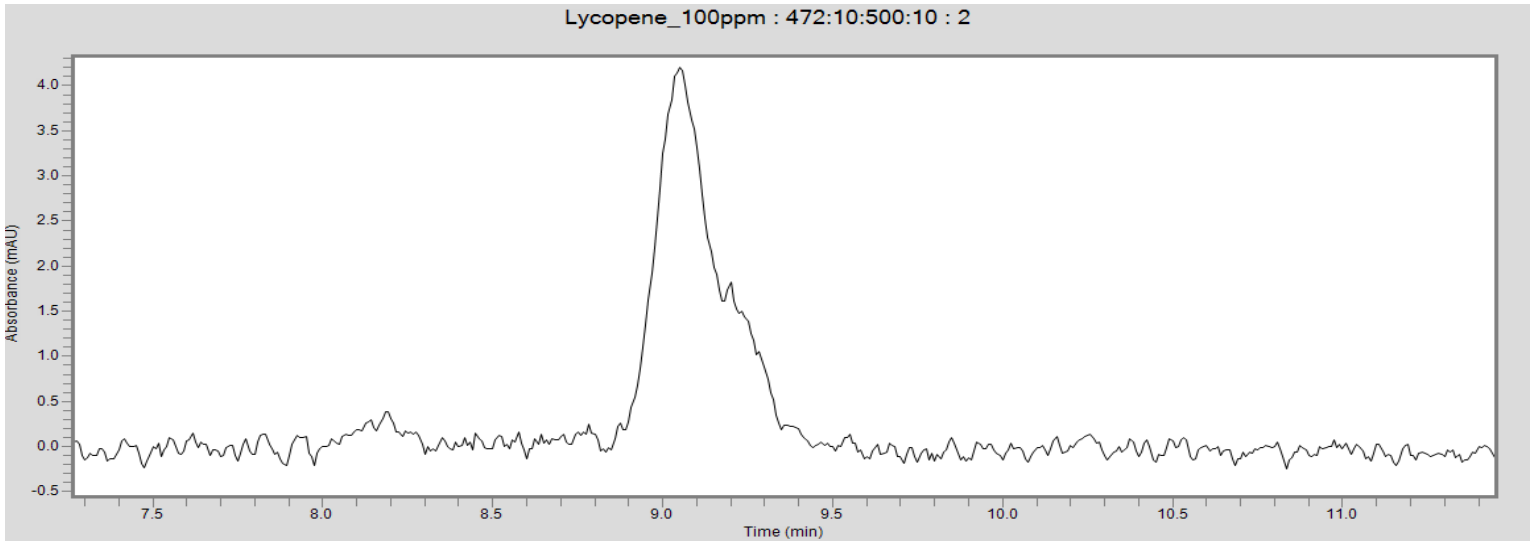

Lycopene_10ppm : 472:10:500:10 : 3
Absorbance (mAU)
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0.0
-0.1
-0.2
-0.3
-0.4
8.0
8.5
9.0
9.5
10.0
10.5
11.0
Time (min)

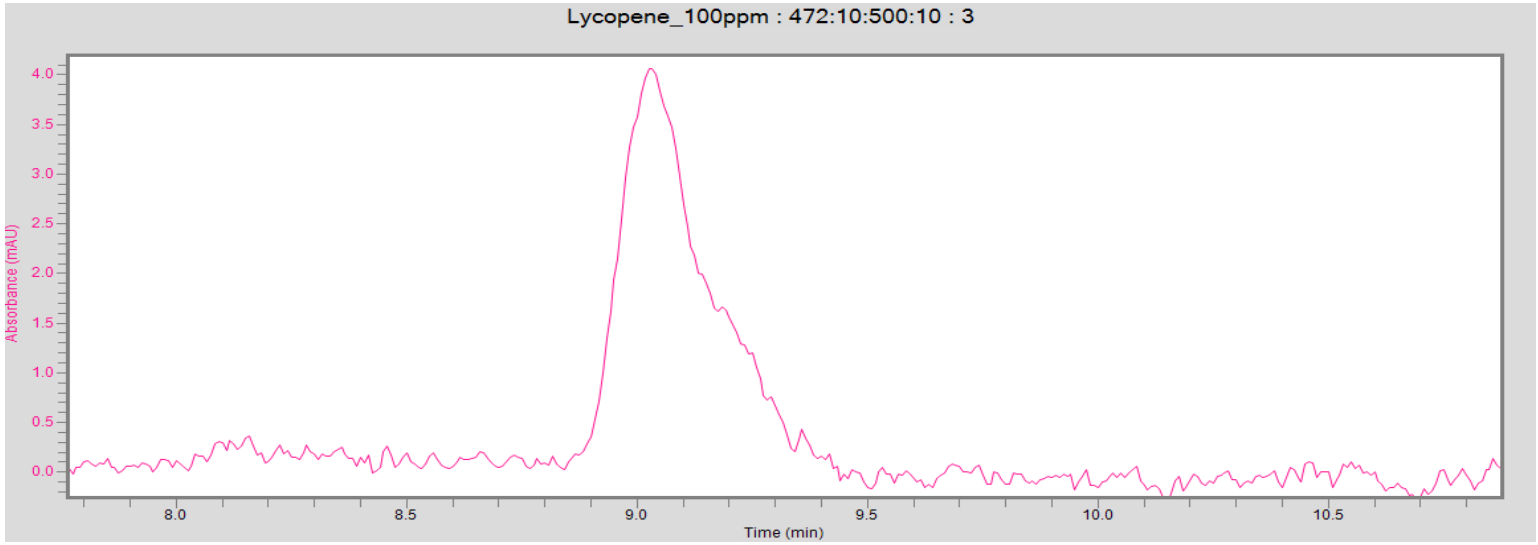

Lycopene_25ppm : 472:10:500:10 : 2
Absorbance (mAU)
1.8
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
-0.2
-0.4
-0.6
7.5
8.0
8.5
9.0
9.5
10.0
10.5
11.0
Time (min)

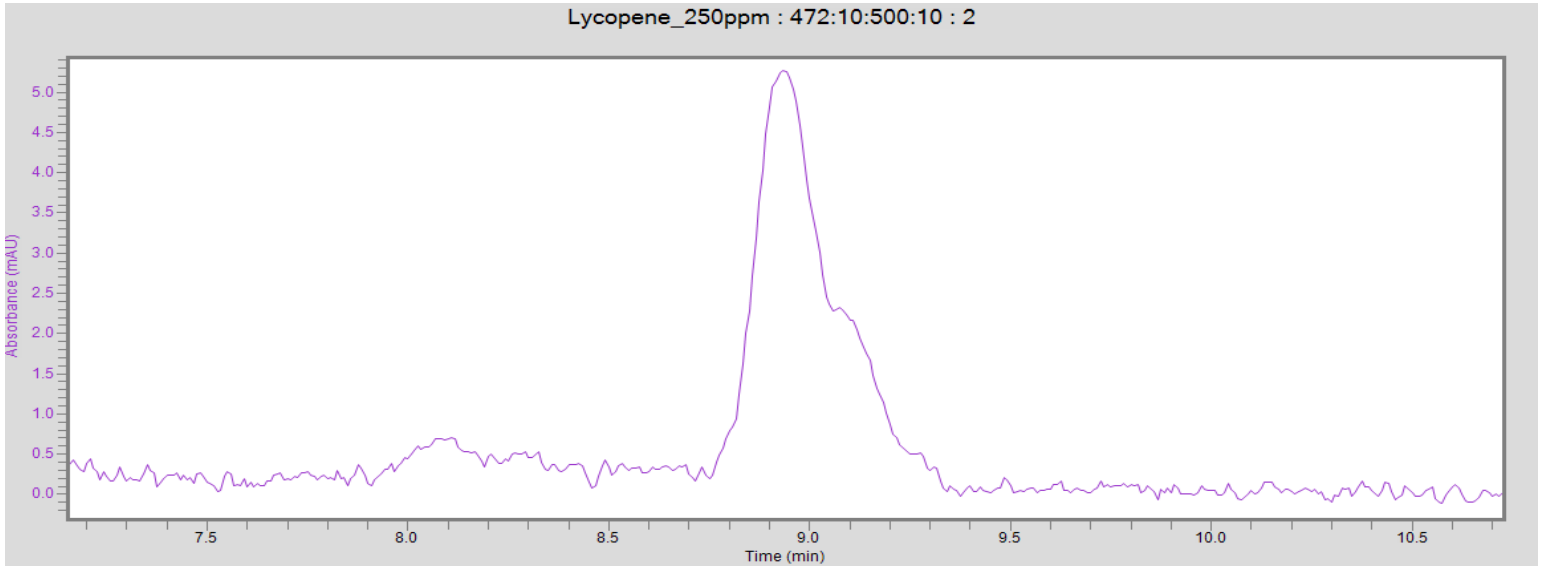

Lycopene_25ppm : 472:10:500:10 : 3
Absorbance (mAU)
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
-0.2
-0.4
-0.6
8.5
9.0
9.5
10.0
10.5
11.0
Time (min)

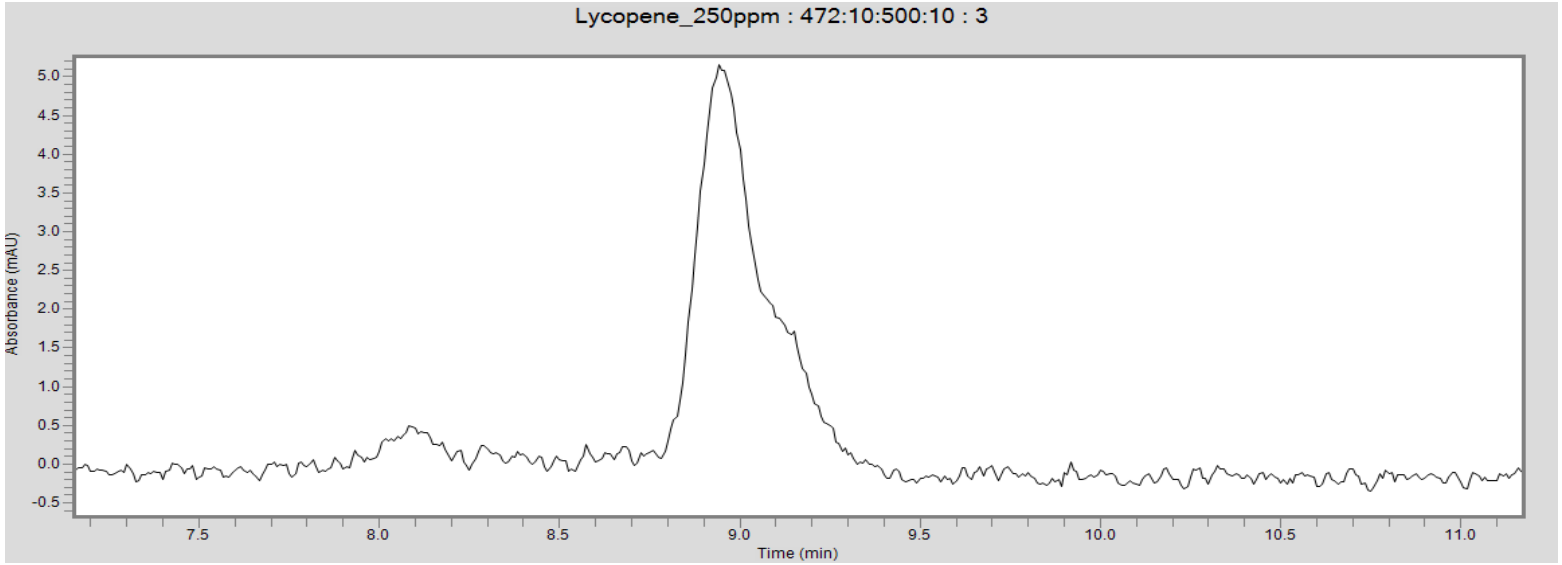

Lycopene_50ppm : 472:10:500:10 : 2
Absorbance (mAU)
2.5
2.0
1.5
1.0
0.5
0.0
7.5
8.0
8.5
9.0
9.5
10.0
10.5
11.0
Time (min)

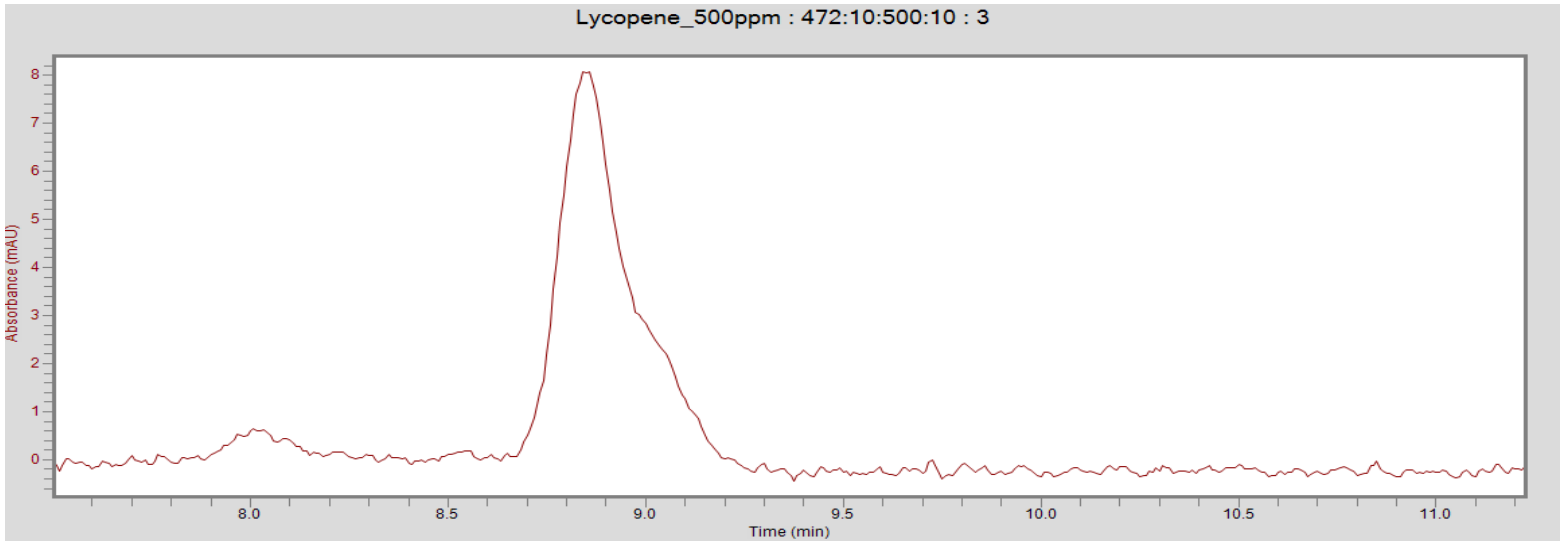

Lycopene_50ppm : 472:10:500:10 : 3
Absorbance (mAU)
3.0
2.5
2.0
1.5
1.0
0.5
0.0
8.0
8.5
9.0
9.5
10.0
10.5
11.0
Time (min)

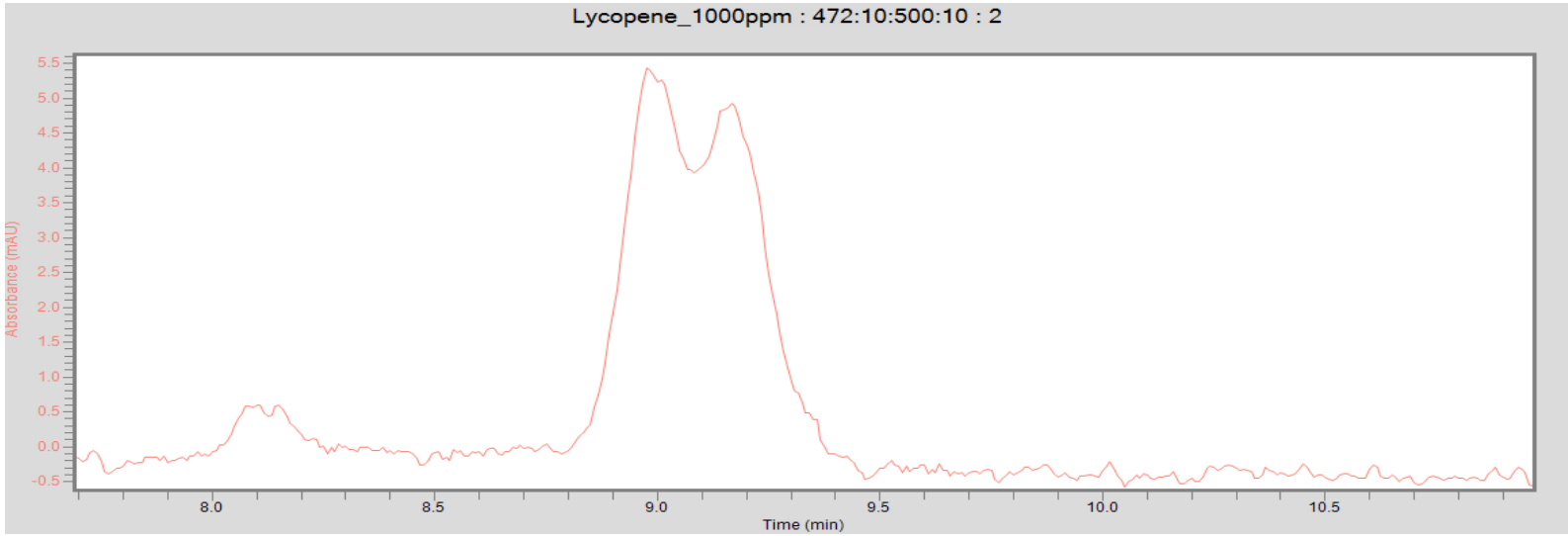

Lycopene_100ppm : 472:10:500:10 : 2
Absorbance (mAU)
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5
7.5
8.0
8.5
9.0
9.5
10.0
10.5
11.0
Time (min)

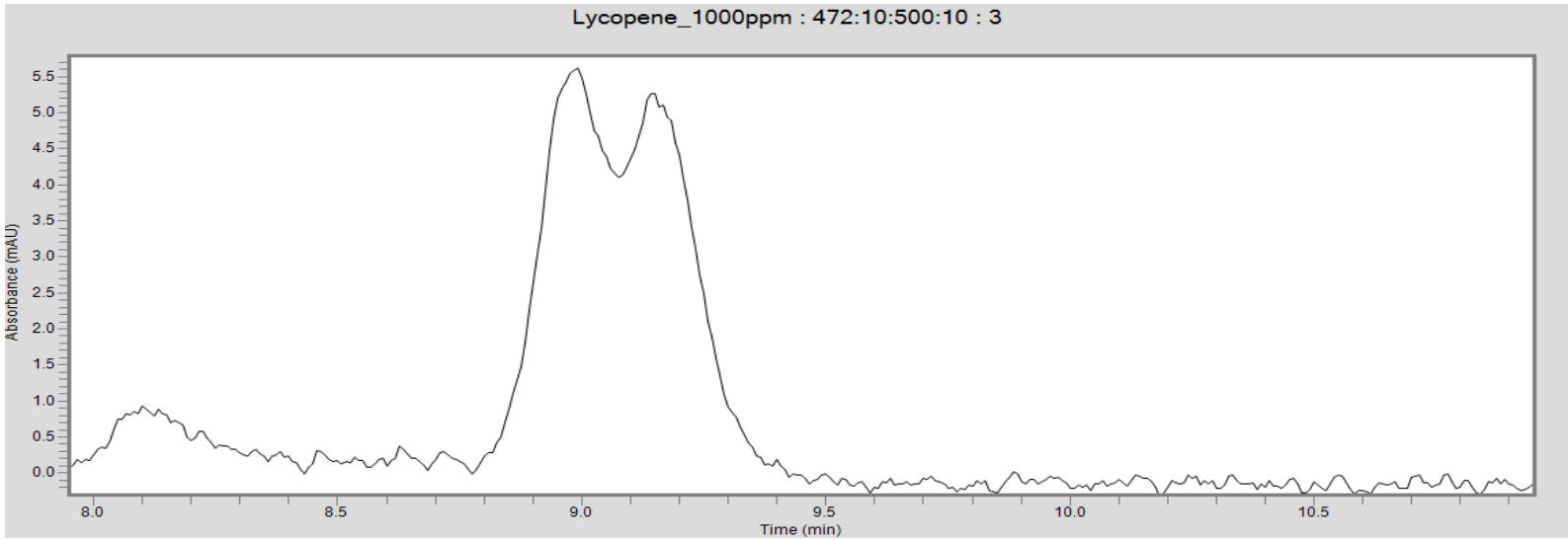

Lycopene_100ppm : 472:10:500:10 : 3
Absorbance (mAU)
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
8.0
8.5
9.0
9.5
10.0
10.5
Time (min)

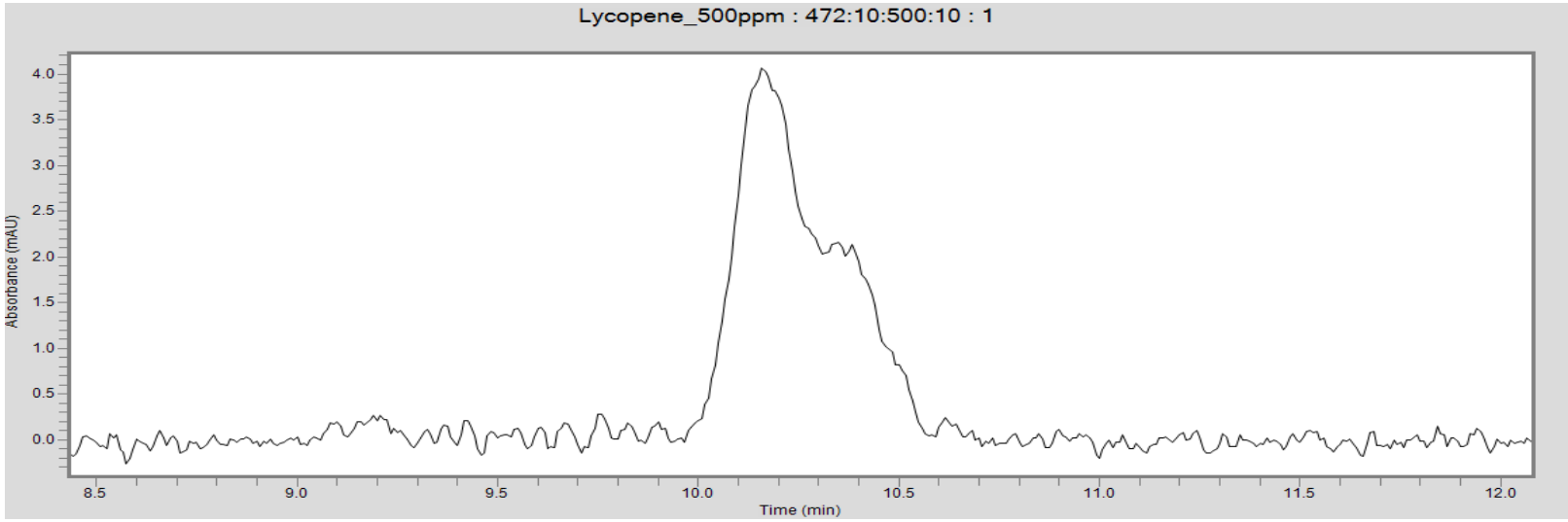

Lycopene_250ppm : 472:10:500:10 : 2
Absorbance (mAU)
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
7.5
8.0
8.5
9.0
9.5
10.0
10.5
Time (min)

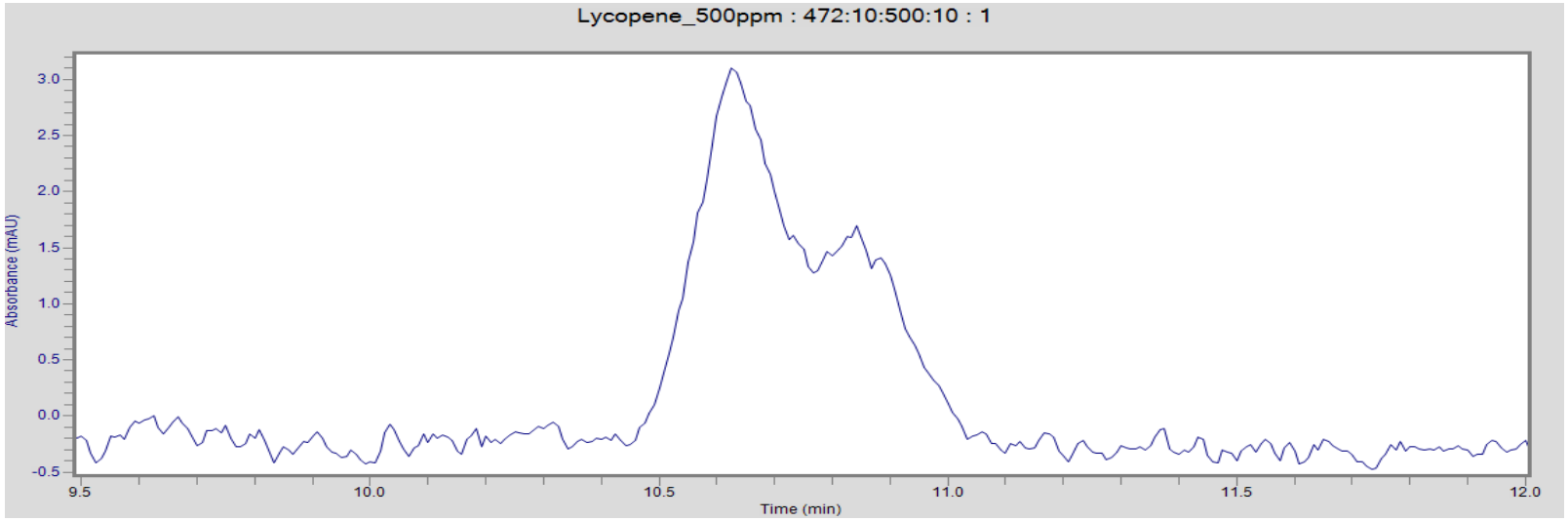

Lycopene_250ppm : 472:10:500:10 : 3
Absorbance (mAU)
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5
7.5
8.0
8.5
9.0
9.5
10.0
10.5
11.0
Time (min)


Lycopene_500ppm : 472:10:500:10 : 3
Absorbance (mAU)
8
7
6
5
4
3
2
1
0
8.0
8.5
9.0
9.5
10.0
10.5
11.0
Time (min)


Lycopene_1000ppm : 472:10:500:10 : 2
Absorbance (mAU)
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5
8.0
8.5
9.0
9.5
10.0
10.5
Time (min)


Lycopene_1000ppm : 472:10:500:10 : 3
Absorbance (mAU)
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
8.0
8.5
9.0
9.5
10.0
10.5
Time (min)


Lycopene_500ppm : 472:10:500:10 : 1
Absorbance (mAU)
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
8.5
9.0
9.5
10.0
10.5
11.0
11.5
12.0
Time (min)


Lycopene_500ppm : 472:10:500:10 : 1
Absorbance (mAU)
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5
9.5
10.0
10.5
11.0
11.5
12.0
Time (min)

## APPENDIX 3: STATISTICAL ANALYSIS TABLES

### *Analysis of Variance in tomato plant height between the 3 treatments*

| | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|
| Between Groups | 32557.481 | 2 | 16278.741 | 551.130 | .000 |
| Within Groups | 1506.389 | 51 | 29.537 | | |
| Total | 34063.870 | 53 | | | |

### *Least Significant Differences in tomato plant height between the 3 treatments*

| (I) trt | (J) trt | Mean Difference (I-J) | Std. Error | Sig. | 95% Confidence Interval | |
|---|---|---|---|---|---|---|
| | | | | | Lower Bound | Upper Bound |
| 1 | 2 | 2.111 | 1.812 | .249 | -1.53 | 5.75 |
| | 3 | 53.111* | 1.812 | .000 | 49.47 | 56.75 |
| 2 | 1 | -2.111 | 1.812 | .249 | -5.75 | 1.53 |
| | 3 | 51.000* | 1.812 | .000 | 47.36 | 54.64 |
| 3 | 1 | -53.111* | 1.812 | .000 | -56.75 | -49.47 |
| | 2 | -51.000* | 1.812 | .000 | -54.64 | -47.36 |

*. The mean difference is significant at the 0.05 level.

### *Analysis of Variance in tomato fruit sizes between the 3 treatments*

| | Sum of Squares | Df | Mean Square | F | Sig. |
|---|---|---|---|---|---|
| Between Groups | 100.990 | 2 | 50.495 | 144.500 | .000 |
| Within Groups | 8.387 | 24 | .349 | | |
| Total | 109.376 | 26 | | | |

### *Least Significant Differences in tomato fruit sizes between the 3 treatments*

| (I) trt | (J) trt | Mean Difference (I-J) | Std. Error | Sig. | 95% Confidence Interval | |
|---|---|---|---|---|---|---|
| | | | | | Lower Bound | Upper Bound |
| 1 | 2 | .9333333* | .2786652 | .003 | .358197 | 1.508470 |
| | 3 | 4.4888889* | .2786652 | .000 | 3.913752 | 5.064026 |
| 2 | 1 | -.9333333* | .2786652 | .003 | -1.508470 | -.358197 |
| | 3 | 3.5555556* | .2786652 | .000 | 2.980419 | 4.130692 |
| 3 | 1 | -4.4888889* | .2786652 | .000 | -5.064026 | -3.913752 |
| | 2 | -3.5555556* | .2786652 | .000 | -4.130692 | -2.980419 |

*. The mean difference is significant at the 0.05 level.

### *Analysis of Variance for for tomato plant diameter between the 3 treatments*

| | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|
| Between Groups | 125.045 | 2 | 62.522 | 110.706 | .000 |
| Within Groups | 28.803 | 51 | .565 | | |
| Total | 153.848 | 53 | | | |

### *Least Significant differences for tomato plant diameter between the 3 treatments*

| (I) trt | (J) trt | Mean Difference (I-J) | Std. Error | Sig. | 95% Confidence Interval | |
|---|---|---|---|---|---|---|
| | | | | | Lower Bound | Upper Bound |
| 1 | 2 | .2722222 | .2505018 | .282 | -.230681 | .775126 |
| | 3 | 3.3555556* | .2505018 | .000 | 2.852652 | 3.858459 |
| 2 | 1 | -.2722222 | .2505018 | .282 | -.775126 | .230681 |
| | 3 | 3.0833333* | .2505018 | .000 | 2.580430 | 3.586237 |
| 3 | 1 | -3.3555556* | .2505018 | .000 | -3.858459 | -2.852652 |
| | 2 | -3.0833333* | .2505018 | .000 | -3.586237 | -2.580430 |

*. The mean difference is significant at the 0.05 level.

### *Analysis of Variance for the number of leaves per plant between the 3 treatments*

| | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|
| Between Groups | 421.778 | 2 | 210.889 | 26.520 | .000 |
| Within Groups | 405.556 | 51 | 7.952 | | |
| Total | 827.333 | 53 | | | |

### *Least Significant differences for the number of leaves per plant between the 3 treatments*

| (I) trt | (J) trt | Mean Difference (I-J) | Std. Error | Sig. | 95% Confidence Interval | |
|---|---|---|---|---|---|---|
| | | | | | Lower Bound | Upper Bound |
| 1 | 2 | 2.556* | .940 | .009 | .67 | 4.44 |

| | | | | | | |
|---|---|---|---|---|---|---|
| | 3 | 6.778* | .940 | .000 | 4.89 | 8.66 |
| 2 | 1 | -2.556* | .940 | .009 | -4.44 | -.67 |
| | 3 | 4.222* | .940 | .000 | 2.34 | 6.11 |
| 3 | 1 | -6.778* | .940 | .000 | -8.66 | -4.89 |
| | 2 | -4.222* | .940 | .000 | -6.11 | -2.34 |

*. The mean difference is significant at the 0.05 level.

### *Analysis of Variance for the number of flowers per plant between the 3 treatments*

| | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|
| Between Groups | 2310.259 | 2 | 1155.130 | 52.459 | .000 |
| Within Groups | 1123.000 | 51 | 22.020 | | |
| Total | 3433.259 | 53 | | | |

### *Least Significant differences for the number of flowers per plant between the 3 treatments*

| (I) trt | (J) trt | Mean Difference (I-J) | Std. Error | Sig. | 95% Confidence Interval | |
|---|---|---|---|---|---|---|
| | | | | | Lower Bound | Upper Bound |
| 1 | 2 | 3.833* | 1.564 | .018 | .69 | 6.97 |
| | 3 | 15.389* | 1.564 | .000 | 12.25 | 18.53 |
| 2 | 1 | -3.833* | 1.564 | .018 | -6.97 | -.69 |
| | 3 | 11.556* | 1.564 | .000 | 8.42 | 14.70 |
| 3 | 1 | -15.389* | 1.564 | .000 | -18.53 | -12.25 |
| | 2 | -11.556* | 1.564 | .000 | -14.70 | -8.42 |

*. The mean difference is significant at the 0.05 level.

### *Analysis of Variance for the number of fruits per plant between the 3 treatments*

| | Sum of Squares | Df | Mean Square | F | Sig. |
|---|---|---|---|---|---|

| | | | | | |
|---|---|---|---|---|---|
| Between Groups | 1633.370 | 2 | 816.685 | 286.480 | .000 |
| Within Groups | 145.389 | 51 | 2.851 | | |
| Total | 1778.759 | 53 | | | |

### *Least Significant differences for the number of fruits per plant between the 3 treatments*

| (I) trt | (J) trt | Mean Difference (I-J) | Std. Error | Sig. | 95% Confidence Interval | |
|---|---|---|---|---|---|---|
| | | | | | Lower Bound | Upper Bound |
| 1 | 2 | 4.667* | .563 | .000 | 3.54 | 5.80 |
| | 3 | 13.278* | .563 | .000 | 12.15 | 14.41 |
| 2 | 1 | -4.667* | .563 | .000 | -5.80 | -3.54 |
| | 3 | 8.611* | .563 | .000 | 7.48 | 9.74 |
| 3 | 1 | -13.278* | .563 | .000 | -14.41 | -12.15 |
| | 2 | -8.611* | .563 | .000 | -9.74 | -7.48 |

*. The mean difference is significant at the 0.05 level.

### *Analysis of Variance for yield per plant between the 3 treatments*

| | Sum of Squares | Df | Mean Square | F | Sig. |
|---|---|---|---|---|---|
| Between Groups | 11000267.890 | 2 | 5500133.945 | 340.779 | .000 |
| Within Groups | 823134.423 | 51 | 16139.891 | | |
| Total | 11823402.313 | 53 | | | |

### *Least Significant differences in yield per plant between the 3 treatments*

| (I) trt | (J) trt | | Std. Error | Sig. | 95% Confidence Interval |
|---|---|---|---|---|---|

| | | Mean Difference (I-J) | | | Lower Bound | Upper Bound |
|---|---|---|---|---|---|---|
| 1 | 2 | 199.6333333* | 42.3476231 | .000 | 114.616932 | 284.649734 |
| | 3 | 1041.5166667* | 42.3476231 | .000 | 956.500266 | 1126.533068 |
| 2 | 1 | -199.6333333* | 42.3476231 | .000 | -284.649734 | -114.616932 |
| | 3 | 841.8833333* | 42.3476231 | .000 | 756.866932 | 926.899734 |
| 3 | 1 | -1041.5166667* | 42.3476231 | .000 | -1126.533068 | -956.500266 |
| | 2 | -841.8833333* | 42.3476231 | .000 | -926.899734 | -756.866932 |

*. The mean difference is significant at the 0.05 level.

## *Analysis of Variance for single fruit weight values between the 3 treatments*

| | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|
| Between Groups | 773.767 | 1 | 773.767 | 29.370 | .000 |
| Within Groups | 895.761 | 34 | 26.346 | | |
| Total | 1669.528 | 35 | | | |

## *Least Significant differences in single fruit weight between the 3 treatments*

| (I) trt | (J) trt | Mean Difference (I-J) | Std. Error | Sig. | 95% Confidence Interval | |
|---|---|---|---|---|---|---|
| | | | | | Lower Bound | Upper Bound |
| 1 | 2 | 5.5778 | 2.8144 | .053 | -.072 | 11.228 |
| | 3 | 102.8667* | 2.8144 | .000 | 97.217 | 108.517 |
| 2 | 1 | -5.5778 | 2.8144 | .053 | -11.228 | .072 |
| | 3 | 97.2889* | 2.8144 | .000 | 91.639 | 102.939 |
| 3 | 1 | -102.8667* | 2.8144 | .000 | -108.517 | -97.217 |

| | | | | | | |
|---|---|---|---|---|---|---|
| | 2 | -97.2889* | 2.8144 | .000 | -102.939 | -91.639 |

*. The mean difference is significant at the 0.05 level.

### *Analysis of Variance for Brix values between the 2 treatments*

| | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|
| Between Groups | 4.720 | 1 | 4.720 | 7.687 | .010 |
| Within Groups | 17.195 | 28 | .614 | | |
| Total | 21.915 | 29 | | | |

### *Analysis of Variance for lycopene content between the 2 treatments*

| | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|
| Between Groups | 374.460 | 1 | 374.460 | 288.046 | .000 |
| Within Groups | 5.200 | 4 | 1.300 | | |
| Total | 379.660 | 5 | | | |

### *Analysis of Variance for source of microorganisms between the 3 treatments*

| | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|
| Between Groups | 852.343 | 2 | 426.172 | 396.029 | .000 |
| Within Groups | 16.142 | 15 | 1.076 | | |
| Total | 868.485 | 17 | | | |

### *Least Significant Differences for source of microorganisms between the 3 treatments*

| (I) trt | (J) trt | Mean Difference (I-J) | Std. Error | Sig. | 95% Confidence Interval | |
|---|---|---|---|---|---|---|
| | | | | | Lower Bound | Upper Bound |
| 1 | 2 | -3.783* | .599 | .000 | -5.06 | -2.51 |

| | | | | | | |
|---|---|---|---|---|---|---|
| | 3 | 12.333* | .599 | .000 | 11.06 | 13.61 |
| 2 | 1 | 3.783* | .599 | .000 | 2.51 | 5.06 |
| | 3 | 16.117* | .599 | .000 | 14.84 | 17.39 |
| 3 | 1 | -12.333* | .599 | .000 | -13.61 | -11.06 |
| | 2 | -16.117* | .599 | .000 | -17.39 | -14.84 |

*. The mean difference is significant at the 0.05 level.

## *Analysis of Variance for the concentration of microorganisms between the 4 treatments*

| | Sum of Squares | Df | Mean Square | F | Sig. |
|---|---|---|---|---|---|
| Between Groups | 113.415 | 3 | 37.805 | 328.500 | .000 |
| Within Groups | 2.302 | 20 | .115 | | |
| Total | 115.716 | 23 | | | |

## *Least Significant Differences for the concentration of microorganisms between the 4 treatments*

| (I) trt | (J) trt | Mean Difference (I-J) | Std. Error | Sig. | 95% Confidence Interval | |
|---|---|---|---|---|---|---|
| | | | | | Lower Bound | Upper Bound |
| 0 | 1 | -1.3833* | .1959 | .000 | -1.792 | -.975 |
| | 5 | -2.7667* | .1959 | .000 | -3.175 | -2.358 |
| | 10 | -5.8667* | .1959 | .000 | -6.275 | -5.458 |
| 1 | 0 | 1.3833* | .1959 | .000 | .975 | 1.792 |
| | 5 | -1.3833* | .1959 | .000 | -1.792 | -.975 |
| | 10 | -4.4833* | .1959 | .000 | -4.892 | -4.075 |
| 5 | 0 | 2.7667* | .1959 | .000 | 2.358 | 3.175 |
| | 1 | 1.3833* | .1959 | .000 | .975 | 1.792 |
| | 10 | -3.1000* | .1959 | .000 | -3.509 | -2.691 |

| | | | | | | |
|---|---|---|---|---|---|---|
| | 0 | 5.8667* | .1959 | .000 | 5.458 | 6.275 |
| 10 | 1 | 4.4833* | .1959 | .000 | 4.075 | 4.892 |
| | 5 | 3.1000* | .1959 | .000 | 2.691 | 3.509 |

*. The mean difference is significant at the 0.05 level.

### *Analysis of Variance for manure for optimal nitrate generation between the 3 treatments*

| | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|
| Between Groups | 136098.778 | 2 | 68049.389 | 82762.770 | .000 |
| Within Groups | 12.333 | 15 | .822 | | |
| Total | 136111.111 | 17 | | | |

### *Least Significant Differences for manure for optimal nitrate generation between the 3 treatments*

| (I) trt | (J) trt | Mean Difference (I-J) | Std. Error | Sig. | 95% Confidence Interval | |
|---|---|---|---|---|---|---|
| | | | | | Lower Bound | Upper Bound |
| .25 | .50 | 182.167* | .524 | .000 | 181.05 | 183.28 |
| | 1.00 | 186.667* | .524 | .000 | 185.55 | 187.78 |
| .50 | .25 | -182.167* | .524 | .000 | -183.28 | -181.05 |
| | 1.00 | 4.500* | .524 | .000 | 3.38 | 5.62 |
| 1.00 | .25 | -186.667* | .524 | .000 | -187.78 | -185.55 |
| | .50 | -4.500* | .524 | .000 | -5.62 | -3.38 |

*. The mean difference is significant at the 0.05 level.