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Editor's Note

It is with great pleasure and privilege to publish the Volume 5 of the Journal of Dry Zone Agriculture (JDZA) of Faculty of Agriculture, University of Jaffna, Sri Lanka. The objective of the JDZA is to publish up-to-date and high quality research findings, focusing all aspects of agriculture in the dry zone of Sri Lanka. Since 2/3 of Sri Lanka is under dry zone and agriculture in these areas witnessed severe losses due to climate change at recent times especially abnormal floods and prolonged severe drought that are given prime importance in the Sustainable Development Goals to be achieved in 2030. Mitigating climate change is the prioritized area to be considered in the next decade since water will be an expensive commodity in near future. Solutions through research to support the activities identified against SDGs will contribute immensely to the regional, national and by global contexts.

Agriculture needs further advancement in technological applications into diversifying the habitat, resource management, marketing, value addition and storage. Integration of all the sectors in agriculture with the livestock and fisher sectors is the real-time development expected in Northern Province to increase the employment opportunities as well as to uplift its GDP contribution. Researches should also need to focus on agro based industrial development, automation, use of renewable energy and labour consuming technologies to uplift the agricultural development to the next level.

This volume consists of twelve full research articles are selected from the abstracts presented at 4th International Conference on Dry Zone Agriculture (ICDA 2018) and submitted research papers in 2019. The Research papers submitted to JDZA are not published previously in the same, or any other form or being considered for publication elsewhere. To ensure the quality of the research papers, all the papers are peer reviewed and finalized by the team of experts.

I would like to thank the contributions made by the authors, and voluntary support given by the reviewers, editors and the editorial board for their tireless efforts to finalize the articles and messages to this volume. Further I also would like to acknowledge the financial support provided by the University of Jaffna to publish the journal. The Editorial Board encourages publications relating to research and development especially on dry zone agriculture containing new methodological approaches to disseminate the knowledge to the community.

At last I thank all who witnesses the release of this journal on the inaugural day of the 5th International Conference on Dry Zone Agriculture in December, 2019.

Prof. Thushyanthy Mikunthan
Editor-in-Chief

List of Content

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Repeated Batch Process for Glucoamylase Production by *Aspergillus niger*

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Abstract: Production of glucoamylase (amyloglucosidase) by *Aspergillus niger* was studied in batch and repeated batch submerged fermentations. In batch culture, growth (biomass 35.8 gL^{-1}) and glucoamylase production (53.2 UmL^{-1}) were highest at 108 h (4.5 days). Addition of spores on the 4th and 8th days to augment the old mycelium extended the glucoamylase production up to 9 days. When the spent medium was supplemented with soluble starch from manioc (50 gL^{-1}) on the 4.5th day, glucoamylase activity increased by 14.6 UmL^{-1} on the 6th day [one and a half day (36 h) after the addition of starch]. Replacement of the spent medium with the fresh fermentation medium on the 4th day, led to 70.3 UmL^{-1} glucoamylase activities on the 7th day (3rd day after the addition of the fresh fermentation medium). The spent medium was replaced with the fresh fermentation medium along with the addition of spores led to an increase in the glucoamylase production by 1.62 and 1.66 folds in the 2nd and the 3rd cycles of that obtained in the first cycle. Thus fed batch process was not possible under the conditions considered for glucoamylase production while repeated batch process seems to be promising.

Keywords: *Aspergillus niger*, Biomass, Glucoamylase, NADPH, Repeated batch process

Introduction

Glucoamylase (1, 4,-α-D- Glucangluco-hydrolase, E.C. 3.2.1.3) is an exo-acting carbohydrase which cleaves glucose units consecutively from the non-reducing end of starch molecule. It is also known as gluc - amylase, amyloglucosidase or λ-amylase. Glucoamylase is produced mainly by fungi such as *Rhizopus* (Inshik and Chunk, 1989) and *Aspergillus* (Lasater and Simith, 1979). Glucoamylase is used for the production of high purity glucose syrups (Mc Mullen, 1977), high fructose syrups (Norman, 1979) high conversion syrups (Norman, 1979), levulose from granular starch (Hebeda and Leach, 1975) and sugar syrups of different dextrose equivalent (DE) values (Normanand Nelson, 1984; Saha *et al.,* 1989; Kumar and Satyanarayan, 2009; Nandy, 2016).

Glucoamylase is produced by solid state fermentation (Ramadas *et al.,* 1996; Arasaratnam *et al.,* 2001; de Souza and Peralta, 2001; Bertolin *et al.,* 2003; Costa *et al.,* 2007; Varzakas *et al.,* 2007; Aithal *et al.,* 2011), continuous cultures (Metwally, 1997; Pedersen *et al.,* 2000), fed batch cultivation (Pedersen *et al.,* 2000; Pederson *et al.,* 2012; Luo *et al.,* 2015; Rani *et al.,*

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1999) cell recycling fermentation (van Verserveld *et al.,* 1991) and with immobilised cells (Konwarth *et al.,* 2010).

The productivity obtained with conventional fermentation processes is limited due to low concentration of microorganism in the system (Arasaratnam *et al.,* 1994). Cell mass and enzyme productivity can be increased by repeated batch process or cell recycling (Ohleyer *et al.,* 1985). Additional advantages of these methods are avoiding product inhibition, broth viscosity (Afschar *et al.,* 1985) and elimination of the time needed for spore germination (Roukas and Alichanidis, 1991). This paper describes the different trails made for the production of glucoamylase continuously from *Aspergillus niger* based on cell recycling, supplementation of nutrients and addition of spores.

Materials and Methods

Chemicals

Manioc flour and soya flour were prepared in the laboratory from local sources. All the other chemicals used were of analytical grade and were purchased from standard sources.

Microorganism

Aspergillus niger CISIR N_4 was used. The organism was maintained on Potato Dextrose Agar (PDA) slants at 4°C and subcultured monthly.

Analytical methods

Reducing sugar in the medium was estimated by dinitrosalicylic acid method (Miller, 1959).

Measurement of glucoamylase activity

The activity of glucoamylase was determined with p-nitrophenyl α-D-glucopyranosideas substrate (Miranda *et al.,* 1987). One unit of glucoamylase is the amount of enzyme which release one m mole of p-nitrophenol from p-nitrophenyl α-D-glucopyranoside in one minute at 60°C and pH 4.0.

Analysis of growth - Biomass

Microbial growth was estimated by measuring biomass dry weight. Samples were centrifuged at 6000 rpm for 10 min, washed with distilled water and dried at 105°C to obtain constant weight (Stone *et al.,* 1992).

NADPH level

Aliquots (5 mL) removed were centrifuged (MSE model 2) at speed 7 for 10 min. Residues were washed three times with 8 mL of 0.02M acetate buffer (pH 4.0). The residue (mycelium) was homogenized with Triton-100 (5 mL, one gL^{-1}) and centrifuged as above. The NADPH content of the supernatant was estimated in a fluorescence spectrophotometer at excitation and emission wave lengths of 360 and 450 nm respectively against Triton X-100 (1 gL^{-1}) 0.02M acetate buffer (pH 4.0) as blank (Taya *et al.,* 1986).

Batch Cultivation

The fermentation medium contained (gL^{-1}) soluble starch from manioc flour, 20;$(NH_4)_2SO_4$, 5;KH_2PO_4, 0.5;$MgSO_4.7H_2O$, 0.5;KCl, 0.5;$FeSO_4.7H_2O$, 0.01;soya flour,20 and peptone, 10.0 at pH 6.5. Fermentation medium (200 mL) was inoculated with spores (1.25 mL, 108 spores mL^{-1} in 0.01%, v/v, Tween). The flasks were incubated at

37°C in an orbital shaker (150 rpm) and biomass, NADPH level, reducing sugar and glucoamylase activity were monitored.

Addition of spores

Experiment was carried out as said in batch cultivation and spores (108 spores mL^{-1}) were added to the medium when the NADPH level reached about 82 mL^{-1}. Samples were analysed.

Supplementation of spent medium with soluble starch

The spores were cultivated as said above in 200 mL fermentation medium. Soluble starch solution (50 gL^{-1}) was added to bring back the starch concentration to 10 gL^{-1} when the reducing sugar concentration was decreased to about 0.0 gL^{-1}. Samples were taken and analysed for reducing sugar, glucoamylase activity and NADPH level.

Replacement of the spent medium with fresh fermentation medium

Batch cultivation was carried out and when the reducing sugar concentration was decreased to about 0.0 gL^{-1}. The spent medium was strained through a sterile muslin cloth and the enzyme trapped in between the mycelia was removed by pressing the mycelia in between the muslin cloth. Fresh sterile fermentation medium was added to the mycelium. All these steps were carried out under aseptic conditions. The cultivation was continued and samples were analysed.

Addition of spores with replacement of spent medium with fresh fermentation medium

Batch cultivation was carried out by replacing the spent medium with fresh fermentation medium and the spores were added as said in the above experiment. The replacement was carried out, when the reducing sugar level was decreased to about 0.0 gL^{-1}.

Results and Discussions

The aim of this work is to produce higher titer of glucoamylase by *Aspergillus niger*. The experiments were carried out to improve the glucoamylase production by supplementing the spent medium with the carbon source or by repeated batch cultivation and by introducing the spores into the medium during the replacement of the spent medium with fresh fermentation medium.

Batch Cultivation

The fungus was cultivated in batch process at the laboratory level in shake flasks containing 200 mL of the fermentation medium. Samples were withdrawn time to time under aseptic conditions for enzyme activity, residual sugar content, NADPH level and the dry weight measurements.

Growth of the *Aspergillus niger* was monitored by measuring the NADPH level as well as the dry weight of the mycelium. NADPH level and dry weight of the mycelium showed good correlation (Figure 1). Growth of the mycelium (biomass 35.8 gL^{-1}) and glucoamylase production (53.2 UmL^{-1}) reached maximum at 108 h (Figure 1). Thereafter, no increase in enzyme production was observed. The activity of the enzyme produced in the medium remained constant while growth (both the dry weight and NADPH level) decreased after 108 h. Reducing sugar level of the medium declined rapidly after

48 h (Figure 1). Decrease in the viability of the mycelium could be due to the lack of nutrients or the accumulation of the toxic products would have led to autolysis or death of the mycelium. Glucoamylase production by *Aspergillus niger* requires energy source (van Verseveld *et al.,* 1991). The lack of carbon source was evidenced by a decrease in the reducing sugar level to 0.0 gL^{-1} at 108 h and no further increase in the glucoamylase titre beyond 108 h. As the reducing sugar level was reduced to 0.0 gL^{-1} at 108 h, a study was carried out to supplement the spent medium with carbon source (soluble starch, which also would dilute the toxic substances in the spent medium).

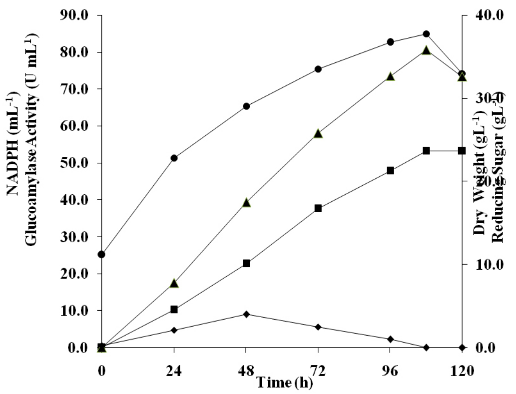

Figure 1: Growth of and glucoamylase production by *Aspergillus niger* in batch cultivation at pH 6.5 and 37°C and changes in reducing sugar and dry weight.
(■) - Glucoamylase activity;
(♦) - Reducing Sugar;
(●) - NADPH and
(▲)- Dry weight

Addition of Spores

Glucoamylase production is also depending on the specific growth rate of the mycelium (Metwally, 1998). The NADPH level increased from 82.6 to 89.2 mL^{-1}, spores were added on the 4th and 8th days (Figure 2). The NADPH level of the mycelium was increased up to 24 hours (5th day of the commencement of the experiment), after the addition of spores on the 4th day. Glucoamylase production (53.8 UmL^{-1}) was increased up to the 6th day, i.e., 48 h after the addition of the spores and stated to decrease on the 7th day to 52.7 UmL^{-1} (Figure 2). Addition of spores on the 8th day showed a slight increase in the NADPH level with slight increase in glucoamylase production (Figure 2). This indicated the inhibition of the germination of the spores as well as the glucoamylase production by the accumulated toxic substance in the spent medium as well as by the deficiency of nutrients.

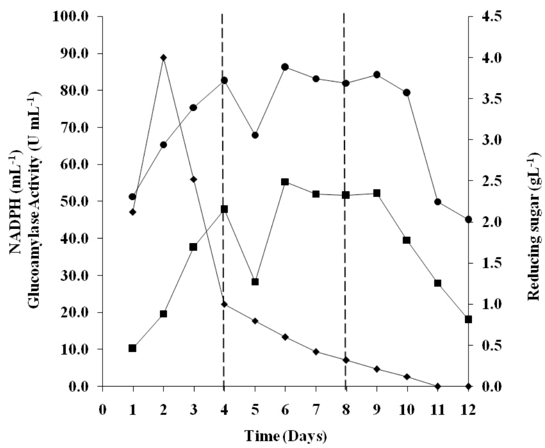

Figure 2: Effect of addition of spores on the changes in glucoamylase production, reducing sugar and growth (NADPH).
The lines indicate the addition of spores (1.25 mL; 108 spores mL^{-1}) to the spent medium on the 4th and 8th days.
(■) - Glucoamylase activity;
(♦) - Reducing Sugar and
(●) -NADPH

Germination of spores is considerably repressed by carbon dioxide (Meyrath and Bayer, 1987) and such toxic substances would have affected the germination of the spores. Spent medium rich in by-products, with increased osmolarity and limited dissolved oxygen would affect the glucoamylase production (Pederson *et al.,* 2012). Hence to have better glucoamylase production it was decided supplement the spent medium with the carbon source, i.e. the soluble starch.

Supplementation of spent medium with soluble starch

Since the growth of the fungus started to decrease at 108 h (Figure 1), and addition of spores at 4th and 8th days did not improve the glucoamylase production (Figures 1 and 2), soluble starch from manioc (50 gL^{-1}) was added at 108 h (4.5 day) when reducing sugar concentration decreased to 0.0 gL^{-1}. When soluble starch (40 mL) was introduced into the spent medium (160 mL) a fall in glucoamylase activity from 53.2 to 42.4 UmL^{-1} was observed (Figure 3). This was due to the dilution of the spent medium. On the 1.5th day after the addition of soluble starch (6th day after the commencement of the experiment) the glucoamylase activity was increased to 54.2 UmL^{-1}. If the dilution factor is considered, addition of soluble starch had increased the enzyme production by 14.6 UmL^{-1}. NADPH level was increased on the 5th day (12 h after the addition of soluble starch) and the levels of NADPH and reducing sugar continued to decrease after the addition of the carbon source (Figure 3). Beyond 7th day, there was decrease in the glucoamylase production. This indicated that in presence of toxic substances in the spent medium, addition of only the carbon source is not useful.

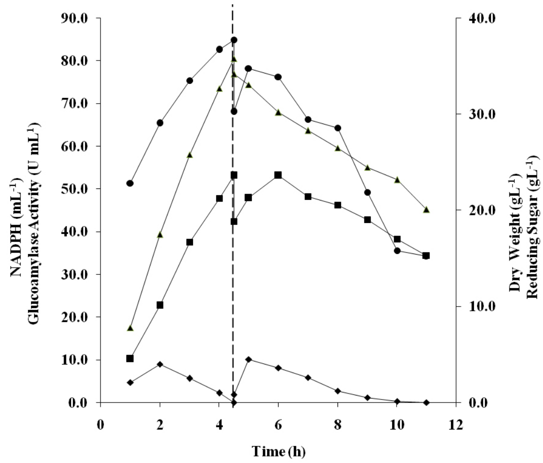

Figure 3: Effect of soluble manioc starch supplementation (50 gL^{-1}) on the changes in glucoamylase production, reducing sugar and growth (NADPH).

The line indicates the supplementation of manioc starch to the spent medium.

(■) - Glucoamylase activity;
(♦) - Reducing Sugar;
(●) - NADPH and
(▲) - Dry weight

Even though there has been a decrease in the glucoamylase production, utilization of the reducing sugar was observed. But the rate of utilization of the carbon source was not the same as that observed in the initial four days (Figure 3). The increase in glucoamylase activity in the spent medium could be either due to the release of intracellular enzyme from the remaining mycelia or the dead or autolysed cells. However other studies had reported an increase in glucoamylase production in fed batch process under controlled conditions (Ramadas *et al.,* 1996, Pedersen *et al.,* 2000) and reduction in cost of enzyme production (Luo *et al.,* 2012). Hence it can be concluded that the addition starch alone had no significant effect on repeated batch glucoamylase production. Another experiment was carried out by replacing the spent medium with fresh fermentation medium to provide all the required nutrients while removing the toxic substances released into the spent medium.

Replacement of the spent medium with fresh fermentation medium

As the supplementation of soluble starch to the spent medium did not show significant effect and the toxic substances seems to get accumulated in the spent medium, the spent medium was completely replaced with fresh medium after the reduction of the reducing sugar to 0.0 gL^{-1} (Figure 4). The spent medium after four days of fermentation was strained and replaced with fresh medium as the extracellular glucoamylase production requires energy and requires the supply of sufficient energy sources (van Verseveld *et al.,* 1991). When the spent medium was replaced with the fresh fermentation medium, glucoamylase production started to increase and reached a maximum value (70.3 UmL^{-1}) on the 3rd day (7th day after the commencement of the experiment).

These results emphasized that the reason for the decreased glucoamylase production as the toxic substances present in the medium (Figure 4). Thereafter from the 3rd day (7th day after the commencement of the experiment) the enzyme activity started to decrease. Decrease in the enzyme production beyond the 7th day, i.e. 3rd day after the replacement of the spent medium with the fresh medium could be due to the aging of the mycelium. With increase in the age of the mycelium the viability of the cells might have decreased (Rhem and Reed, 1988). Maximum activity in the 2nd cycle obtained was 70.3 UmL^{-1} and was 1.32 times higher than that of the highest activity (53.2 UmL^{-1}) obtained in the 1st cycle. The time required for maximum glucoamylase production was decreased in the 2nd cycle from 4.5 to 3 days.

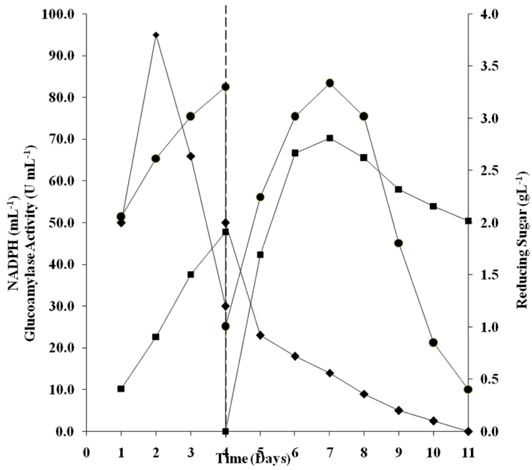

Figure 4: Effect of replacement of spent medium with fresh medium on the changes inglucoamylase production, reducing sugar and growth (NADPH).

The line indicates the replacement of spent medium with fresh fermentation medium.

(■) - Glucoamylase activity;

(♦) - Reducing Sugarand

(●) - NADPH

The results showed that even though the mycelium was relieved from toxic effect which could have been brought about by the removal of the spent medium, the efficiency of glucoamylase production was high for a short while and the enzyme production was reduced. The oxygen limitation, high osmolarity and by-products metabolism can affect the glucoamylase production

(Pedersen *et al.,* 2012). This may be due to the reduced enzyme production and release of the enzyme from the dead mycelium. *Botryodiplodia theobromae* retained the capacity to produce glucoamylase through four cycles with an increase in the age of the mycelium (8 days) (Navaratnam *et al.,* 1996). In continuous batch citric acid production, 70% of the original citric acid production was obtained in the second cycle (Roukas and Alchsndis, 1991). In this studies the rate of reducing sugar utilization as well as the NADPH level were reduced. These can be considered as the indications of the loss of the mycelial viability. Hence a study was carried out to have young mycelium by introducing the spores into the medium while replacing the spent medium with fresh medium to remove the toxic substances and to provide nutrients.

Replacement of the spent medium with fresh fermentation medium and addition of spores

On the 4th and 8th days the spent medium was replaced with fresh fermentation medium and spores were added to the medium. When additional spores were introduced on the 4th day the NADPH level and glucoamylase activity started to increase and reached the maximum value of 86.3 UmL^{-1} on the 7th day, i.e., 3 days after the replacement of the medium and addition of spores. Similarly when the spent medium was replaced with the fresh medium and spores were also added on the 8th day, the glucoamylase production has reached the maximum value on the 10th day to 88.5 UmL^{-1} which is 2 days after the replacement of the spent medium and addition of spores (Figure 5).

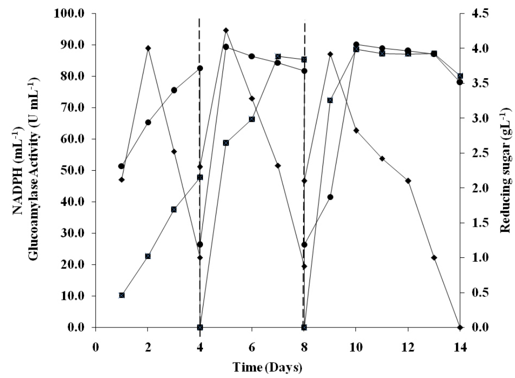

Figure 5: Effect of addition of spores and replacement of spent medium with fresh medium on glucoamylase production, reducing sugar and growth (NADPH).

The lines indicates the replacement of spent medium with fresh medium and the addition of spores (1.25 mL; 108 spores mL^{-1}) on the 4th and the 8th days.

(■) - Glucoamylase activity;
(♦) - Reducing Sugarand
(●) - NADPH

Highest activity was obtained in the 2nd and 3rd cycles were 86.3 and 88.5 UmL^{-1}, respectively. These values were equivalent to 1.62 and 1.66 times of the maximum activity (53.2 UmL^{-1}) obtained in the 1st cycle (Figure 5). Thus the added spores would have grown in the fresh fermentation medium and have given young mycelium to produce glucoamylase in addition to the mycelium which existed. This is somewhat similar to the inoculation of the fresh fermentation medium with the spores, as well as the viable mycelium producing the enzyme.

The glucoamylase production could have been the double the amount of what was produced in the first cycle. Decreased glucoamylase production than that was expected could be because of the degradation of the old mycelium as well as the inhibitory effect of the derivative products of the mycelium on the growth of the spores.

When the fresh medium was introduced along with the spores, the NADPH level did not reduce substantially by the fourth day (i.e. by the 8th day of the commencement of the experiment) of the introduction of the medium and the spores, indicating the germination and the of the spores and the growth of the mycelium. When the fresh medium was again replaced for the spent medium and the additional spores were introduced, the NADPH level has started to declaim by the 12th day and did not decrease as it was observed in the previous experiments. The decrease in the NADPH level was observed when the reducing sugar level has reached 0.0 gL^{-1} on the 14th day of the commencement of the work and 6th day after the introduction of the fresh medium and the spores (Figure 5). Even though the addition of the spores had not increased the glucoamylase production substantially, it has helped the medium to have viable mycelium and to produce glucoamylase continuously (Metwally, 1998) and has provided sufficient energy for the production of the extracellular glucoamylase (van Verseveld *et al.,* 1991).

Conclusions

This work indicates that glucoamylase production in repeated batch culture was not possible by adding spores to the spent medium or supplementing the spent medium with carbon source or replacing the spent medium with fresh fermentation medium. This study clearly indicated that addition of fresh medium and introducing the spores to the pre-existing mycelium can allow the glucoamylase production to continue at least for three cycles. The study also has indicated that mycelial death after the first cycle and deficiency of nutrients affect the enzyme production substantially. Hence for the production glucoamylase, the organism could be cultured by replacing the spent medium with the fresh medium to remove the inhibitory products and maintaining the viable mycelium by adding spores intermittently.

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Effect of Variety and Polishing on Cooked Red Rice Colour Change after Blending with Iron and Folic Acid Fortified Kernels

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Abstract: Micronutrient malnutrition is a global issue and iron deficiency is a major public health concern, particularly, in young children and pregnant women in Sri Lanka. Rice fortification with micronutrients has been recognized as a key approach to alleviate micronutrient deficiency in rice eating populations. Sri Lankans consume both parboiled (55%) and raw rice (45%) in red (23%) and white (77%) forms. Therefore, this study was conducted to investigate the issue of iron and folic acid fortified kernel blending with red pericarp rice. The objective was to investigate the influence of the variety and degree of polishing on the intensity of blue colour spot development in cooked rice around kernels during cooking. Nine red rice varieties; Bw 361, Attakari, Bg 252, Ld 365, Ld 356, Bg 406, At 303, At 362 and Bw 364 were polished to 2 and 8 %. Rice blending was done at 1: 99 ratio (kernel: red rice) with kernels having 8 mg/100 g of ferric pyrophosphate and folic acid (13.5 mg/100 g). Fifty grams of blended and normal rice (control) in raw and parboiled forms were cooked under same conditions and blue color spot development was visually observed. Results revealed that no colour change in controls and blended parboiled red rice after cooking. However, Attakari and Bg 252 raw rice developed high and slightly greenish blue color spots, respectively under both polishing levels. The blue color intensity decreased with the increasing degree of polishing for both varieties. Therefore, it is assumed that the phenolic compounds (antioxidants) in the bran reduce Fe^{3+} to Fe^{2+} during cooking and the variety and polishing influences the intensity of colour change.

Keywords: Antioxidants, Iron Fortification, Red Rice Blending.

Introduction

Food fortification has the advantage to deliver nutrients to large segments of the population without requiring radical changes in food consumption patterns. The main reason for the increased interest is the realization of public health implications of micronutrient malnutrition are potentially huge (Allen *et al.,* 2006). Micronutrient deficiencies in iron, zinc, vitamins A and B, iodine, and folic acid affect more than 2 billion people worldwide today (Tontisirin *et al.,* 2002), of which iron deficiency is the most prevalent. Especially, in pregnant women and young children the effects are serious as they affect fetal and child growth, cognitive development, education, economic development, productivity and resistance to infection. People in different age groups in many regions of Sri Lanka are affected by iron deficiency. According to WHO mortality data, around 0.8 million

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deaths (1.5% of the total) are attributed due to iron deficiency each year (Allen *et al.,* 2006). Iron-deficiency anemia results in 25 million of the global population (WHO, 2002). According to the national nutrition and micronutrient survey, 15.1% of children between 6-59 month ages suffer from anemia in Sri Lanka.

The highest iron deficiencies of children under above age group were reported in Kilinochchi (26.9%), Moneragala (25.6%), Trincomalee (23.1%) and Puttalam (20.3%) districts in Sri Lanka (Jayatissa *et al.,* 2012). Iron is considered as one of the most limiting micronutrients, especially in diets with polished rice. Unpolished rice contains about 2.6 mg iron/100 g whereas polished rice contain as low as 0.4–0.6 mg/ 100 g (Steiger *et al.,* 2014).

Iron fortification is the optimal approach to reduce high iron deficiency anemia (Cook and Reusser, 1983). The bioavailable forms of iron are chemically reactive and produce undesirable effects when added to food. Iron compounds that have a high bioavailability and whose absorption is not so susceptible to the negative effects of inhibitory ligands would be the ideal way of administering iron via food (Davidsson *et al.,* 1994).

Although such compounds are commercially available for fortification at present, they develop quality issues in cooking locally available red pericarp rice. Therefore, the objective of this study was to investigate the influence of variety, parboiling and degree of polishing of red pericarp rice on cooked rice colour change when blended with iron and folic acid fortified kernels.

Materials and Methods

Collection and preparation of samples

Nine varieties of red pericarp rice (*Oryza sativa* L.) samples were collected from Rice Research Station, Paranthan. For this experiment, two different degree of polishing (2% and 8%), form of rice (parboiled or raw) were used for blending while unblended normal rice of the same sample was used as the control. The nine popular red rice varieties were selected; Bw 361, Attakari, Bg 252, Ld 365, Ld 356, Bg 406, At 303, At 362 and Bw 364. Freshly de-husked red rice of each variety was used for the study. They were milled and raw rice was produced using a SATAKE laboratory rice mill and a polisher. Parboiled rice was also produced from all above varieties in the laboratory and milled using the same milling machines.

Fortificants and rice blending

The source of iron used for the fortified kernel production was Ferric pyrophosphate (8 g/100 g) and hot extruded kennels were used. These kernels were also fortified with folic acid as well (13.5 mg/100 g). Rice fortification is usually done by blending fortified kernels with normal at a 1: 99 ratio (Global Alliance for Improved Nutrition, 2015). The two different levels of polished raw and parboiled rice samples were blended with kernels at 1% level.

Cooking fortified rice

Water (75 mL - 100 mL depend on variety) was added to fifty grams of normal (Control) or blended, raw and parboiled rice and cooked for 20 min in a 250 mL glass beaker. Observations were made for 2% and 8% polished rice of both parboiled and raw forms. Blue colour development of cooked rice was visually observed. The colour comparison of cooked rice was done by a qualitative duo-trio test with 30 untrained panelists. Significance was tested at three different significance levels; 90%, 95% and 99.1%.

Results and Discussions

Polished Raw Rice cooking

It was observed that normal rice (unblended) had no any greenish blue color development in both 2% and 8% polishing levels after cooking. Two rice varieties Attakari and Bg 252 have shown hard greenish blue color at 2% polishing level and slight greenish blue color at 8% polishing level. The sensory panel test results revealed that there was a "Perceptually different" colour between the control and the blended rice at three different significant levels. Other seven tested rice varieties developed undetectable blue color after cooking and the sensory result revealed that it was "Perceptually similar" at three different significant levels;

Table 1: Visual observation of blue color development of rice bran and different raw rice varieties after cooking

Varieties	Samples	2 % Polished Rice		8 % Polished Rice	
		Blended	Unblended	Blended	Unblended
Bw 361	Rice	VSGBC	NGBC	VSGBC	NGBC
	Rice bran	DGBC	NGBC	DGBC	NGBC
Attakari	Rice	DGBC	NGBC	MGBC	NGBC
	Rice bran	SGBC	NGBC	DGBC	NGBC
Bg 252	Rice	DGBC	NGBC	MGBC	NGBC
	Rice bran	SGBC	NGBC	MGBC	NGBC
At 362	Rice	VSGBC	NGBC	NGBC	NGBC
	Rice bran	VSGBC	NGBC	NGBC	NGBC
Ld 365	Rice	VSGBC	NGBC	VSGBC	NGBC
	Rice bran	NGBC	NGBC	NGBC	NGBC
Bg 406	Rice	VSGBC	NGBC	NGBC	NGBC
	Rice bran	GBC	NGBC	VSGBC	NGBC
Bw 364	Rice	VSGBC	NGBC	NGBC	NGBC
	Rice bran	VSGBC	NGBC	NGBC	NGBC
Ld 356	Rice	VSGBC	NGBC	VSGBC	NGBC
	Rice bran	VSGBC	NGBC	NGBC	NGBC
At 303	Rice	VSGBC	NGBC	VSGBC	NGBC
	Rice bran	VSGBC	NGBC	NGBC	NGBC

* DGBC-Dark Greenish Blue Colour, MGBC- Moderate Greenish Blue Colour, SCBS- Slight Greenish Blue Colour, VSGBC- Very Slight Greenish Blue Colour, NGBC- No Greenish Blue Colour

Bg 252 at 2% Polishing
Hard Blue Colour Spots

At 362 at 8% Polishing
No Blue Colour development

Parboiled Blended Rice (Bg 406)- at 2% Polishing
No Blue Colour Appearance

Iron Fortified Kernels
No Blue Colour Appearance

Plate 1: Blue color development of different rice varieties at different polishing levels

90%, 95% and 99.1% . It may be due to the lower available phenolic compounds and antioxidant properties of those rice varieties. The concentration of total phenolic compounds in the grain has been positively associated with the antioxidant activity (Goffman and Bergman, 2004; Itani *et al.,* 2002; Zhang *et al.,* 2006). It is also reported that the grains with a darker red pericarp colour, such as red and black rice, contain higher amounts of polyphenols and anthocyanin (Acquaviva *et al.,* 2003; Tian *et al.,* 2004; Zhou *et al.,* 2004; Tian *et al.,* 2005).

Besides the difference in the content of total phenolics related to the colour of the grains, variation was also observed in the content of total phenolics among the genotypes with the same pericarp colour (Goffman and Bergman, 2004). Minerals have also been reported to interact with anthocyanins containing vicinal hydroxyl groups causing a red to blue colour change (Clydesdale, 1991). The cooking parameters for control and fortified brown rice such as cooking time and water uptake were quite close to the control (normal rice) sample. Water uptake was higher for some rice varieties as Attakari, Bg 252 and Bg 406 compared to other rice varieties and this may be due to the internal grain structural differences.

Parboiled Rice Cooking

None of the blended parboiled rice samples developed even a trace of green colour after cooking at both 2% and 8% polished levels and the test result was “Perceptually similar”. This could be due to the reduction of phenolic compounds and antioxidant activity due to parboiling process. The polyphenols are water soluble due to their chemical characteristics. A portion of these compounds may be solubilized in the parboiling water. Goffman and Bergman (2004) and Zhang *et al.* (2006) have reported that a small part of reduction in the polyphenol concentration in the parboiled grains is due to the loss of polyphenols in the water.

Conclusion

The present study led to draw the following conclusions on the influence of blending iron and folic acid fortified kernels with red pericarp rice. The results revealed that, there is an effect of red rice bran on blue colour development during cooking of red pericarp rice. No colour development was observed for parboiled rice for all rice varieties at levels of polishing. A significant blue colour development was observed in two locally grown rice varieties; Attakari and Bg 252 when blended with iron fortified kernels at 1%. However, this change was significant with cooked raw rice which is polished to either a lower degree or to a relatively higher degree. It may be due to the reducing phenolic compounds available in the red rice bran facilitating the reduction of ferric to ferrous. Reducing ability depends on the antioxidant capacity available in the rice bran of a particular variety and polishing or parboiling reduces that capacity. Therefore further studies are needed to develop stable forms of iron compounds to be used for the fortification of some red rice varieties in future.

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Identification of Suitable Mulch for Radish (*Raphanus sativus* L.) to Cope with Temperature Stress

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Abstract: Although mulching provides numerous benefits in cropping activities, very few studies were reported on crop specific influence of different mulching materials. Thus, a pot experiment was conducted in a polytunnel to assess impact of different mulching materials on growth and yield of radish (cultivar, *Beeralu*). Treatments were arranged in two-factorial experiment in Completely Randomized Design with three replicates. Two temperature regimes viz. ambient (32-33°C) and temperature stress (35-36°C) condition and three types of mulching materials *viz*. coir dust (M2), gliricidia (M3) and straw (M4) were used as treatments. Crops without a mulch was the control (M1). Raddish seeds were sown in pots filled using reddish brown earth soil. One week after the emergence of seedlings, mulching materials were applied according to treatments. Growth parameters (number and fresh weight of leaves) and yield parameters (root fresh weight, root dry weight, root length and girth of the root) were measured. Data were statistically analyzed using SAS and mean separation was done by Least Significant Difference test. The results revealed that the temperature and mulching material and their interaction were significantly ($p<0.05$) influenced on most of growth and yield parameters tested. Highest number of leaves (17.2), leaf fresh weight (59.8 g), plant height (45.1 cm), length of tuber (16.1cm), fresh weight of tuber (77.3 g) and dry weight of tuber (18.8 g) were recorded in gilicidia mulch under the temperature stress. Coir dust mulch under the ambient temperature indicated the second best performance among the tested treatments. Lowest performance in raddish crop was recorded in temperature stress condition without having a mulch.

Keywords: Growth, Mulch, Temperature, Yield

Introduction

Agriculture is one of the main sources for income generation in Sri Lanka. When considering agriculture, it mainly depends on climatic conditions. Especially, rainfall and temperature have influence on agriculture. Irregular weather conditions result in decline of yield of agriculture products and drastic changes on cultivation. The temperature is a primary factor affecting the rate of plant development. Due to temperature stress and water stress, the food productivity is decreasing. In general, high temperature may lead to significant losses in crop productivity in many species due to limited vegetative and reproductive growth (Spears *et al.,* 1997; Cross *et al.,* 2003; Godawatte *et al.,* 2014). Yan *et al.* (2008) and Zhou *et al.* (2010) reported that the high temperature stress reduced photosynthetic capacity by accelerated leaf senescence and decreased relative chlorophyll content.

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Net photosynthetic rate was decreased by 50−60% in maize under high temperature conditions of 35/30°C (day/night) and 40/35 °C (day/night) compared to that under low-temperature conditions of 25/20°C (day/night) and 30/25°C (day/night) (Ben-Asher *et al.,* 2008). De Silva (2006) reported that the Sri Lanka's dry zone agricultural output will decline significantly in the next 20 to 30 years because of reduced rainfall and increased temperature. Chandrapala and Fernando (1995) reported that, there has been an increase in temperature in Colombo by 0.0164°C / year during the period of 1961 to 1990. According to a report of the Intergovernmental Panel on Climatic Change (IPCC) (IPCC Expert Meeting Report, 2007) the global mean temperature will rise 0.2°C per decade in the coming years. De Silva *et al.* (2007) predicted using HadCM3 general circulation model that, by 2050, rainfall will decrease by 9% to 17% in the main *Maha* cultivation season.

Radish is one of the popular root vegetables in Sri Lanka and it belongs to the family Brassicaceae. Radish is a good source of vitamin C (ascorbic acid) and minerals of calcium, potassium and phosphorus. It is known to have refreshing and diuretic properties. Radish is also used to treat for neurological, headache, sleeplessness, chronic diarrhoea, urinary complaints and piles (Dhananjaya, 2007). Radish cultivated in many parts of Sri Lanka due to its' short life span. However, only few studies were conducted to evaluate the effect of temperature stress and water stress on plant growth and yield. Shaw *et al.* (2002) reported that the water shortage strongly affects crop growth, root yield and biomass partitioning. Alfaro *et al*. (2006) reported that the maximum temperatures are affected by local conditions, especially soil water content and evaporative heat loss as soil water evaporates. Hatfield and Prueger (2011) reported that an increasing water vapor demand will cause more water to be transpired by the leaf until the water supply becomes limited and the stomatal conductance will decrease leading to a higher leaf temperatures and a reduction in photosynthesis. Evidence shows a strong correlation between climatic conditions and national agricultural growth and the growth rate declined to 1.5% in 2011 due to adverse climatic conditions.

In this study temperature stress and water stress affects the growth and yield of radish. Therefore, solutions are very important to overcome such impacts on radish imposed by high temperature and water stress. As an answer for it, use of mulching material is important to conserve soil moisture and vital to avoid wilting cases due to higher transpiration from plant and evaporation from soil under temperature stress condition. Kumar *et al.* (2014) reported that the use of mulch as a soil cover is effective in improving yield and soil fertility. Yoo-Jeong *et al.* (2003) reported that the mulching as a one of important agronomic practice beneficial in conserving soil moisture, suppressing the weeds, improving soil fertility, and modifying the soil physical and chemical environment. Knavel and Mohar (1967) mentioned that the moisture distribution in the upper soil layer is more uniform compared with un-mulched soil and more roots developed in the upper soil layer which usually has richer nutrients and useful microorganisms. Also, positive response of mulch has been reported. Different mulching materials have different effectiveness for enhancing performance because of their capacities in absorbing moisture due to their aggregate nature in allowing air circulation. Mulching could change the physical and chemical environment of the soil resulting in increased availability of phosphorus and potassium (Muralidharan and Kamalam, 1973).

The study intends to identify the suitable mulch to mitigate the consequences of higher temperature stress due to unexpected weather events by evaluating the effect on growth and yield parameters of Raddish variety *Beeralu rabu*.

Materials and Methods

Collection and preparation of samples

The study was conducted under the conditions of two protected structures i.e. a polytunnel and a net house at the Open University of Sri Lanka (Figure 1). The treatments were designed in a two-factor factorial experiment in Completely Randomized Design with three replicates. One set of plants kept in temperature regulated polytunnel (35 – 36°C) and the other set of plants were kept in ambient air temperature (32- 33°C) at the net house. The average diurnal fluctuation of temperature within the polytunnel and net house interior is shown in the Figure 2. It is indicated that the temperature at the polytunnel was higher than the temperature at the net house. Godawatta and De Silva (2014) reported that there were no significant differences in RH observed in the inside and outside environment although elevated day time air temperatures in the poly tunnel resulted in higher partial pressure of water. This condition was regulated with the open structure of the top vent roof structure. Same polytunnel with similar conditions was used in this study.

The pots (40 cm in diameter with 45 cm depth) were filled using compost and red-dish-brown earth soil in the ratio of 1:5. Seed were sown in individual pots and excess plants were removed to maintain 3 plants/pot at one week after seedling emergence. Coir dust, straw and gliricidia were used as mulching materials.

Figure 1: Polytunnel with open top

Front view of the net house

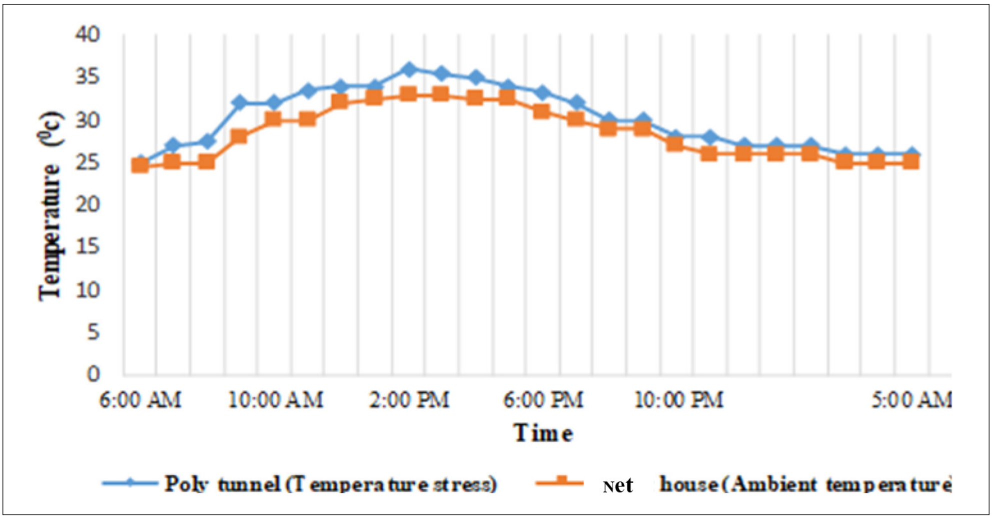

Figure 2: Temperature variations inside of the polytunnel and net house

Mulches were applied as a uniform layers to an average depth of 2.2 inches each on the soil surface. A pot without mulch was used as the control in the experiment. Soil moisture in all pots was maintained at field capacity throughout the growing season to avoid the water stress for plants. Fertilizers were applied according to the recommendation of Department of Agriculture, Sri Lanka. Plot layout of the study is indicated in the Figure 3.

Data collection

Plant growth parameters were measured at two weeks interval. Plant height (cm) was measured from each replicate by measuring the height from ground level to the terminal growing point of the shoot. The number of leaves of the plants, whose plant heights were taken, was recorded on weekly basis. Leaf width was measured by using Vernier caliper in three replicates.

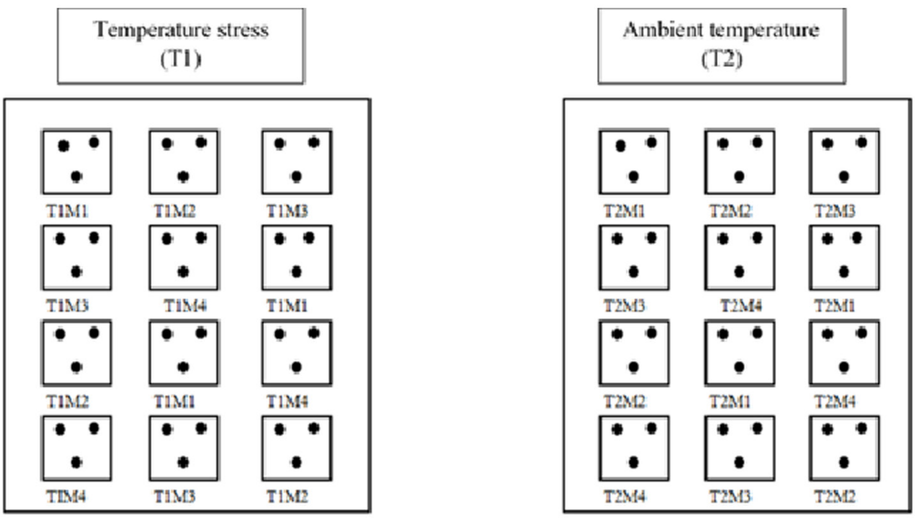

Figure 3: Layout of the study

M1- Control M2- Coir dust M3- Gliricidia M4- Straw

Leaf area was measured from randomly selected leaves from each replicate and grid counting method was used (Chaudhary *et al.,* 2012). Yield parameters were measured after harvest. Fresh weight of root (g) was measured by using electronic balance. Leafy part and other soil and debris were removed to measure fresh weight of roots. Weights of roots in all replicates were measured. The length of root (cm) from each replicate was recorded.

Data Analysis

All the data will be analyzed by using SAS software and multiple comparison of the various means were carried out by LSD (Least Significant Difference) test at P = 0.05.

Results and Discussion

Average number of leaves

Number of leaves has shown an increasing trend pattern during the study period. Effect of temperature stress, mulching material and interaction between temperature and mulching material were significant ($p<0.05$) on average number of leaves (Figure 4). All the treatment at ambient temperature has shown better performances and the treatment with gliricidia mulch at ambient temperature showed the highest number of leaves.

Treatment with straw at ambient temperature showed the second highest number of leaves. Treatment with gliricidia mulch in temperature stress has shown better performances than the other treatments at temperature stress. Treatments without mulch have shown the lowest number of leaves either at temperature stress or ambient temperature (Figure 4).

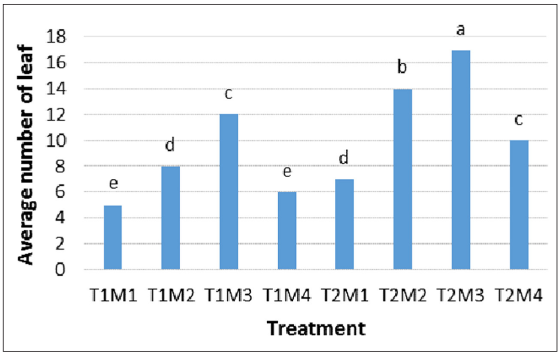

Figure 4: Number of leaves at harvest

T1- Temperature stress T2- Ambient temperature M1- Control M2- Coir dust M3- Gliricidia M4- Straw

Values followed by same letter are not significantly different at p =0.05 level

Average Leaf area

Effect of temperature stress, mulching material were significant ($p<0.05$) on leaf area (Figure 5). However, effect of interaction between temperature and mulching materials was not significant on measured parameter. Treatments with gliricidia mulch have shown the highest leaf area.

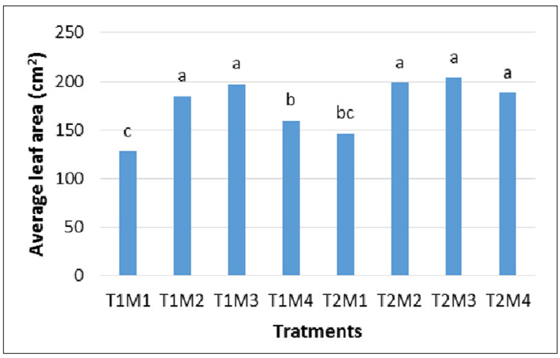

Figure 5: Average leaf area

T1- Temperature stress T2- Ambient temperature M1- Control M2- Coir dust M3- Gliricidia M4- Straw

Values followed by same letter are not significantly different at p =0.05 level

But it was not significantly different from the treatments with coir dust either at temperature stress or ambient temperature and the treatments with straw at ambient temperature. Treatments without a mulch presented the lowest leaf area at temperature stress and as well as at ambient temperature.

Average fresh weight of leaves

Average fresh weight of leaves of radish due to the effect of mulching material, temperature stress and the interaction between temperature stress and mulching material showed a significant variation (Figure 6).

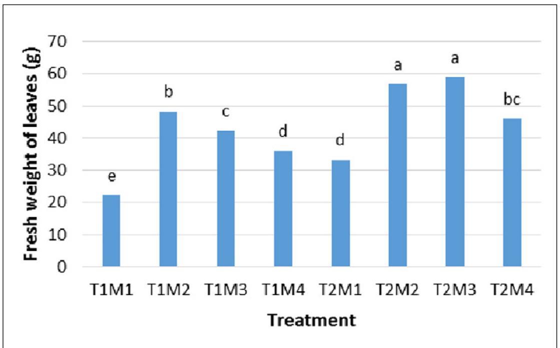

Figure 6: Average fresh weight of leaves
T1- Temperature stress T2- Ambient temperature M1- Control M2- Coir dust M3- Gliricidia M4- Straw
Values followed by same letter are not significantly different at p =0.05 level

All the treatments at ambient temperature have shown the better performances than treatments under temperature stress conditions. Treatment with gliricidia mulch at ambient temperature gave the highest fresh weight of leaves. Maximum fresh weight of leaves has obtained from gliricidia mulch at ambient temperature which was statistically similar to that of coir dust mulch at ambient temperature. Non-mulched treatment at temperature stress as well as at ambient temperature resulted lower weight of fresh leaves.

Plant height

Plant height has shown a pattern of increment. Effect of temperature, mulching material and the interaction effect of temperature and mulching materials were significant ($p<0.05$) on plant height (Figure 7).

The tallest plants were obtained from gliricidia mulch treatment at ambient temperature and it was not significantly different from the treatment with coir dust at ambient temperature. The plants grown without mulch at temperature stress gave minimum plant height (Figure 8). Mohanty *et al.* (1991) reported that the plant height was low in treatment without mulch which agrees with this study.

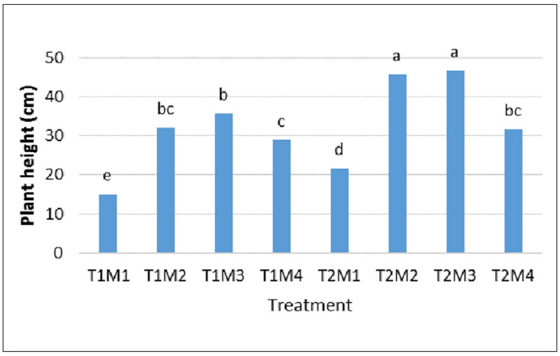

Figure 7: Plant height
T1- Temperature stress T2- Ambient temperature M1- Control M2- Coir dust M3- Gliricidia M4- Straw
Values followed by same letter are not significantly different at p =0.05 level

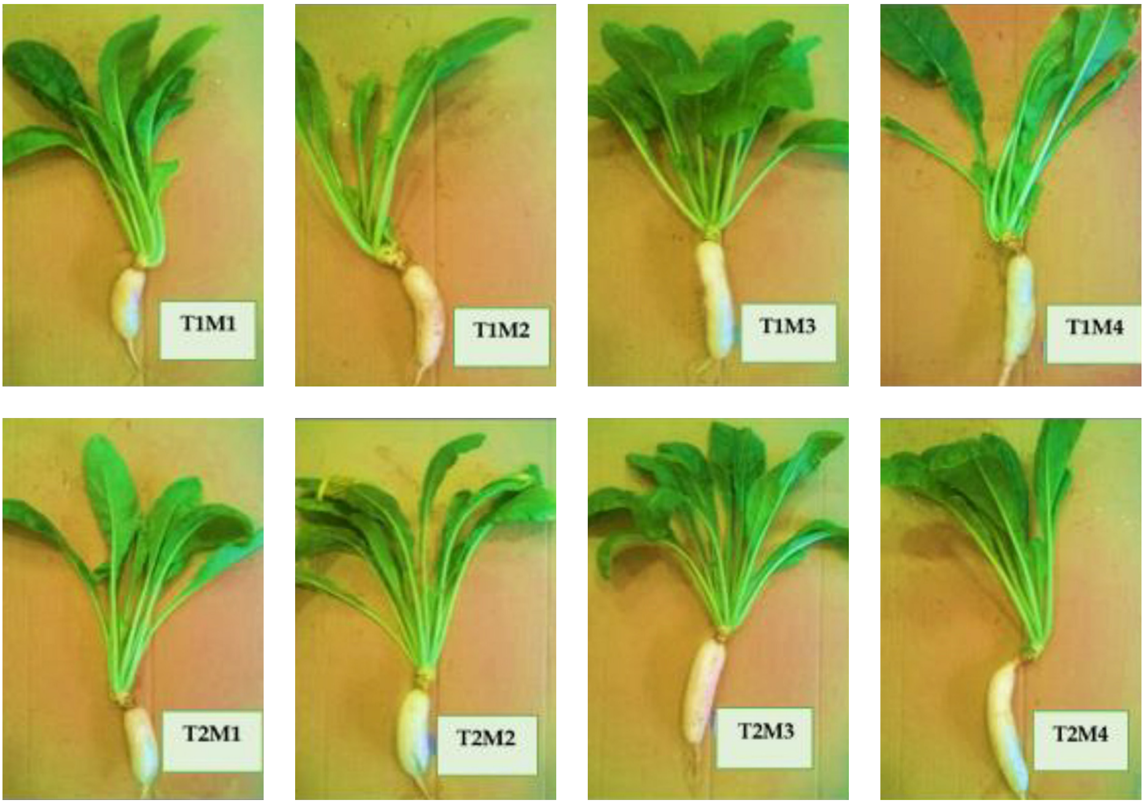

Figure 8: Growth performances of radish at harvest
T1- Temperature stress T2- Ambient temperature M1- Control M2- Coir dust
M3- Gliricidia M4- Straw

Length of Tuber

Effect of temperature, mulching material and their interaction significantly influenced ($p<0.05$) on length of tuber (Figure 9). Treatment with gliricidia at ambient temperature has shown the longest tuber and it was significantly different ($p<0.05$) from all other treatments. Treatments without mulch at temperature stress showed the lowest tuber length.

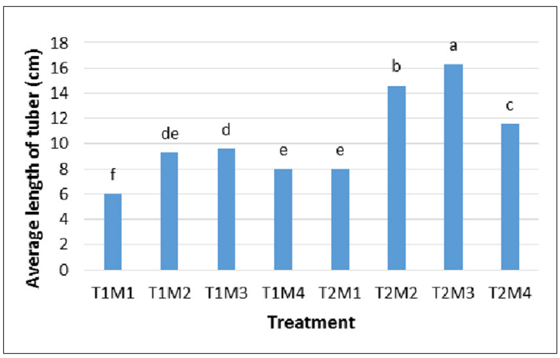

Figure 9: Root length
T1- Temperature stress T2- Ambient temperature M1- Control M2- Coir dust
M3- Gliricidia M4- Straw
Values followed by same letter are not significantly different at p =0.05 level

Girth of the tuber

Girth of the tuber was significantly varied ($p<0.05$) according to temperature and mulching material. However, the interaction effect was not significant ($p>0.05$) on the girth of the tuber

(Figure 10). Treatments at ambient temperature with gliricidia and coir dust have shown the higher value for girth and both of these treatments were not significantly differ from each. Third highest girth of the tuber has shown in temperature stress with gliricidia mulch.

Treatments at ambient temperature have shown higher performances than the treatments at temperature stress. Treatment without mulch at temperature stress has shown the lowest girth of tuber and it was significantly varied from all other treatments.

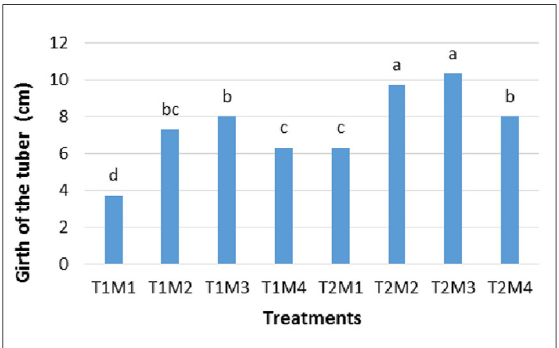

Figure 10: Girth of tuber
T1- Temperature stress T2- Ambient temperature M1- Control M2- Coir dust M3- Gliricidia M4- Straw
Values followed by same letter are not significantly different at p =0.05 level

Fresh weight of tubers

All the treatments at ambient temperature have shown better performances than the treatments at temperature stress. According to Table 1, treatment with gliricidia mulch at ambient temperature has shown the highest fresh weight and it was significantly different ($p<0.05$) from all other treatments apart from the treatment with coir dust at ambient temperature. Treatment with temperature stress and gliricidia mulch showed the third highest fresh weight of tubers.

Kumar *et al.* (2014) reported that the gliricidia mulch was produced pronounced efect ($p<0.05$) with regard to yield of ginger against the other treatments. All the control treatments at temperature stress and ambient temperature have shown the lowest fresh weight due to absence of mulch. Treatment without mulch at temperature stress has shown the lowest fresh weight and it was significantly different from all other treatments.

Dry weight of tubers

Dry weight of tubers also varied approximately same manner as fresh weight (Table 1). Dry weight of tuber also significantly varied ($p<0.05$) according to the both factors (Temperature and Mulching material) and interaction effect of temperature and mulching material. As similar to the fresh weight variation, all the treatments which were kept at ambient temperature have shown better performances than the treatments which were kept at temperature stress.

Treatment at ambient temperature with gliricidia mulch has shown the highest tuber dry weight and it was significantly different from all other treatments apart from the treatment with coir dust mulch at ambient temperature which has shown second highest dry tuber weight. Treatment without mulch at temperature stress has shown the significantly lowest dry weight of roots.

Table 1: Fresh and Dry weight of tubers

Treatment	Tuber fresh weight (g)	Tuber dry weight (g)
T1M1	32.00^{g}	6.00^{f}
T1M2	55.00^{d}	11.55^{de}
T1M3	67.66^{bc}	13.83^{cd}
T1M4	50.00^{e}	9.16^{ef}
T2M1	41.00^{f}	8.73^{f}
T2M2	74.00^{ab}	16.5^{ab}
T2M3	77.33^{a}	18.83^{a}
T2M4	62.00^{cd}	15.5b^{c}
CV (%)	5.43	7.158

T1- Temperature stress T2- Ambient temperature M1- Control M2- Coir dust M3- Gliricidia M4- Straw

Values followed by same letter are not significantly different at p =0.05 level

Conclusion

This study showed that the mulches of gliricida, coir dust and straw improved the yield of radish at temperature stress and ambient temperature. All the treatments without mulch have shown the lowest growth and yield performance either at the temperature stress or at ambient temperature. The consistent good results showed under the gliricidia and coir dust mulches for tested parameters in both ambient temperature and temperature stress condition and the crop recorded a significantly higher yield than the other treatments. Even though there is temperature stress to plants, along with gliricidia mulch, yield could be obtained without a significant reduction. These findings of this study will help the farmers in dry zone to cope with temperature stress in coming years due to climate change.

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Effect of Reverse Osmosis Wastewater on Seed Germination and seedling performance of four Different Crops

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Abstract: Wastewater reclamation and its reuse for beneficial purposes is a common goal of many countries particularly in water stressed countries around the world. Cutting edge technologies such as reverse osmosis (RO), micro- and ultra-filtration are often used to purify underground water and wastewater generated from the industries. Wastewater is generated during the process of water purification with RO. However, the wastewater from these processes is usually laden with concentrated nutrients, salts, and other materials. Using RO rejection water as irrigation water for agriculture is a viable alternative. Hence, a study was conducted to assess the suitability of five different level of diluted RO wastewater along with control treatment (100% pure water) on seed germination and seedling performance of Amaranthus, cabbage, green gram and maize. This study was carried out in the laboratory with completely randomized design comprising three replicates. Purified water was used as control and used to prepare dilution levels of 20%, 40%, 60% and 80% along with RO wastewater. Seed germination percentage, germination time, shoot length, root length and fresh weight were recorded seven days after sowing and analyzed statistically. Significant differences ($p<0.05$) were found among treatments on germination percentage, germination time, root length, shoot length and fresh weight. Germination percentage decreased with the increasing percentage of RO rejected wastewater, while other parameters have shown slight fluctuations. Amaranthus and maize performed well with the combination of 40% wastewater and 60% purified water. Meanwhile, combination of 20% wastewater and 80% purified water showed positive effect on the germination of green gram (96.67%) and maize (100%). Furthermore, seven water quality parameters have shown a linear relationship with the fresh weight of sprouts. The results indicated that there is a possibility to use diluted reverse osmosis wastewater as source of irrigation water. Further, field studies needed to recommend the usage of RO rejected wastewater as part of irrigation water.

Keywords: Germination, Irrigation, Reverse osmosis, Wastewater

Introduction

A scarcity of fresh water limits sustainable agriculture development worldwide (Wang *et al.,* 2017). It is estimated that about 2/3 of world population will face a water deficiency at medium or high level by the year 2025.World total water resources is 1.4 billion m^3 and 1% of the water can only be used as drinking water (Bozdogan, 2015). Groundwater is the major source of water especially in rural areas, and it is estimated that about 72% of the rural

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population relies on groundwater for domestic use (Central bank report, 2010).

Eco-soft reverse osmosis system can be used to demineralize low to medium salinity water. Usually Eco-soft reverse osmosis plant is desalinated 10000 (± 10%) L/day. Reverse osmosis (RO) plants reject 15 - 20% of the feed water as waste of concentrated saline streams which considered as major environmental and economic drawback of the RO process (Ghamdi, 2016). Various alternative options used for the treatment and disposal of generated concentrate including deep well injection, evaporation ponds, disposal into surface water bodies, disposal through pipelines to municipal sewer systems, ion exchange procedures, shrimp breeding and hydroponic cultivation of salt tolerant plants (Qurie *et al.,* 2013).

Wastewater repossession and its reuse for beneficial purposes is a common aim of many countries particularly in water stressed countries around the globe. Cutting edge technologies such as RO, micro and ultra-filtration are often used to filter wastewater effluent generated in traditional systems. However, the rejected wastewater from these processes is usually laden with concentrated nutrients, salts, inorganic materials while it is rich in organic materials those are removed in the course of treatment (Shahalam *et al.,* 2010).

Using RO rejection water as irrigation water for agriculture is a viable alternative. In the present study RO rejected wastewater from an Eco-soft reverse osmosis plant was used as irrigation source with several dilutions with pure water to assess the suitability of Reverse Osmosis rejection water and its different dilution levels of water as source of irrigation for different crops such as Amaranthus, cabbage, green gram and maize.

Materials and Methods

Sample collection and preparation

The feed water was taken from the ground well, which is located in the Kilinochchi premises of the University of Jaffna. Reverse osmosis desalination (RO) plant is located in the Faculty of Agriculture, University of Jaffna, Kilinochchi premises. The RO plant consists of two membranes for desalination with a capacity of 6000 (±10 %) L/ day.Electric conductivity of the feed water is about 3.25 dS/m.

The different concentrations (20%, 40%, 60% and 80%) of wastewater (Table 1) were prepared by mixing reverse osmosis rejected water with pure water. Reverse osmosis wastewater (100%) and its dilutions (20%, 40%, 60% and 80%) were analyzed for electrical conductivity (EC), sodium (Na^+), potassium (K^+), hardness (calcium plus magnesium), alkalinity, phosphate and sulphate in triplicate.

Table 1: Treatments and different concentrations of RO wastewater

Treatment	Concentrations of RO wastewater
T_1 (control)	0%
T_2	20%
T_3	40%
T_4	60%
T_5	80%
T_6	100%

Germination tests

Germination test was carried out in laboratory conditions with three replicate and Completely Randomized Design. Seeds were surface-sterilized with 5% sodium hypochlorite (NaOCl) (Sauer and Burroughs, 1986) and washed thoroughly using distilled water. All petridishes were washed by tap water, followed by rinsing with distilled water. Hot air sterilizer was used for sterilization at 170^{o}C in 4 hours (Muhammad and Hussain, 2010).

Ten seeds were put in each petridish on one- layer filter paper. Initially, the seeds were immersed with 5 mL of appropriate solution in petri dish. Then, 1 mL of prepared solution was added to each Petri dish every day to provide moisture for the germination and was monitored daily.

Data collection

When the radical was two mm in length, it was considered as germinated seed (Panuccio *et al.,* 2014). Data were recorded at seven days after seed placement. The germination percentage, mean germination time, root length, shoot length and fresh weight of the germinated seeds were measured.

Statistical Analysis

The collected data were subjected to analysis of variance. Dunnett's test was used to compare mean at 5% probability level.

Moreover, correlation and regression analysis were performed to find the strength and relationship between the water quality parameters and seed germination and seedling performance.

Results and Discussions

Impact of different concentration of RO wastewater on seed germination and seedling performance

The highest germination percentage (80%), shoot length (2.16 cm), root length (1.25 cm) and fresh weight (0.06 g) were recorded in the (T_1) control treatment where the sole pure water was used as the irrigation source. Despite, the lowest mean germination time (2.33) was noted in the T_1. Amaranthus has shown significant difference ($p<0.05$) on germination percentage, shoot length, mean germination time and fresh weight.

However, there was no any significant ($p<0.05$) difference was recorded on the shoot length and fresh weight of sprout up to the combination of 40% wastewater and 60% pure water (T_3). These results indicated that 60% and 40% pure and RO wastewater combination were performed well for Amaranthus. This might be due to the undisturbed physiological activities of the seeds. Therefore that level could be used for irrigation without any impact on growth of germinated seed.

Salinity stress had a significant impact on the growth of different Amaranthus cultivars, where the yield was reduced from 17.8% to 25.2% for A0020 cultivar; from 6.9% to 28.3% for A0057 cultivar, from 3.6% to 8.6% for A211cultivar respectively for 3 dS/m and 6 dS/m compared to the control (0.92 dS/m). While, the same wastewater had been used since beginning of study, which ensures the suitability of diluted wastewater irrigation for the seed germination and seedling growth of Amaranthus (El Youssfi *et al.,* 2012)

Table 2: The effect of different dilution levels of RO wastewater on seed germination of Amaranthus.

Treatment	Germination %	Mean germination time	Root length (cm)	Shoot length (cm)	Fresh weight(g)
T_1 (100% control)	80	2.33	1.25	2.16	0.06
T_2 (80% pure:20% RO)	70	3.45	0.72	1.61	0.06
T_3 (60% pure:40% RO)	70	3.41	0.79	0.96	0.05
T_4 (40% pure:60% RO)	66.67	3.45	0.81	0.81*	0.04*
T_5(20% pure:80% RO)	53.33	3.85*	0.93	0.73*	0.04*
T_6 (100% RO)	40*	5.36*	1.18	0.39*	0.03*
F test	**	**	NS	**	**

*Dunnett's test significant at α = 0.05 level is indicated by *; F test significant at α = 0.05 level is indicated by **; NS- Not significant*

Table 3: The effect of different dilution levels of RO wastewater on seed germination of Cabbage

Treatment	Germination %	Mean germination time	Root length (cm)	Shoot length (cm)	Fresh weight(g)
T_1 (100% control)	83.33	2.68	10.35	3.45	1.77
T_2 (80% pure:20% RO)	73.33	3.05	9.19	2.83	1.06
T_3 (60% pure:40% RO)	73.33	3.53*	8.32*	2.53	1.08
T_4 (40% pure:60% RO)	66.67	3.79*	8.58*	2.85	0.94*
T_5 (20% pure:80% RO)	66.67	4.30*	7.52*	2.05	0.97*
T_6 (100% RO)	66.67	4.13*	7.33*	1.47*	0.78*
F test	NS	**	**	**	**

*Dunnett's test significant at α = 0.05 level is indicated by *; F test significant at α = 0.05 level is indicated by **; NS- Not significant*

Table 3 shows the germination parameters of cabbage. The significant difference ($p<0.05$) was observed for all the parameter except germination percentage. The highest germination percentage (83.33%), shoot length (10.35 cm), root length (3.45 cm) and fresh weight (1.77 g) were recorded in the T_1. However, there was no any significant difference ($p<0.05$) observed up to combination of 80% and 20% of pure water (T_2) and RO wastewater respectively for all the parameters. Kiziloglu1 *et al.* (2007) stated that cabbage could perform well under wastewater irrigated field (3510 ± 54.2 kg ha^{-1}) than non-wastewater irrigated land (2780 ± 42.1 kg ha^{-1}). If cabbage can survive with the wastewater application as irrigation source at the germination and seedling stage, it would provide beneficial agronomic and yield

Table 4: The effect of different dilution levels of RO wastewater on seed germination of green gram.

Treatment	Germination %	Mean germination time	Root length (cm)	Shoot length (cm)	Fresh weight(g)
T_1 (100% control)	100	1.03	6.78	16.48	3.97
T_2 (80% pure:20% RO)	96.67	1.07	5.7	15.16	3.87
T_3 (60% pure:40% RO)	83.33	1.2	4.52	9.66*	3.73*
T_4 (40% pure:60% RO)	80	1.62*	5.28	8.33*	3.23*
T_5 (20% pure:80% RO)	80	1.62*	5.62	7.57*	3.23*
T_6 (100% RO)	80	3.14*	3.37*	5.81*	3.43*
F test	NS	**	**	**	**

*Dunnett's test significant at α = 0.05 level is indicated by *; F test significant at α = 0.05 level is indicated by **; NS- Not significant*

attributes at the reproductive and harvesting stage. Hence, application of diluted wastewater could enhance the plant growth, reduce the requirement of fertilizer application and enhance the productivity of marginal soil.

The significant difference ($p<0.05$) was observed in germination parameter of green gram (Table 4). The lowest performance was recorded in the T_6, where the 100% of wastewater was used as the treatment that inhibits the physiological process of the green gram. Moreover, there was no any significant difference ($p<0.05$) observed up to combination of 80% pure water and 20% RO wastewater (T_2) for all the parameters. In this treatment, the germination percentage of maize seeds and shoot length of sprouts have recorded as 96.67% and 15.16 cm, respectively.

Similar results have reported from the previous studies, where the germination percentage was dropped due to the raising concentration of sugar mill effluent. Higher amount of solids found in the effluent leads to manipulations in osmotic relationship of seed and water, which leads to reduction in absorption of water by the seed. Furthermore, this can be altered by the effluent salinity as well (Baskaran *et al.,* 2009). Moreover, Divya *et al.* (2015) reported that the maximum germination percentage and seedling performance could be achieved at lesser dilutions of 10–30% for the wastewater.

The germination parameters of maize is given in table 5. The significant difference ($p<0.05$ was observed for all the parameter except germination percentage in the germination study of maize. The highest germination percentage 100%), shoot length (16.7 cm), root length (9.94 cm) and fresh weight (11.31 g) were recorded in the treatment 1.

However, there was no any significant difference ($p<0.05$) observed up to

Table 5: The effect of different dilution levels of RO wastewater on seed germination of maize.

Treatment	Germination %	Mean germination time	Root length (cm)	Shoot length (cm)	Fresh weight(g)
T_1 (100% control)	100	1.77	16.7	9.94	11.31
T_2 (80% pure:20% RO)	100	2.07	15.32	9	10.54
T_3 (60% pure:40% RO)	100	2.87	14.21	8.24	9.51
T_4 (40% pure:60% RO)	100	3.17*	10.77*	6.02*	10.5
T_5 (20% pure:80% RO)	100	3.27*	7.84*	4.23*	9.5
T_6 (100% RO)	96.67	3.37*	11.80*	3.97*	8.64*
F Test	NS	**	**	**	**

combination of 60% and 40% of pure water and RO wastewater respectively for all the parameters. Comparison of corn yields obtained from plots irrigated with effluent (1.46 dS/m) and fresh water (0.51 dS/m) demonstrated that the high quantity of corn gained from the plot which was irrigated by the effluent. This could be due to the availability plant nutrient and dormancy breaking substances ability in the source of irrigation. (Hassanli *et al*., 2009)

Measures of relationships between water quality parameters and seed germination and seeding performance

Table 6 shows the chemical quality of the different combination of the RO wastewater and the pure water. Electric conductivity was increased with increasing percentage of reverse osmosis rejected water. Electric conductivity of control treatment and 100 % of RO rejected water were recorded as 0.38 dS/m and 11.34 dS/m, respectively. Total sodium, potassium, phosphate and sulphate were increased with increasing concentration of RO wastewater which may be due to presence of salt ions in the water. Water hardness and alkalinity were increased with increasing RO wastewater amount due to presence of calcium, magnesium and carbonates in the water.

To study the patterns of the relationships between the water quality variables and germination and seedling performance of Amaranthus, cabbage, maize, and green gram, the Pearson's correlation

Table 6: Water quality parameters of different treatments

Treatment	EC (dS/m)	Na^+ (ppm)	K^+ (ppm)	Hardness (ppm)	Alkalinity (ppm)	PO_4^{3-} (ppm)	SO_4^{2-} (ppm)
T_1	0.38	51.24	22.52	55	0	3	1
T_2	2.91	153.73	23.51	850	84	9.57	38
T_3	5.13	237.31	24.01	1500	132	11.57	59
T_4	7.37	316.92	24.5	1950	184	12.71	90
T_5	9.34	355.72	24.75	2075	246	13.86	127
T_6	11.34	419.4	25.25	2550	384	14.43	161

coefficient (r) was used. The correlation coefficient (Table 7) shows the linear relationship between initial growth of germinated seeds and seven water quality parameters. No significant relationships ($p<0.05$) were seen between root length of Amaranthus and water quality parameter except concentration of PO_4^{3-}, while this was shown weak negative correlations, respectively. Root length of cabbage has shown strong but negative correlation with EC, hardness, alkalinity and concentration of cation and anion of RO wastewater.

Furthermore, EC and concentration of anions and cations of wastewater have moderately and negatively correlated with root length of green gram, while strong negative relationships were recorded in between the hardness and alkalinity of the water with root length of green gram. There were strong negative relationships were found in between the root length of maize and EC, Na^+ and K^+ concentration, hardness and concentration of SO_4^{2-} of the irrigated wastewater. Whereas PO_4^{3-} concentration and alkalinity moderately but negatively related with root length of maize. Strong positive relationships were observed in between the quality of the irrigation water and time taken for the seed germination of four different crops.

However, average germination time of green gram seeds has given moderately positive relationship with PO_4^{3-} concentration of water. There was significant relationship observed at 0.05 levels of significance of water quality variables and shoot length of seeds of cabbage, maize, Amaranthus and green gram, which could be recorded with strong negative relationship. The fresh weight of the germinated seeds of four crops were strongly but negatively correlated with water quality parameters. Meanwhile, Zhao *et al.* (2014) stated that the concentration of Na^+ had significantly impact on the germination of *Sorghum bicolor* (L.).

Influence of the water quality on fresh weight of sprout

Electrical Conductivity (EC):

Fresh weight of sprouts of Amaranthus, cabbage, maize and green gram have slightly decreased with increasing EC. The significance ($p<0.05$) of EC on fresh weight of sprouts of all the crops have illustrated in figure 1, while the maize and cabbage have exhibited the highest and the lowest sensitivity to the increasing EC of irrigation water, respectively. The principal effect of increasing EC of irrigation water on crop productivity is the incapability of the plant to compete with ions in the soil solution for water. The negative effect of EC on germination was reported by many authors from several plant species (Cavalcante *et al.,* 2005; Mostafa *et al.,* 2012).

Salt stress:

The negative effect of Na^+ and K^+ are given in figure 2 and figure 3, respectively. Salinity can enhance the osmotic pressure, which disrupts the absorption of water at the completion of cell division and differentiation, and length of plumule and radicle could be decreased due to the rising Na^+ and K^+ concentration of irrigation water (Eskandari

Table 7: Correlation coefficient between water quality parameters and seed germination and seedling parameters

Parameter	*Amaranthus*				Cabbage				Green gram				Maize			
	Mean germination time	Root length (cm)	Shoot length (cm)	Fresh weight (g)	Mean germination time	Root length (cm)	Shoot length (cm)	Fresh weight (g)	Mean germination time	Root length (cm)	Shoot length (cm)	Fresh weight (g)	Mean germination time	Root length (cm)	Shoot length (cm)	Fresh weight (g)
EC (dS/m)	0.8844	0.0301	-0.9639	-0.9684	0.9678	-0.9549	-0.9148	-0.8493	0.8346	-0.7634	-0.9636	-0.8554	0.9543	-0.8343	-0.9787	-0.8606
Na^+ (ppm)	0.8673	-0.0525	-0.9822	-0.9506	0.9693	-0.9568	-0.8913	-0.8820	0.8017	-0.7754	-0.9750	-0.8723	0.9721	-0.8372	-0.9633	-0.8498
K^+ (ppm)	0.8836	-0.1200	-0.9855	-0.9384	0.9579	-0.9647	-0.8992	-0.9205	0.7875	-0.7999	-0.9621	-0.8476	0.9614	-0.8144	-0.9427	-0.8625
Hardness (ppm)	0.8608	-0.1170	-0.9938	-0.9277	0.9584	-0.9578	-0.8793	-0.9066	0.7780	-0.8043	-0.9781	-0.8588	0.9773	-0.8135	-0.9356	-0.8554
Alkalinity (ppm)	0.9556	0.1404	-0.9234	-0.9848	0.8978	-0.9240	-0.9531	-0.8241	0.9227	-0.8298	-0.9135	-0.7397	0.8846	-0.7110	-0.9409	-0.8926
PO_4^{3-} (ppm)	0.8085	-0.3607	-0.9637	-0.8270	0.9261	-0.9543	-0.8389	-0.9685	0.6265	-0.7619	-0.9112	-0.8129	0.9295	-0.7999	-0.8576	-0.8286
SO_4^{2-} (ppm)	0.9103	0.0866	-0.9377	-0.9806	0.9498	-0.9458	-0.9359	-0.8316	0.8651	-0.7587	-0.9353	-0.8199	0.9201	-0.8102	-0.9792	-0.8668

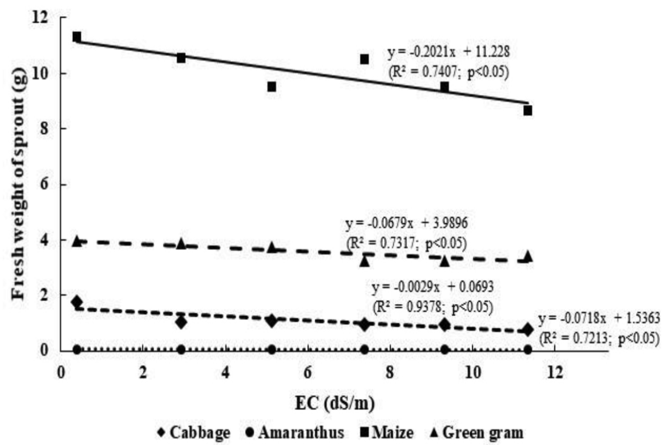

Figure 1. Fresh weight of sprout as affected by the EC of water

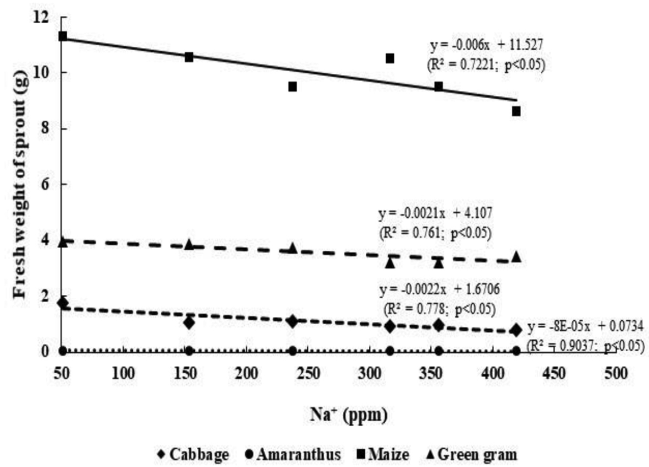

Figure 2. Relationship between fresh weight of sprout and Na^+

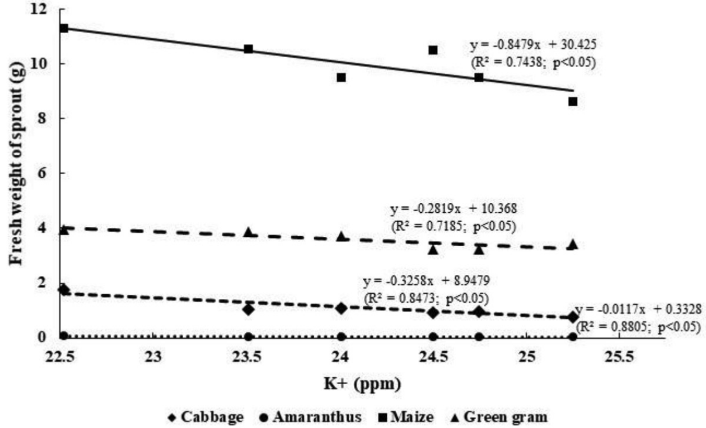

Figure 3. Relationship between fresh weight of sprout and K^+ concentration

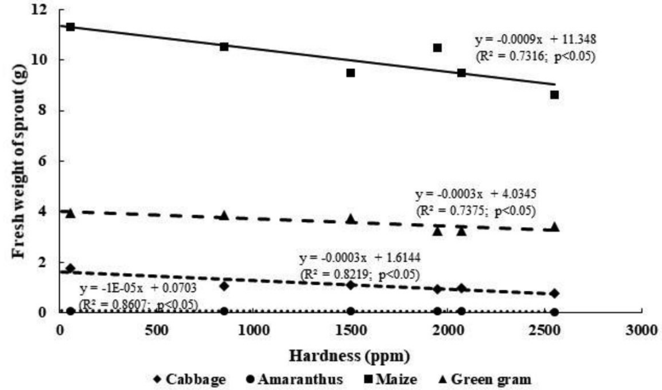

Figure 4. Relationship between fresh weight of sprouts and hardness of water

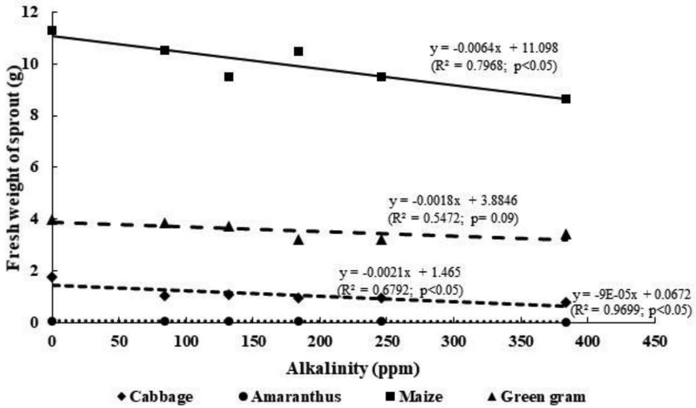

Figure 5. Relationship between fresh weight of sprouts and alkalinity of water

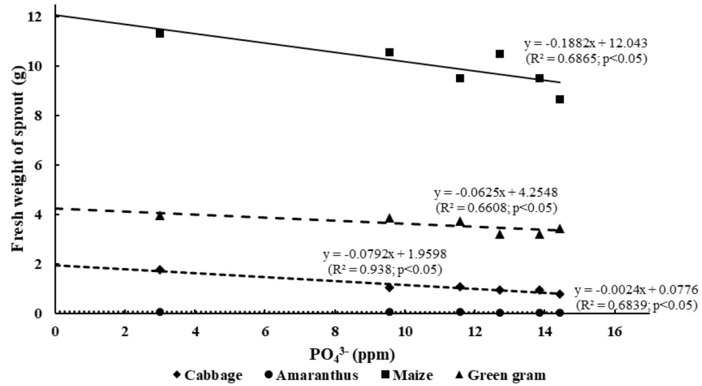

Figure 6. Relationship between fresh weight of sprouts and PO_4^{3-} concentration

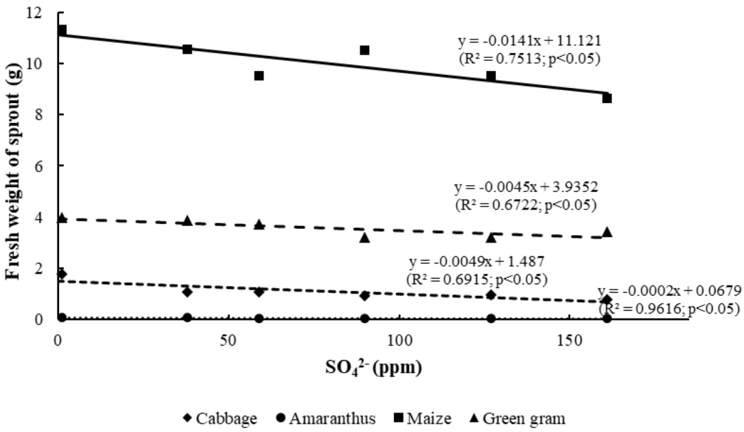

Figure 7. Relationship between fresh weight of sprouts and SO_4^{2-} concentration

and Kazemi, 2011). Among seed of four different plant species, fresh weight of germinated maize seeds have considerably decreased with increasing Na^+ (y = -0.006x + 11.527) and K^+ (y = -0.8479x + 30.425) concentrations. On the other hand, sprouted seeds of Amaranthus have shown slight decrease of fresh weight with increasing concentration of Na^+ and K^+, where the increase of one unit concentration of Na^+ and K^+ have reduced the fresh weight by 8.45×10^{-5} and 0.0117, respectively. From previous studies, similar relationship had been reported on the fresh weight of alfalfa sprouts (*Medicago sativa* L.) with salinity (Zhang *et al*., 2017).

Hardness:

Hard water contains high quantity of dissolved calcium and magnesium (Sengupta, 2013). Compare to other water quality parameters, hardness of irrigation water has less effect on the fresh weight of the sprouts, where the increment of one unit hardness drop the fresh weight by less than 0.001 g. Fresh weight of maize sprouts have shown a significant ($p<0.05$) linear relationship (y = -0.0009x + 11.348; R^2 = 0.73) with the water hardness (Figure 4). Meanwhile, similar pattern of relationship have observed on fresh weight of cabbage and green gram with water hardness as y = -0.0003x + 1.6144 and y = -0.0003x + 4.0345, respectively. Although, Salahshoor and Kazemi (2016) found that the impact of calcium on reducing the salt stress in seed germination and early growth stage of *Festuca ovina* L., the salinity can dominate than the concentration of calcium, where the seed germination and early growth could be altered even with increasing hardness of irrigation water (hardness 2). Furthermore, considerable abnormalities in seedlings of Haloxylonammodendron had recorded while treating with Mg^{2+} salts. This had happened due to the disruption of membrane permeability and functions of the plasma membrane and cell wall (Tobe *et al*., 2004).

Alkalinity:

The fresh weights of sprouts have significantly ($p<0.05$) decreased with alkalinity stress except fresh weight of green gram sprouts (Figure 5).The highest and the lowest sensitivity have noted on fresh weights of maize and Amaranthus, respectively with the alkalinity.

Furthermore, relationship of alkalinity with fresh weight of germinated seeds of maize and Amaranthus can be estimated by using linear equations $y = -0.0064x + 11.098$ and $y = -9 \times 10\text{-}5x + 0.0672$, respectively. Some sesame cultivars also have given the similar trend for the alkalinity stress, where the dry weight of sesame cultivars have declined with rising alkalinity stress (Mahdavi, 2016). The impact of alkaline stress generally added the impact on high pH, which can obstruct the ionic uptake and ionic balance of plant cells (Lin *et al.,* 2012).

Concentration of PO_4^{3-} and SO_4^{2-}:

The concentration of PO_4^{3-} and SO_4^{2-} of wastewater are given in figure 6 and figure 7, respectively. Sprout fresh weights of Amaranthus, cabbage, maizeand green gram have decreased with increasing concentration of PO_4^{3-} and SO_4^{2-}. Fresh weight of germinated seeds of Amaranthusis minimally threatened by both PO_4^{3-} ($y = -0.0024x + 0.0776$) and SO_4^{2-} ($y = -0.0002x + 0.0679$) concentration, while fresh weight of maize sprout has been adversely altered by the concentration of both anions (PO_4^{3-}; $y = -0.1882x + 12.04$, SO_4^{2-}; $y = -0.0024x + 0.0776$). This could be due to the occurrence of osmotic imbalance with high concentration of salts (PO_4^{3-} and SO_4^{2-}), which leads to poor germination and seedling performance.

Conclusions

The study showed that the combination of 40% of RO rejected wastewater and 60% pure water recorded good results on germination and seedling growth of Amaranthus and maize. At the same time green gram and cabbage showed good germination and seedling growth in the combination of 20% of RO rejected wastewater and 80% pure water.

Furthermore, the chemical analysis of the treatments indicated that the addition of pure water to the sole RO rejected wastewater helps to reduce the harmful effect of chemical parameters of the RO rejected wastewater. Meanwhile, the estimated relationships between water quality parameters and seed germination and seedling growth variables have given an idea to find the appropriate concentration of RO wastewater for the irrigation purposes.

The findings indicated that the possibility of using RO rejected water as a part of irrigation water source. The germination and seedling stage are the most critical period in the plant growth cycle. However, seeds and seedlings of four different plant species have showed good performance in germination and seedling growth. Further, field studies needed to commercialize the usage of RO rejected wastewater as irrigation water.

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Impact of Farming Practices on Productivity of Paddy Cultivation in Minor and Major Irrigation Schemes of Mahagirilla Agrarian Services Division of Kurunegala District in Sri Lanka

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Abstract: Food supply for ever growing population has become one of the challenges in Sri Lanka. Considering the paddy production statistics over 10 years period of *Maha* and *Yala* seasons, though the districts of Anuradhapura, Polonnaruwa and Ampara maintained higher average yield than national average yield, Kurunegala district had less average yield than national average yield during this period. Therefore, this study investigated the problems at farmers' level for low productivity of paddy and identified the extent of adaptation of modern technological practices to overcome prevailing problems in Mahagirilla Agrarian Services Division. Two hundred farmers engaged in paddy cultivation from 16 Grama Niladhari divisions were selected from major and minor irrigation schemes as of 100 in each using random sampling method including. Major scheme is irrigated under Deduru Oya irrigation Scheme. Minor scheme is irrigated by *wewa*/ surface storage systems. The study revealed that the yield is positively correlated with method of planting, method of fertilizer application, duration of land preparation and irrigation method. The majority (79%) of farmers are a risk avert cluster who hesitate to adapt new technological practices to overcome prevailing problems. It could be recommended to organize awareness campaigns, build the confidence of farmers through activity oriented and result oriented trials, create a competitive work environment throughout the paddy farming areas, promote paddy cultivation among young generation and consistently providing subsidiaries through the government and non government organizations to reduce the yield gap.

Keywords: Fertilizing technology, Water management, Yield gap

Introduction

Before six decades, the population of Sri Lanka had been six million and now it has exceeded twenty million. It has become a big challenge to supply enough food for the ever growing population (Datta, 1981; FAO, 2000). This problem can be addressed by increasing the total cultivating area and introducing improved rice varieties which have qualities like higher yield, disease resistance and suitable for most existing soil problems. But, land has become a limiting factor and increasing the cultivating area could not be further proceeded with ever growing population. Also, improved varieties have reached its highest

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potential yield in research fields but not in farmer's fields so far. However, rice production has to be increased in Sri Lanka to meet the food demand in the future. Literature review proves that there is a gap between potential yield and the farmers yield mainly due to poor nutrient and water management, but other factors too contribute (FAO, 2000; Khan *et al*., 2002). Therefore, reducing yield gap between potential yield and the farmers yield need to be considered to address this problem. It is known that management of water and fertilizer is important to reduce the above yield gap or to increase yield per unit area (Bandara *et al.,* 2006).

One way of increasing the productivity of unit land area can be done by efficient use of fertilizer. Over use of fertilizer leads to environmental pollution, on the other hand increase the cost of production. Climate change prediction using general circulation model have shown that there will be water shortage in dry and intermediate zone due to temperature increase (De Silva *et al*., 2007). Therefore, understanding of constraints faced by the farmers of Mahagirilla agrarian services division in intermediate zone in relation to water and fertilizer along with other factors including new technologies in planting and seed varieties will help to introduce better management options to achieve the above task. Therefore, socio-economic survey can be used as a tool to find out the constraints faced by the farmers.

Methodology

Sample design

Target population was farmers who are engaged in paddy cultivation in Mahagirilla Agrarian service Division in Kurunegala District under major and minor irrigation systems and rainfed cultivation. This area is selected for this study to represent the Kurunegala District where the average yield is less than national average yield during the study period. There are 16 Grama Niladhari divisions which belong to two Agriculture Instructor divisions. According to Mahagirilla Agrarian Service Division, in farmers registered book, there were 3522 farmers who are engaged in paddy farming. Farmers were selected using random sampling method and 100 from major schemes and 100 from minor schemes. Data collected from the farmer's field during *maha* season of year 2016/2017. Hidogama and Hulawa are under major irrigations schemes and all other 14 GN divisions are under minor irrigation scheme.

Questionnaire Survey

Data were collected by method of Pre-tested questionnaire filled by interviewee. Questionnaire was designed to get basic information about the farmer age, gender, educational level, socio-economic constraints regarding paddy farming, water management and fertilizer problems, adaptation of new technologies with reference to water management and fertilizer (use of leaf colour chart, soil testing, use of parachute technique and use of transplanter machine)

Both primary as well as secondary data were used for analysis. Secondary data were used to supplement the findings. Key Informant Discussions (KID) were used to strengthen the quantitative findings. Secondary data were collected using the publications of Sri Lanka Census and

Statistics Department, Hector Kobbekaduwa Agrarian Research and Training Institute (HARTI) and World Bank publications. Data analysis was conducted using the Statistical Package for Social Sciences (SPSS) and Microsoft Excel 2007. Descriptive statistics along with frequency tables were used. Correlation tests were used for analysis of data. Results obtained from the quantitative analysis were supported by the qualitative data obtained through discussions, interviews and observations. When it comes to the data collection educational, psychological, economical levels of the farmers have not been considered.

Results and Discussion

Basic information about the farmers

Age distribution

Mahagirilla Agrarian service Division area belonged to rural, and dry climatic zone in Kurunegala District of Sri Lanka. It is shown that, out of the total number of respondents 45% represents were above the 61 age category, which reflects the reduction of new entries from young ages to the paddy cultivation (Figure 1).

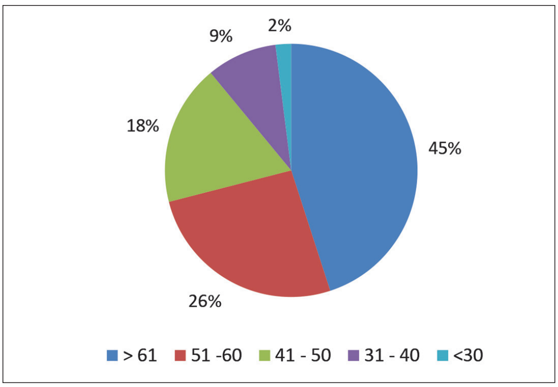

Figure 1: Age distribution

The age category of 41-50 years and 51-60 years were represented by 18% and 26%, respectively. The age category of 31-40 years and below 30 years was just 9% and 2%, respectively. It shows that the younger generation has not been involved in paddy cultivation in the study area.

Source of Income

The results show that, 93% of the correspondence Mahagirilla Agrarian service division depends on paddy farming (Figure 2). There are only 7% of farmers who are having alternative income sources such laborer in other paddy farms and or in shops.

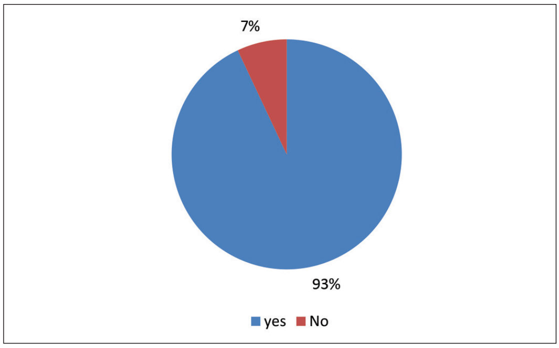

Figure 2: Source of income

Farming Practices

Planting methods

Out of the four planting methods, such as broadcasting, transplanting manually, transplanting using transplanter machine and transplanting by parachute method, only 13% of farmers have adapted transplanting by Transplanter machine and use of parachute technology in major and minor irrigation schemes which were considered as modern technological planting methods in use recently (Figure 3). It reflects that more than 75% of farmers are reluctant in practicing these planting methods although they have been made aware by the Department of Agriculture.

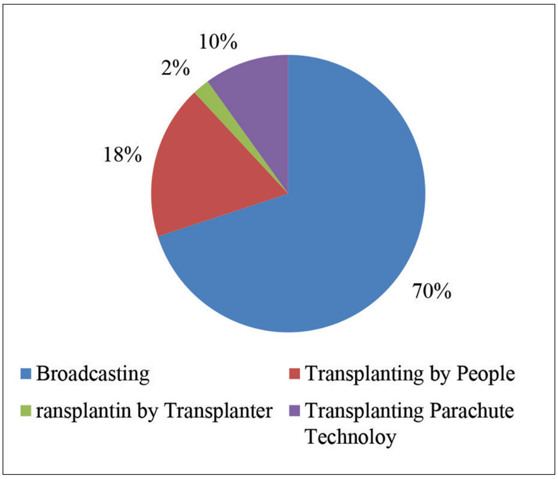

Figure 3: Planting methods used by farmers in percentage

Though broadcasting method requires more seeds than other methods, highest number of farmers tends to use broadcasting as it is the easiest method. Besides that, water requirement of rice crop varies with the method of crop establishment, and water has been identified as a scare resource in Mahagirilla Agrarian Services Division. But, these analyzed data showed that farmers did not get the benefit of practicing transplanting by machine and parachute methods which have the ability of saving water because the farmers consider the broadcasting was the easiest and low cost method compared to transplanting.

Selection of Seed variety

Although Bg300 has the potential yield of obtaining 100-150 bushels per acre, farmers tend to use this variety more than the other varieties. It is tragic that there are 6% of farmers; they do not have knowledge or not even aware of the variety, but they are using different varieties (Figure 4). Though the farmers who use major irrigation system have the ability to cultivate four or four and a half months varieties such as BG401, BG403, BG405 and BG450 which give higher yield, but farmers are reluctant to cultivate it because they think about short time benefits and the uncertainty of the availability of irrigation water.

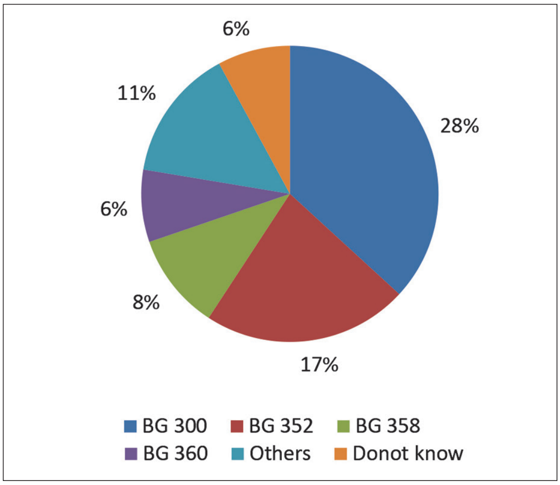

Figure 4: Selection of Seed variety

Duration of land preparation

The prevailing cultural practices shows that the majority (67%) of farmers are not following the standard land preparation times due to various reasons. This was because farmers have waited until filling the tanks/*wewa* with water completely to start cultivation and limited time was remained to allocate land preparation. But, filling of tanks entirely depend on rainfall pattern, which is also uncertain due to the prevailing climate change impacts. Only one third (33%) of farmer group has been following 21 days land preparation time, which is recommended by the Department of Agriculture.

Fertilizer Application

All the farmers in the major and minor irrigation schemes use the inorganic fertilizers recommended by the Department of Agriculture. As per the Table 1,

Table 1: Average yield per farmer (Bushel/Ac) against method of fertilizer usages

	Average yield per farmer (Bushel/Ac)		
	Major Irrigation Scheme	**Minor Irrigation Scheme**	**Rain fed**
Inorganic and organic fertilizer	108.64	98.74	58.26
Only inorganic fertilizer	87.48	74.48	44.78

comparison between the average yields of minor and major irrigation systems, both schemes shows almost the same average yield. It is obvious that the farmers who use major irrigation system get higher average yield, because there is no water shortage.

However, the reason for yield in minor irrigation scheme is because almost 80% of the farmers in minor irrigation scheme use of both organic and department recommended rates of inorganic fertilizer which gave yield almost similar to the major irrigation scheme. These results of this study agree with the findings of Premaratne and Sanggakkara (2014). As it takes about 3 months to make compost, farmers did not concern about pre preparation of compost which is enough for the next season. Though there is enough material available for compost preparation, farmers do not use it as farmers do not consider the advantage of it. Compost contributes to nutrient supply as well as moisture conservation (Premaratne and Sanggakkara, 2014) and the farmers use organic fertilizers in addition to the department recommended inorganic fertilizers have obtained higher yield.

Use of leaf colour chart

According to Khan *et al.* (2002) the use of leaf colour chart limits the excess usage of urea, by working as an indicator of applying the sufficient amount of urea. In this study, only 9.7% of respondents used the leaf colour chart as an indicator for urea application. Therefore, this is evidence that farmers show slow movement of adapting new technological practices to overcome prevailing problems. The average yield of farmers who used leaf colour chart was higher than the average yield obtained by the farmers who did not use leaf colour chart in both major and minor irrigation schemes.

Water Shortage

As shown in Figure 5, 16% of the considered sample in minor irrigation scheme faced the problem of water shortage at the flowering stage of their cultivation which was highly

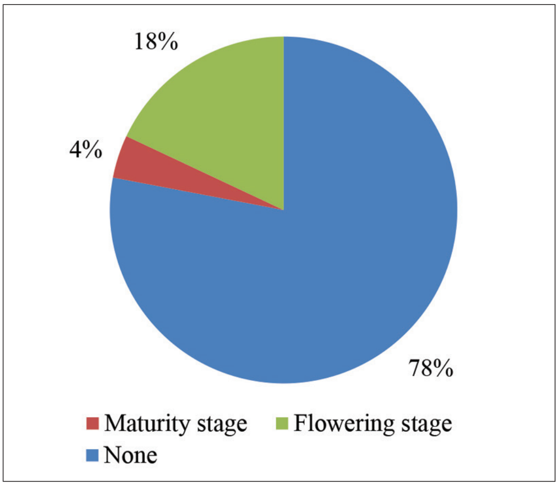

Figure 5: Farmers perception on shortage of water during different growth stages.

affected by the reduction of panicle number and potential spikelet number which in turn negatively affected the yield as suggest by Khan *et al.* (2002). About 8% of the respondents faced the water shortage problem at maturity stage which might have resulted in increase in unfilled spikelet as reported by Tuong and Bouman (2003).

Water supply method

In minor irrigation scheme when the available water was not sufficient, only 16% of the sample population overcame the water shortage problem by having alternative water storing sources like rain water harvesting ponds and agro wells (Figure 6). Unfortunately, 5% of farmers of the considered sample had to abandon their cultivation due to heavy water shortage. However, 79% of the sample population did not do any water supply method and received very low yields.

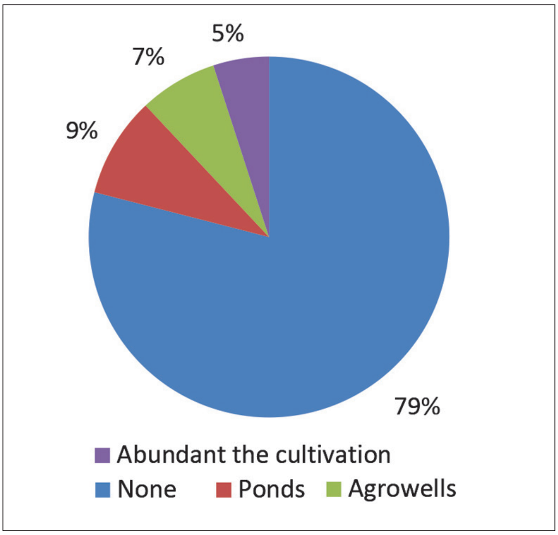

Figure 6: Water supply method when the available water is not sufficient in minor irrigation scheme.

Other problems identified in paddy cultivation

Salinity has been identified in 8% of the farmer's field in Minor irrigation scheme of Mahagirrilla Agrarian Services division, but farmers did not know how to reduce salinity from the soil. In addition they do not know about the use of liquid fertilizer to supply nutrient through leaves because it is difficult to absorb nutrients from the soil (Figure 7). Though some farmers do not know that there is salinity problem in their fields because they have not tested the soil samples through Department of agriculture prior to cultivation, they tend to use urea instead of using liquid fertilizer. Nearly, 5% of the farmers indicated the iron toxicity and 18% of the farmers indicated the drought as the problem. Almost 75% of the farmers do not know what the problems in soil or paddy cultivation are as they have never tested their soils through Department of Agriculture service.

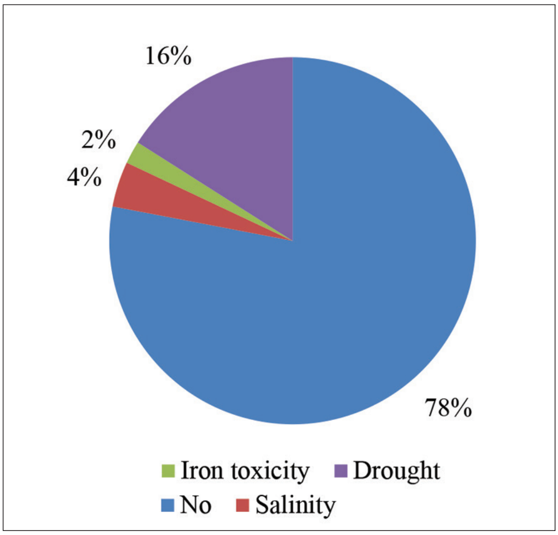

Figure 7: Other problems identified in the farmer's field

Table 2: GN Division of the study area and the average yield during the year 2016/2017.

GN Division	Average yield (bussel/Ac)	GN Division	Average yield (bussel/Ac)
Paluus *wewa*	62.66	Nagala	67.5
Galkadawala	68.23	Hulawa	93.88
Mewellewa	64.11	Sirisethagama	71.66
Mahagirilla	74.5	Dodangollegama	54.66
Hidogama	84.61	Ethewa	70
Yakadapotha	68.84	Diwllepitiya	69.95
Elagammillawa	68.24	Mahakirinda A	72.4
Nawana	69.41	Mahakirinda B	63.5

Table 3: Correlation between cultural practices and the farmer's yield

Cultural practices	**Pearson correlation**	**Sig. (2-tailed)**	**N**
Planting method	0.501**	0.00	300
Duration of land preparation	0.179**	0.002	300
Fertilizer application	0.208**	0.00	300
Water supply method when the available water is not sufficient	0.435**	0.00	300
Way of supplying water	0.737**	0.00	300

***. Correlation is significant at the 0.01 level (2-tailed).*

The average yield in minor irrigation scheme of Mahagirilla Agrarian services division in 2015/16 *Maha* season was 75.35 bushel/Ac (Paddy Statistics, 2015). In this study, only Hidogama and Hulawa GN divisions belong to major irrigation scheme therefore exceed the average yield of Mahagirilla Agrarian services division, indicating that scarcity of water had been one of the major problems of minor irrigation schemes to cause low productivity (Table 2).

Correlations were sought using Pearson`s correlation factor. Table 3 indicates significant correlations between cultural practices done and the yield. Planting method, water supply method when available water is not sufficient and way of supplying water were strongly correlated with obtained yield. Fertilizer application also influences the yield with correlation coefficient of 0.208. Further, there is a significant difference between duration of land preparation and farmer's yield, because farmers do not follow proper land preparation due to various factors such as financial constraints and labour availability (Table 3).

Conclusions and recommendations

Yield of paddy is significantly correlated with method of planting, method of fertilizer application, duration of land preparation done and the way of water supplying. As far as the Mahagirlla Agrarian division concerns, 93% of the farmers are fully depend on the paddy farming which indicate that the most influencing socio economic factor is paddy farming. Age distribution showed a declining trend of involvement into the agricultural occupations by the new generation of below 30 age category, which is only 2% of the sample farmer population. The above argument is strengthened by the fact that in the age category above 61 years represents almost half of the sample population. By representation only 13% of the farmers are adapting new technologies such as transplanting using transplanter machine and parachute planting for the paddy cultivation. It is identified that majority (79%) of farmers are a risk averters who have planted three months varieties although there is the opportunity of having more yield by planting four months varieties.

Finally, it was revealed that the inefficient cultural practices related to transplanting, fertilizing and water management in the paddy cultivation have contributed to the low productivity of Mahagirilla Agrarian Service division. Further, the prevailing cultural practices are being practiced without any developments with respect to new innovations. It has been proved that the trend of newcomers for joining the paddy farming is less which could be a negative sign to the future of the industry.

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Impact of Water Management on Nitrogen Dynamic in Low Land Paddy Soil

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Abstract: Lowland paddy cultivation requires relatively higher amount of water. The nitrogen dynamics in lowland paddy can be affected by various factors such as climatic and agronomic individually or in combination. In order to understand the effect of these factors and to reduce the complexity of the field conditions, a controlled experiment was carried out. An experiment was designed to identify the impact of alternative wetting and drying (AWD) cycles with different drying periods on the variation of NO_3^- -N and NH_4^+-N using a physical model (Lysimeter) together with simulations using Hydrus-1D. The experiment was conducted with following treatment combinations for 98 days. The data were analyzed using complete randomized design with three replicates. 4 days dry spell (D_4), 12 days dry spell (D_{12}), 20 days dry spell (D_{20}) and 4 days dry spell with plant (D_{4p}) were arranged using complete randomized design with three replicate to clarified the effect of length of dry spells and the plant on nitrogen. NH_4^+-N showed a decreasing trend over the study period in all treatments. On the other hand, NO_3^--N increased in leachate with time. The NO_3^--N loss in the leachate was higher than NH_4^+-N regardless of the length of the AWD cycles. With the presence of paddy, the nitrogen retention and leaching loss was low. Therefore, the irrigation interval would have to be decided along with the rainfall variability to reduce the nitrogen loss in paddy field. The Hydrus-1D could be used to simulate the impact of AWD on NO_3^--N loss. The measured and the simulated concentration of NO_3^--N correlated with R^2 values of 0.89, 0.79, 0.74 and 0.69 for D_4, D_{12}, D_{20} and D_{4p}, respectively. The NO_3^--N loss in the leachate was higher than NH_4^+-N regardless of the length of the AWD cycles. The length of the dry spells up to 12 days did not show significant variation in nitrogen loss in this study compared to 20 days dry spell. Therefore, the irrigation interval has to be decided along with the rainfall variability to reduce the nitrogen loss in paddy field.

Keywords: Alternative Wetting and Drying, Hydrus -1D, Nitrogen loss, Paddy

Introduction

Lowland paddy cultivation requires relatively higher amount of water. The ponded water plays a central role in weed control, soil biomass accumulation and decomposition, nutrient availability, crop growth and the yield. However, under minor irrigation systems, maintaining a continuous flooded condition is difficult due to high rainfall variability and lack of enough water in the tanks throughout the growing season. Alternate wetting

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and drying (AWD) is therefore a common phenomenon in paddy fields.

The extension of a dry spell in an AWD cycle will cause irregular and extreme water stresses for soil organism and plants. This affects nutrient availability, plant productivity, biogeochemical processes, gaseous and leaching losses and carbon and nitrogen pools in soils (Borken and Matzner, 2008). The drying and rewetting impose a significant stress on the microbial community of soil. While wetting events are common in most environments, the short and long-term effects of soil drying on microbial processes have not been well understood. Therefore, it is important to understand the consequences of varying dry spells on the soil microbial community and other soils processes.

Nitrogen plays an important role in crops grown under supplementary irrigation (Tavakkoli and Oweis, 2004). Nitrogen accumulation and leaching in soil profiles with different irrigation schedules and rate of fertilizer application have been reported (Wei *et al*., 2010; Sepaskhah, 2012). Nitrogen losses in paddy fields under AWD cycle can be extremely large compared to continuous flooding conditions because of the difference in nitrogen transformations between anaerobic and aerobic environments (Tan *et al.,* 2015).

Nitrate-N (NO_3^--N) formed by nitrification of ammonium nitrogen (NH_4^+-N) during drying regimes can be quickly lost by denitrification in the following wetting regimes (Buresh *et al*., 2008; Dandeniya and Thies, 2010). The formation of aerobic and anaerobic condition depends on the number dry days in the AWD cycles. Short period may not create effective aerobic conditions. This also may depend on the infiltration and drainage characteristics of the soil.

Therefore, owing to these different soil processes, environmental conditions and management, the amount of nitrogen can vary with plant type, growth stage, fertilizer, time and the soil depth. Objective of this study was to identify and assess the impacts of AWD cycles with different drying period on the variation of NO_3^--N and NH_4^+-N. For achieving this objective under similar conditions to lowland paddy, a physical model (Lysimeter) and Hydrus-1D model was used.

Materials and Methods

Preparation of Lysimeter and treatment combinations

Rectangular plastic containers were used to prepare Lysimeters for the experiments. Twelve containers each having 54 cm length, 36 cm width and 30 cm depth with the surface area of 0.194 m^2 were used to simulate the field condition. A drainage system with perforated PVC pipe was placed at the bottom of the Lysimeter. The pipes were connected to a common pipe for drain water to a common outlet. The metal aggregates with the diameter of 1 - 2 cm were placed up to 3 cm from the bottom to cover drainage pipes to facilitate free water flow. Then a plastic mesh was placed on top of the aggregates to prevent soil movement. Soil collected from the command area of Bayawa minor irrigation system was used to fill the Lysimeters. Sieved soil (< 2 mm) was filled up to 15 cm depth leaving 12 cm from top of the container. The effective depth of the Lysimeters was 20 cm.

A piezometer was installed vertically in each Lysimeter in order to monitor the water level. The Lysimeters were allowed to settle down with adding water from the bottom and allowed for pre-settling for 10 days. The outlet of the Lysimeter was connected to a plastic collector with the capacity of 5 liters.

The experiment was conducted with following treatment combinations for 98 days based on the growth period of paddy. The experiment was arranged with complete randomized design with three replicates. The treatment was designed based on the probability analysis of last 80 years of rainfall data. The 6 mm of rainfall was simulated during the experimental period. This amount of rainfall has high probability of occurring in the study area.

D_4 - 4 days interval of simulated rainfall to represent 4 days dry spell

D_{12} - 12 days interval of simulated rainfall to represent 12 days dry spell

D_{20} - 20 days interval of simulated rainfall to represent 20 days dry spell

D_{4p} - 4 days interval of simulated rainfall to represent 4 days dry spell with paddy

Agronomic practices

A 10 cm depth of irrigation was provided at 0, 15, 30, 75 days and urea fertilizer was applied at 0, 16, 31, 46 days at the rate of 125 kg/ha to simulate the field conditions.

Soil and water sampling

Soil samples were collected from the surface at the beginning (0 days), air dried, sieved (2 mm) and analyzed for texture and NH_4^+-N and NO_3^--N were measured. Soil core sample was used to measure the saturated hydraulic conductivity and bulk density. This procedure was continued at 14 days interval until end of the experiment.

Leachate volumes were measured at weekly intervals and subsamples were used to measure NO_3--N (Keeney and Nelson, 1982) and NH_4^+-N (Searle, 1984) in the laboratory. Data were analyzed statistically at 95% confidence level using SAS statistical software.

HYDRUS -1D simulation

The HYDRUS – 1D model was used to simulate a 15 cm soil profile as a single soil layer with observation modes at 5 cm and 10 cm depths. The model printing times were 0, 20, 40, 60, and 80 days represented as (T0), (T1), (T2), (T3) and (T4), respectively. Boundary conditions and initial conditions are shown in Table 1.

Figure 1: The experimental set up with and without plant

Table 1: Initial and boundary conditions used in Hydrus-1D simulation

	Boundary Condition		Initial Condition	
	Upper	Lower	Upper	Lower
Water flow	Atmospheric boundary with surface layer (10 cm)	Free drainage	Pressure head (-15)	Pressure head (-15)
Solute	Concentration flux boundary	Zero gradient	Liquid phase concentration NH_4^+ = 6.59 µg, NO_3^- = 0.93 µg	Liquid phase concentration NH_4^+ = 6.59 µg, NO_3^- = 0.93 µg

Table 2: Input parameters used in solute transport using Hydrus-1D

Solute transport and root up-take parameters	NH_4^+–N	NO_3^-–N
Adsorption isotherm coefficient (K_d)	3.5	0
First-order rate constant for dissolved phase (Sinkwater*)	1	0
Molecular diffusion coefficient in free water, (Diffus.W)	1	1
Longitudinal dispersivity, (Disp) (1/10 of profile length)	1.5	1.5
Maximum allowed concentration for passive up take (cRoot)	30	70
N-fertilizer Applications (µg-N/cm^2)	$cTop_1$	$cTop_2$
0 day – Precipitation assume 1 cm (real 0 cm)	172.5	402.5
16th day – Precipitation 10 cm	172.5	402.5
31st day – Precipitation 10 cm	172.5	402.5
46th day – 17/10/2014 Precipitation assume 1cm (real 0 cm)	172.5	402.5

Results and Discussions

Climatic conditions and management practices

The treatment combinations were subjected to several cycles of varying length of dry days in AWD cycles. Figures 2, 3 and 4 show management practices and climatic conditions for D_4, D_{12} and D_{20}, respectively.

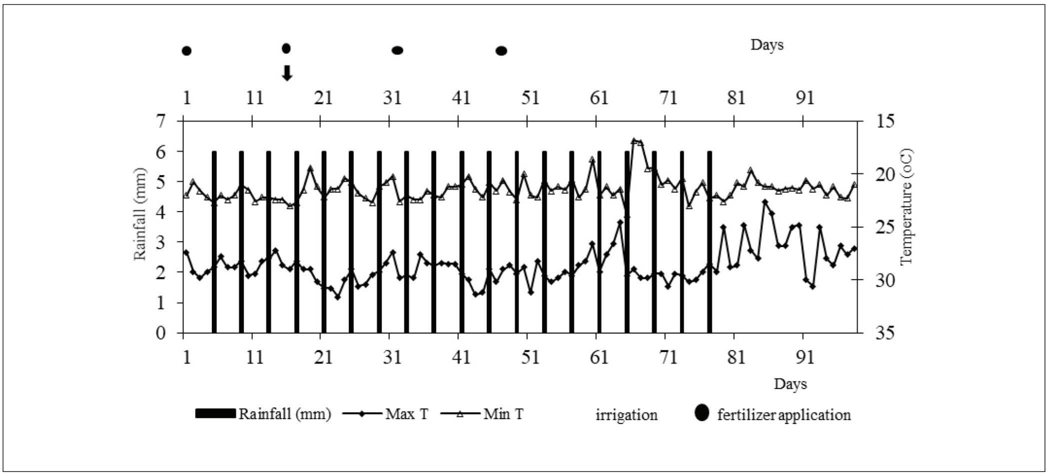

Figure 1: The climatic conditions and management practices for D_4

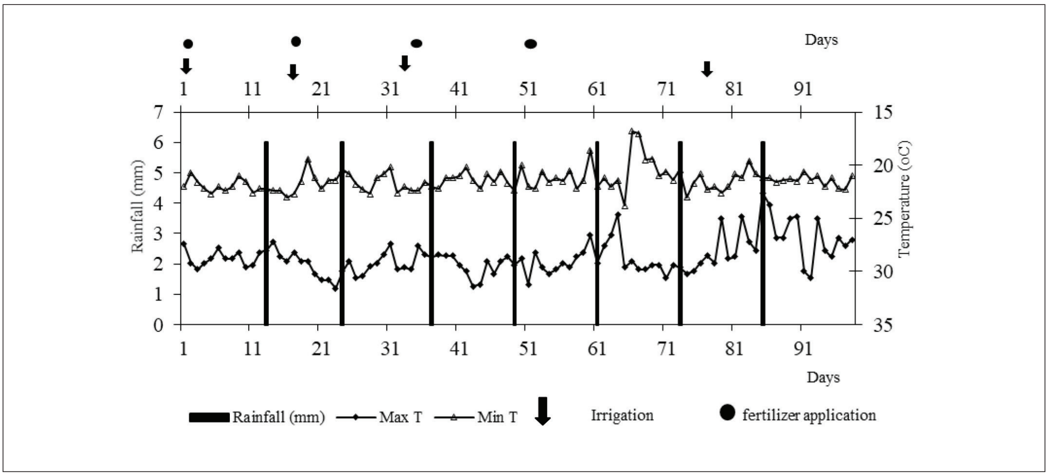

Figure 2: The climatic conditions and management practices for D_{12}

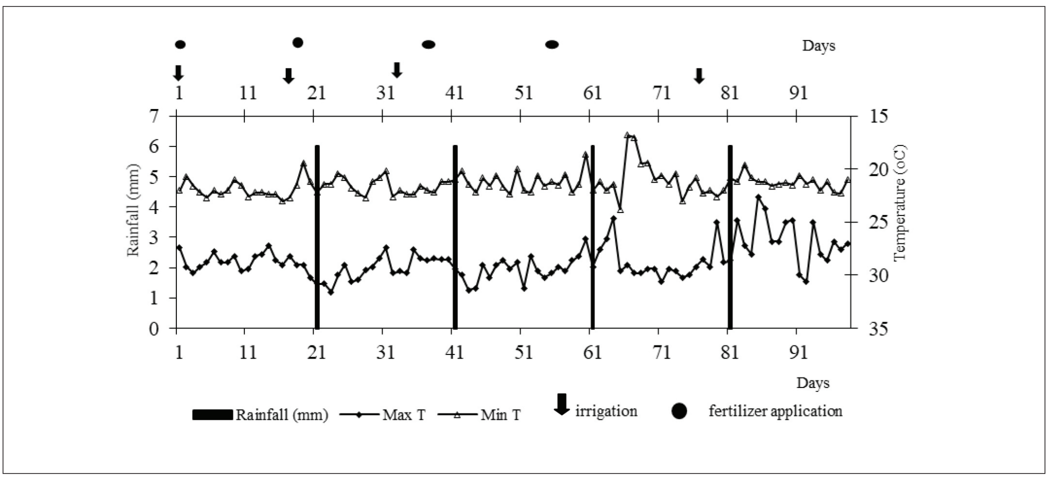

Figure 3: The climatic conditions and management practices for D_{20}

Variation of NO_3^--N and NH_4^+-N concentrations

Table 3 shows the average NO_3^--N and NH_4^+-N concentration in 12 leachate samples in all 4 treatments. ANOVA shows no significant difference between D_4 and D_{12}, (shorter dry spells), but D_{20} (longer dry spell) was significantly different. These results indicate that the shorter and frequent wetting and drying cycle increase the nitrogen loss (both NO_3^--N and NH_4^+-N) through leaching when compared to longer days dry cycles (20 days in this case as tested). Based on these results, we cannot say exact number of dry days needed to have a significant nitrogen leaching since we do not have data in between 12 and 20 days.

Table 3: Nitrate nitrogen (NO_3^--N) and ammonium nitrogen (NH_4^+-N) concentrations (mg/L) in the leachate

Treatment	NO_3^-–N	NH_4^+–N
D_4	2.07+ 1.31[a]	0.68±0.47[a]
D_{12}	2.90+1.66[a]	0.64±0.46[a]
D_{20}	0.92+1.24[b]	0. 46+0.33[b]
D_{4p}	1.28+1.21[b]	0.43+0.43[b]

The means with the same letters are not significantly differ from each other at α=0.05, mean comparison is at column wise

The effect of irrigation on nitrogen leaching was observed since higher amounts of NO_3^--N and NH_4^+-N were measured after the irrigation. Except the samples collected following irrigation, NO_3^--N loss is high in D_4 followed by D_{12}. The NH_4^+-N loss in leachate is high in D_4 followed by D_{12}, D_{20} and D_{4p}. These results agree with the study by Wijler and Delwiche (1954). Higher nitrogen loss has been observed in soils subjected to periods of alternate drained (aerobic) and flooded (anerobic) conditions (Sepaskhah and Trfteh, 2012)

In the presence of paddy, the loss of nitrogen was less compared to the treatment without plant in the 4 days cycle. The plant should have utilized the nitrogen and the loss was minimized as expected. Therefore, the effect of plant factor on nitrogen loss is cleared in this comparison.

The cumulative leaching of NO_3^--N and NH_4^+-N are shown in Figure 5 and Figure 6, respectively. The total loss of nitrogen was high in D_{12} followed by D_4, D_{20} and D_{4p}. The total loss of nitrogen was calculated based on concentrations and the leachate volumes. The amount of leachate also plays a role in nitrogen losses in AWD cycles. The amount of water applied per irrigation and frequency in AWD cycles is a management option to minimize the effect of nutrient loss. When AWD has a higher frequency, such as D_4 in this case, soil is in saturated conditions. In the saturated condition, nitrification is less and amount of NO_3^--N available for leaching is less. On the other hand, nitrification adversely is affected by extreme drought condition as in D_{20}.

Figure 6 shows the cumulative NH_4^+-N loss in all treatments. The cumulative loss of NH_4^+-N is varied as $D_4 > D_{12} > D_{20} > D_{4p}$, where a significantly low NH_4^+-N loss was in D_{4p} compared to D_4. Table 4 summarizes the inorganic – N recovery efficiency for different drying cycles. The recovery efficiency is varied as $D_4 > D_{4p} > D_{12} > D_{20}$.

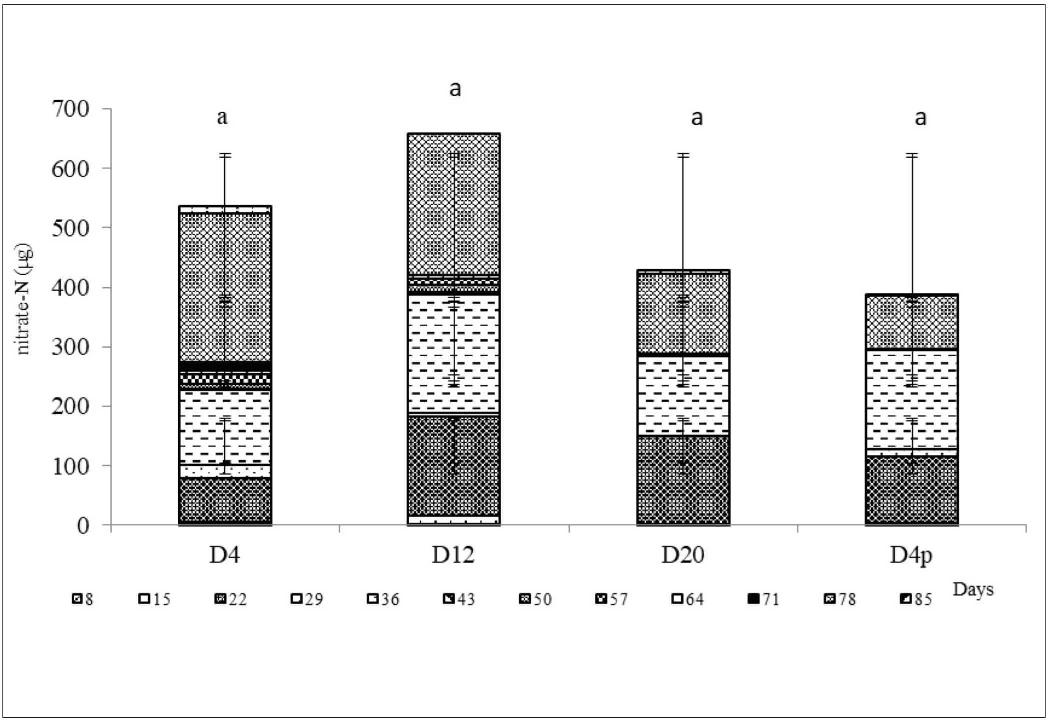

Figure 5: Cumulative nitrate – N loss through leachate for treatments

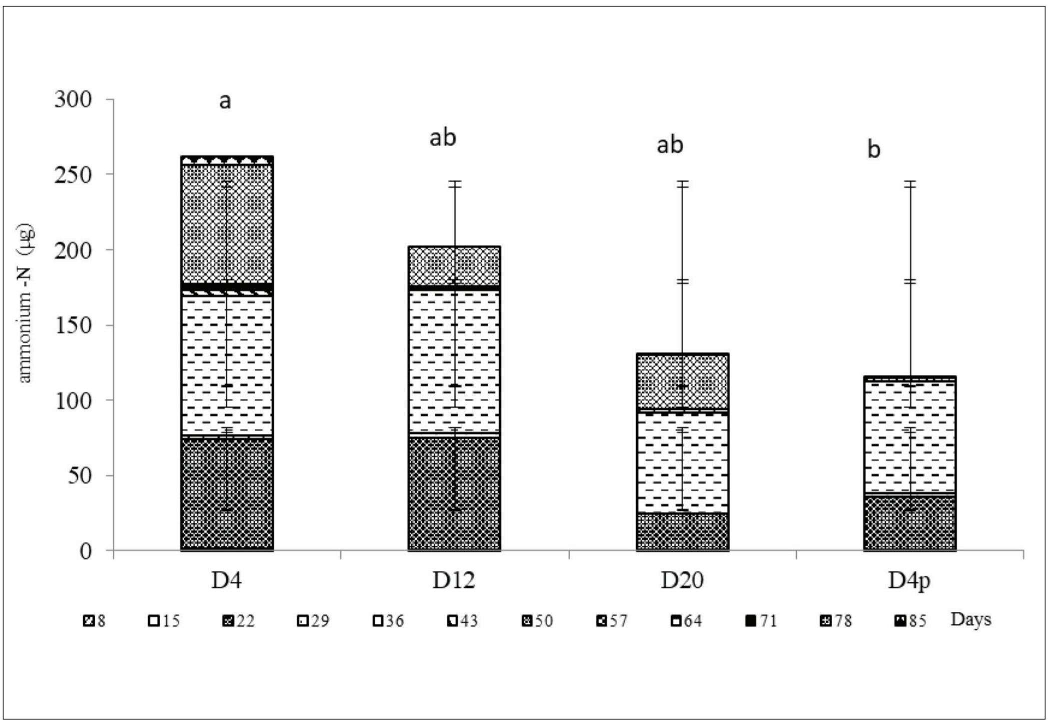

Figure 6: Cumulative ammonium – N loss through leachate for treatments

Table 4: The recovery of inorganic – N with treatment

AWD intervals	Paddy plant	N-added (mg/g of soil)	Initial total mineral N (ppm)	Final total mineral N (ppm)	Nitrogen retained (%)
4 days (D_4)	No	26.07	26.04	20.24	38.85
12 days (D_{12})	No	26.07	70.40	21.31	22.09
20 days (D_{20})	No	26.07	90.11	22.02	18.95
4 days (D_{4p})	Yes	26.07	30.07	20.70	36.87

Simulated water flow

Water flow was simulated with Hydrus-1D. A remarkable increase in the observation node's water content for all treatment is observed during the irrigation day and decreases with time. In all treatments, the water content at 10 cm observation node was higher than that at 5 cm node.

Also the 5 cm node shows a quick loss of water and quick recovery in simulation of water. All treatments shows similar pattern of the fluctuations with their respective water application. The D_{4p} (4 days dry spell with plant) shows a reduced amount of water at the 5 cm observation node at 40 to 98 days. This may be associated with the root water uptake by the plant with the growth stages.

Simulated water flux

The water flux value remains positive, means there are no water stress in the experimental setups during the study period. The effective root zone depth was 12 cm, because the model simulates the water flux above 12 cm depth in negative direction.

Model validation

The model was validated by assumption of fate of Urea with different combinations of NH_4^+-N and NO_3^--N. This fraction in the soil depends on soil pH, moisture level, aeration and microbial activity. The fertilizer applied undergoes through the hydrolysis process resulting NH_4^+, NO_2^-, NO_3^- and N_2. The NH_4^+ and NO_3^- can be taken up by plants. The Hydrus-1D simulated the mean concentration of nitrogen in the leachate and solute balance error (%) for assumptions on fate of Urea. With increase in the fraction concentration, NH_4^+-N and NO_3^--N show an increasing trend.

Error balance increases for NO_3^--N and decrease for NH_4^+-N. Most suitable fraction for NO_3^--N leaching was 30n_70N (urea hydrolyzed into 30% of ammonium and 70% of nitrate) for all treatments. The Hydrus-1D simulated leaching loss of NO_3^--N was 285, 1150, 1140 and 1090 kg/ha for D_4, D_{12}, D_{20} and D_{4p}, respectively. Except for the D_4, other treatments show a similar pattern of changes for both simulated and measured values. The correlation between the measured and simulated values (R^2) were; 0.89, 0.79, 0.74 and 0.69 for D_4, D_{12}, D_{20} and D_{4p}, respectively.

The measured fertilizer used efficiency was calculated using fertilizer input and leachate output, for all treatments. The simulated fertilizer use efficiency was also calculated indirectly from fertilizer input and bottom solute flux (represent leachate). The measured fertilizer use efficiency was 65.3, 27.9, 38.3 and 53.8% for D_4, D_{12}, D_{20} and D_{4p}, respectively. These values varied as 74.0, 82.9, 83.3 and 77.1% under simulated conditions for D_4, D_{12}, D_{20} and D_{4p}, respectively.

According to Patrick and Reddy (1974), the NO_3^--N leaching in AWD was higher than continuous aerobic conditions, but it was higher under continuous aerobic condition than anaerobic condition. The soil used in this experiment was subjected to a 17 mm/day infiltration rate. Therefore, the applied irrigation water could be escaped from the soil column in 9 days.

Simulated solute flux

The solute flux (NO_3^--N and NH_4^+-N) at the surface fluctuates with the split application of the fertilizer. Even though all treatments provided with the same amount of fertilizer, the fluctuation was high during the 2nd and the 3rd application where, the fertilizer was applied with irrigation water. Figure 7 and Figure 8 describe the variation of NH_4^+-N and NO_3^--N at surface with time, respectively. In all treatments, the surface concentration of NH_4^+-N and NO_3^--N shows the similar pattern. The surface NH_4^+-N concentration shows increasing and then decreasing pattern with maintaining $D_{20} > D_{12} > D_4 > D_{4p}$ rates, mainly because of plant uptake of NH_4^+-N.

The concentration of NO_3^--N shows an increasing pattern with time and shows

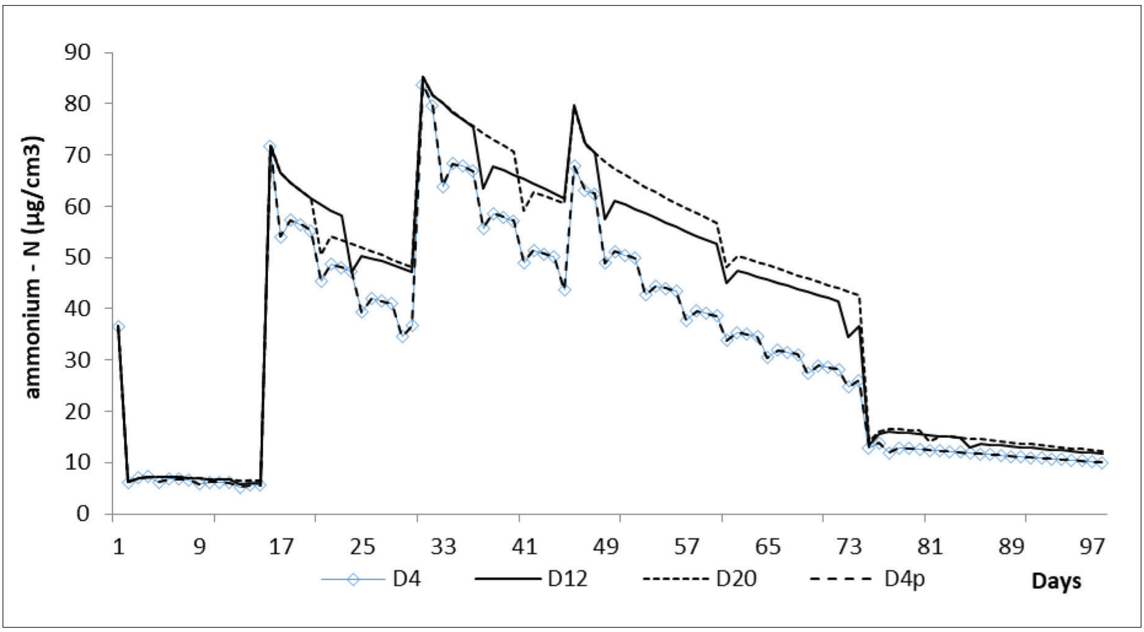

Figure 7: Simulated surface concentration of ammonium – N (NH_4^+-N)

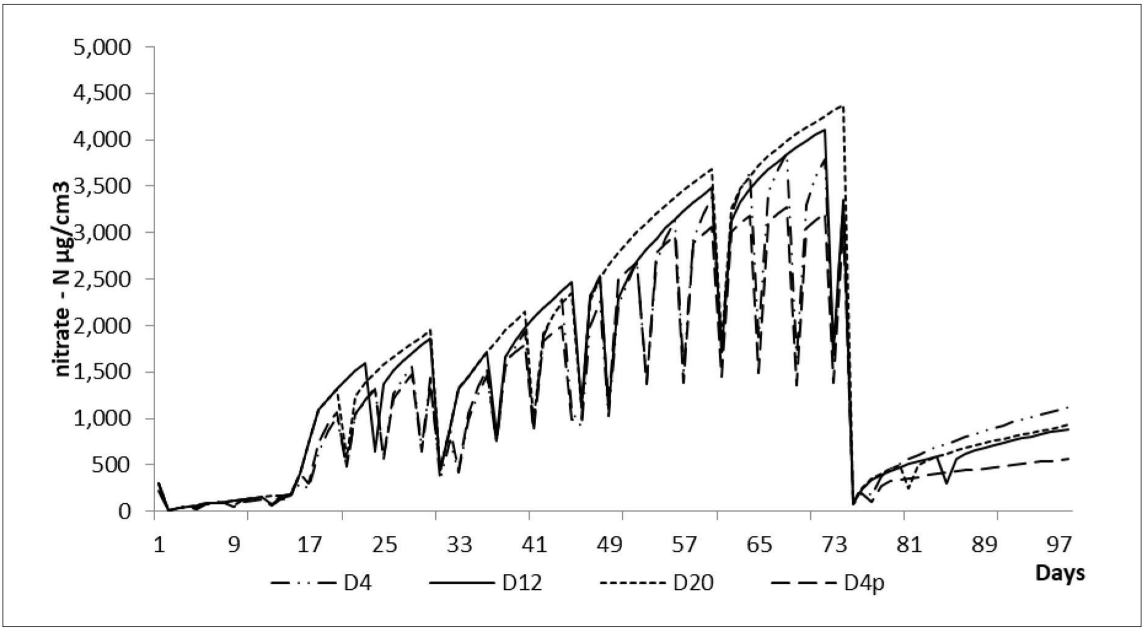

Figure 8: Simulated surface concentration of nitrate – N (NO3-N)

the decreasing in flux for the period of irrigation. The surface concentration is very high than the NH_4^+-N. During the period of experiment for 90 days, the recorded minimum temperature was greater than 20^{o}C. This situation favors the conversion of urea in to NO_3^--N than to NH_4^+-N. With the application of irrigation water, surface concentration reduces considerably. With availability of excess water, the leaching potential of negatively charged NO_3^--N is high. The surface concentration of NO_3^--N varied as $D_{20} > D_{12} > D_4 > D_{4p}$.

In all treatments, the concentration of both nitrogen ions shows similar patterns. The reduction of NH_4^+-N and increases in NO_3^--N concentration in the later part might be due to two factors; scarcity of the water to absorb by plants or leached out and change in the growth stage of the plant. During the later part due to the reduced water, soil leads to more aerobic conditions and formation of NO_3^--N from urea.

Conclusions

The Lysimeter study further clarified the effect of the length of dry spells and the plant on nitrogen dynamics. The aerobic condition towards the end of growing period creates the decreasing trend of NH_4^+-N and increasing trend of NO_3^--N. The irrigation interval would have to be decided between 12 to 20 days along with the rainfall variability to reduce the nitrogen loss in paddy fields. The Hydrus-1D could be usedto simulate the impact of alternative wetting and drying on NO_3^--N loss and for understanding of nitrogen dynamics under different agronomic conditions.

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Page 3 onwards

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…. with low or zero trans fatty acids and the results were promising (Zhang et al., 2001; Zhang et al., 2006; Goli et al., 2008).

……trans-free margarine formulations and most widely used enzyme for the interesterification is Lipozyme TL IM (Ferreira-Dias, 2013).

…..fatty acids can be obtained by enzymatic interesterification (Huang and Akoh, 1994). This result was later contradicted by Becker and Seligman (1996).

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Journals articles

1. Single author
   Gil, A. 2002. Polyunsaturated fatty acids and inflammatory diseases. Biomedicine and Pharmacotherepy, 56:388–396.

2. Two authors
   Huang, K.H. and Akoh, C.C. 1994. Lipase-catalyzed incorporation of n-3 polyunsaturated fatty acids into vegetable oils. Journal of the American Oil Chemists' Society, 71:1277–1280.

3. More than two authors
   Zhang, H., Xu, X., Mu, H., Nilsson, J., Adler-Nissen, J. and Høy, C.E. 2000. Lipozyme IM- catalyzed interesterification for the production of margarine fats in a 1 kg scale stirred tank reactor. European Journal of Lipid Science and Technology, 102:411–418.

Books and other monographs

4. Chapter in book
   Gordon, M.H. 2001. The development of oxidative rancidity. In: Pokorny, J., Yanish, N. and Gordon, M. (eds) Antioxidants in Food – Practical Applications. CRC Press, Washington, pp 7-225.

Web references

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