MOLECULAR ABSORPTION OF UREA BY FLOODED RICE

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By

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THESIS

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requirement for the degree of

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Department of Agronomy
COLLEGE OF HORTICULTURE
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Kerala

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DECLARATION

I hereby declare that this thesis entitled **"Molecular absorption of urea by flooded rice"** is a bonafide record of research work done by me during the course of research and that the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship of any other similar title, of any other University or Society.

Vellanikkara,

22-7-1992

SAFEENA, A.N.

CERTIFICATE

Certified that the thesis entitled "Molecular absorption of urea by flooded rice" is a record of research work done by **Miss. Safeena, A.N.** under my guidance and supervision and that it has not previously formed the basis for the award of any degree, fellowship or associateship to her

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"Oars alone can never prevail-
To reach the distant coast;
The breath of Heaven must swell the sea
Or all the toil is lost"

Cowper

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SAFEENA, A N.

Dedicated to
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Introduction

INTRODUCTION

It has been well established that the plants absorb nitrogen mainly in the form of ammonia and/or nitrate. In the case of flooded rice, ammoniacal form is preferred to nitrate form, primarily because nitrification is rather slow in anaerobic soil environment Urea is the most popular nitrogen fertilizer used in rice cultivation. When urea is applied to soil, it rapidly undergoes enzymatic hydrolysis leading to the production of ammonia and CO_2. This reaction mediated by the enzyme urease present in the soil, forms the most important step in the utilization of N from urea by the plant Though urea is mostly taken up by plant after its hydrolysis, it has been demonstrated by Japanese workers in the early 1960's (Mitsui and Kurihara, 1962) that rice plants are capable of absorbing urea in the molecular form. In a recent study conducted in this lab, it was observed that a drastic decline in urease activity occured following flooding the soil (Saraswathi et al., 1991) It is to be assumed in this context that urea top-dressed to flooded rice can persist in the soil for a considerably longer time without hydrolysis Under such circumstances, there exists a strong possibility for absorbing larger quantities of urea in the molecular form. Hence it was decided to examine the possibility of molecular absorption of urea by flooded and non-flooded rice from top and basal dressings

There are divergent findings on the effect of soil moisture or water level on urease activity While some workers have reported decrease in soil urease activity upon soil submergence (Dalal, 1975), some wokers could not find any such effect of soil moisture (Skujins and Mc Larin, 1969; Delaune and Patrick, 1970; Gould <u>et</u> <u>al</u>., 1973). Most of these studies have done by the non-buffer method of Zantua and Bremner for 5 h incubation period However, not much information is available on the effect of soil submergence upon prolonged incubation with urea. Though the urease activity is mostly estimated employing the non-buffer method, it is possible to estimate the activity using ^{14}C labelled urea. Considering all these aspects an investigation was undertaken with the following objectives.

1 To study the effect of soil submergence on urease activity upon prolonged incubation with the substrate
2 To know the molecular absorption of urea by flooded and non-flooded rice from top and basal dressings.
3 To develop an isotope method for the estimation of urease activity

Review of Literature

REVIEW OF LITERATURE

The literature reviewed in this chapter are classified under the following heads.

1 Origin and location of soil urease
2 Kinetics of urease activity
3 Influence of soil moisture on urea hydrolysis
4 Methods of assay of urease activity in soils
5 Absorption of molecular urea and other forms of nitrogen

2.1 Origin and location of soil urease

The presence of urease in soil was first indicated by Rotini (1935) Later Conard (1940, 1942a, b) hypothesized that urea hydrolysis was catalyzed by extracellular urease derived from dead and ruptured cells of ureolytic microorganisms and plant organs adsorbed into soil functions. Later Mc Garity and Myers (1967) reported that urea could be hydrolysed by urease produced by active soil microorganisms. Paulson and Kurtz (1969) demonstrated that urease activity of soil could be devided into two components. microbial urease, directly associated with microorganisms and adsorbed urease, apparently adsorbed on soil colloids But it was generally assumed that the urease in soils is essentially a microbial extracellular enzyme accumulated through release of urease from living and disintegrated microbial cells (Skujins, 1976). Therefore urease activity refers to the activity of extracellular urease in the soil and exclude the urease activity of metabolizing micro-organisms

Although some of the urease activity produced on treatment of soil with organic material persisted for several weeks, the urease activity of soil amended with organic materials eventually became identical to that of the unamended soil (Zantua and Bremner, 1976, 1977) This indicates that the urease activity of unamended soils reflects their capacity for protection of urease and that urease in excess of their capacity is decomposed or inactivated. Zantua and Bremner (1976, 1977) concluded that native urease in soils was remarkably stable and different soils had different levels of urease activity determined by the ability of their constituents to protect urease against microbial degradation and other processes leading to inactivation of the enzyme.

According to Burns (1982) the location of urease enzyme was atleast partially determined by such factors as the size and solubility of its substrate, the species of microorganisms and the physical and chemical nature of the soil colloids. He also found that enzyme bound to clay and humic colloids had a residual activity not found in enzymes free in the soil-aqueous phase. The results of the studies by Tiwari <u>et al</u>. (1988) demonstrated that there were atleast three different loci of enzyme activity in soil: inside viable cells, on the surface of clay humic colloids and in the soil solution.

2.2 Kinetics of urease activity

According to Delaune and Patrick (1970) urea hydrolysis took place at the initial rate of about 8-12 ppm/h and levelled off after about 24 h. Gould et al. (1973) measured the hydrolysis rate of urea under laboratory conditions and found that equivalent of 200 μg urea N g^{-1} soil added as a solution, hydrolysed in 20 h.

Dalal (1975) studied the effect of varying the period of incubation on the urease activity and found that urea hydrolysis followed a zero order kinetics. The substrate concentration was not a limiting factor in the assay of enzyme activity for periods of incubation extending upto 16 h. Sankhayan and Shukla (1976) found that most of the urea added to five Indian soils was hydrolysed within 24 h and the average half time values ranged from 3.7 to 7.9 h. Hydrolysis was found to follow a first order reaction. Sahrawat (1980a) observed in a non-buffer method of study with three alluvial soils of India that urea hydrolysis followed a zero order kinetics at least up to 12 h and the urea hydrolysis rate coefficient (ko) of the soil ranged from 0.83 to 0.167 μ moles g^{-1} soil h^{-1}.

Singh and Yadav (1981) observed that complete hydrolysis of urea occured after one week in sandy loam and two weeks in sandy soils. Singh and Bajwa (1986) found that urea hydrolysis seemed to follow first order kinetics in salt affected soils and the average time for one half of the hydrolysis to occur (t 1/2) ranged from 0.51 to 4.55 days.

Paulson and Kurtz (1970) obtained Michaelis constant (Km) value of 0.212 M for the total urease activity and 0.057 M and 0.252 M for microbial and adsorbed forms of soil urease respectively. In presence of THAM buffer, Tabatabai (1973) reported Km values ranging from 1.3-7 M urea for different soils. Pal and Chhonkar (1979) evaluated the influence of different soil characteristics on enzyme kinetics and found that Michaelis constant (Km) for the enzyme varied in different soils and was significantly and positively correlated with soluble salt content. Saraswathi (1989) in a study without buffer recorded the Km values of 116, 130.2 and 714.3 μ moles of urea in Karappadam, black and kari soils. The corresponding V max values obtained were 1.[illegible] 2.6 and 7.3 u moles of urea hydrolysed g^{-1} soil h^{-1} respectively. With buffer (pH 7.2) the Km values obtained for Karappadam, black and kari soils were 45.3, 55.1 and 2346.6 μ moles of urea and the corresponding V max values were 4.2, 1.2 and 16.7 μ moles of urea hydrolysed g^{-1} soil h^{-1} respectively.

2.3 Influence of soil moisture on urea hydrolysis

There are divergent findings on the effect of soil moisture or water level on urease activity. Several workers had found that urease activity in soil was not significantly affected by the water level (Skujins and Mc Laren 1969 Delaune and Patrick, 1970; Gould et al , 1973) Skujins and Mc Laren (1969) found that activity in an air dried soil equilibrated at 100 per cent relative humidity was close to that observed when this soil contained 50 per cent water Dalal (1975) found that urease activity increased with moisture content from 25 to 50 per cent WHC beyond which it decreased. According to Wikremansinghe et al. (1981) the rate of urea hydrolysis was independent of soil moisture at 25% or above.

More and Varade (1982) concluded that about 65% of applied urea N hydrolysed within 2-4 days at all levels of soil moisture potentials (zero -0 33, -5.0 and -25 0 bars, Farooqui et al. (1983) observed complete hydrolysis of urea within 11 days and found that mineralisation potential of soil under anaerobic condition was more than that under aerobic condition. According to Vlek and Carter (1983) soil urease is sensitive to the lack of soil moisture and an increased hydrolysis of urea occurs with increasing soil water content upto near field capacity, followed by a decrease thereafter Kuma and Wagenet (1984) also reported a linear increase in urease activity with moisture contents upto field capacity. Sahrawat (1984) observed that urease activity increased with increasing moisture content from air dried state to field capacity and then remained constant with further increase in moisture content. However, urease activity was not detected in soil samples collected in late summer when the soil moisture content was below -15 bar

According to Sahrawat (1980b) flood water of tropical lowland rice soils might have measurable amounts of urease activity though its contribution was far less than that of soil urease. Vlek *et al.* (1980) reported that urease activity in flooded soils was not constant but was dynamic and changed with the duration of flooding. According to them the flood water urea was hydrolysed largely at the soil-flood water interface. Savant *et al* (1985) observed that in a submerged soil system incubated for 24 h, the depletion of O_2 seemed to retard the hydrolysis and with a longer submergence time, soil Eh decreased and soil urease activity also decreased to a stabilized value. However, on reoxidation of the reduced soil under a continuous 1 cm of flood water the soil urease activity showed a marked increase. In general the order of the urea hydrolysis in the main three components of the wetland soil system was oxidised soil > reduced soil > flooded water (without algae) Thus urea hydrolysis in a wet land soil system showed temporal and spatial variation

According to Monoreal *et al.* (1986) urea got hydrolised completely within five days. Flood water concentrations of urea

N decreased rapidly and all urea initially present in the flood water was hydrolysed within 3-4 days. Saraswathi et al. (1991) observed that when urea was added to the soil which was in a reduced condition, there was a steady decrease in urea hydrolysis and no urease activity could be noticed upon flooding for periods longer than two days.

2.4 Methods of assay of urease activity in soils

Numerous methods had been used for assay of urease activity in soils The methods which had been thoroughly evaluated were the buffer method proposed by Tabatabai and Bremner (1972) and the non-buffer method proposed by Zantua and Bremner (1975) The non-buffer method cited was essentially a scaled-down version of the method proposed by Douglas and Bremner (1970), the only significant difference being that, toluene was omitted. It involved determination of the amount of urea hydrolysed or incubation of the soil sample with urea at 37°C for 5 h, urea hydrolysis being estimated by colorimetric determination of urea in the extract (Douglas and Bremner, 1970) obtained by shaking the incubated soil sample with 2 M KCl containing a urease inhibiter (PMA) and filtering the resulting suspension According to Zantua and Bremner (1975), although both methods gave precise results, the buffer method gave markedly higher values than non-buffer method and detected urease activity that did not occur when soils were treated with urea in the absence of buffer. A rapid assay for soil urease had been developed by Kandeler and Gerber (1988). The method involved incubation of soil with an aqueous or buffered urea solution, extraction of ammonium with 1 N KCl and 0.01 N HCl and colorimetric NH_4^+ determination by a modified indophenol reaction. In a modified method of Douglas and Bremner, Praveenkumar and Aggarwal (1989) proposed the use of H_2SO_4 in place of H_3PO_4. The use of H_2SO_4 was found to increase the linear calibration range almost two fold without affecting the precision of the original method and the developed colour remained stable for 72 h in the dark.

Several workers had assayed soil urease activity in soils by estimating the $^{14}CO_2$ released through hydrolysis of ^{14}C-labelled urea (Skujins and Mc Laren, 1969; Pel'tser, 1972; Norstadt et al., 1973) Skujins and Mc Laren (1969) observed a linear rate of $^{14}CO_2$ evolution in all urea-amended soils for several hours, after which time an increase of rate indicating microbial proliferation. Thus an intrinsic urease activity might be distinguished from enzymatic activity due to microbial proliferation. One problem recognised by Skujins and Mc Laren (1968, 1969) was that the $^{14}CO_2$ produced by hydrolysis of ^{14}C labelled urea in soils was not evolved quantitatively They proposed use of an acidic (pH 5.0) buffer for assay of urease activity in soils by determination of the $^{14}CO_2$ released from ^{14}C labelled urea. Another problem in use of ^{14}C labelled urea was the possibility of isotope effects (Rabinowitz *et al.*, 1956)

2.5 Absorption of molecular urea and other forms of nitrogen

Most plant roots were thought to absorb nitrate more rapidly than ammonium (Viets, 1965), although there were reports (Fried *et al.*, 1965, Sprutt and Gasser 1970) that young seedlings preferred ammonium. In several experiments greater amounts of nitrogen were absorbed and better growth was observed when rice was supplied with the ammonium (NH_4^+) rather than the nitrate (NO_3^-) form of nitrogen (Doi, 1951, Patrik and Sturgis, 1958). Tanaka *et al* (1959) obtained comparatively better growth and higher grain yield in a culture solution experiment when ammonium was applied upto ear initiation and nitrate at later growth stages. Fried *et al* (1965) observed that ammonium could be absorbed 5-20 times as fast as nitrate, depending on the pH of the medium.

Mc Carthy (1972) studied urea uptake by different species of marine phytoplankton and found that several had high affinities for urea uptake. Active mechanisms for urea transport in Aspergillus (Pateman *et al*, 1973) and in Saccharomyces (Cooper and Sumrada, 1975) were also reported. Healey (1977) reported that nitrogen deficiency increased the initial saturated rate of ammonium and urea uptake by green and blue green algae.

Several workers (Frieberg and Payne, 1957, Dilley and Walker 1961 Mitsui and Kurihara, 1962) reported that urea can enter the plants not only after its decomposition but also as a whole molecule. Freiberg and Payne (1957) noted the absorption of undecomposed urea by the leaves of banana, and Japanese researchers (Mitsui _et al_, 1960, Mitsui and Kurihara, 1962) by the roots of rice. No activity of urease was found in the tissues of roots of rice or in the assimilatory tissues of banana. Molecules of urea were found in the juice of the crushed leaves and in the guttation exuded by corn grown in a solution of urea, which proved the absorption of urea by corn plants (Korenkov, 1966).

Auto radiographs of rice plants fed with ^{14}C urea confirmed the molecular absorption of urea (Saraswathi _et al._, 1991) Quantitatively the accumulation of ^{14}C amounted to 1.8 µg urea h^{-1} g^{-1} plant tissue. It was also suggested that the molecular absorption of the applied urea could be substantial upon top dressing under anaerobic situation

Materials and Methods

MATERIALS AND METHODS

The experiment was conducted at the Radio Tracer Laboratory, College of Horticulture, Kerala Agricultural University. This is located at 10°32' N latitude and 76°16' E longitude at an altitude of 22 m above M.S.L.

The investigation was mainly aimed to find out the absorption of molecular urea and other forms of nitrogen by rice under flooded and non flooded situations. It was also intended to study the influence of soil submergence on urea hydrolysis and also to develop an isotope method for urease estimation

The experiments undertaken during the course of this investigations were as follows.

1 Effect of soil submergence on urea hydrolysis in five soil types of kerala, ie , laterite, kari, kayal, kole and black cotton soils

2 Pot culture experiment using ^{14}C and ^{15}N labelled urea to study the absorption of nitrogen as molecular urea and other forms by flooded and non flooded rice

3 Development of an isotope method for urease estimation.

3.1 Effect of soil submergence on urea hydrolysis

Laterite, kole, kari, kayal and black cotton soils were included in this study

Five gram samples each of the five soil types were submerged for varying period of 0, 5, 10, 20 and 30 days prior to application of urea. The soil samples were taken in glass tubes (30 mm x 120 mm, 30 ml capacity and flooded with 5 ml of distilled water and were kept for the specified period of submergence On the elapse of the time specified for submergence, 5 ml of the urea solution (2000 $\mu g\ g^{-1}$ soil) was added and incubated for different intervals of time namely, 5, 12, 24, 48, 72, 120, 240, 480 and 720 h Sufficient number of replications were kept to allow the removal of samples in duplicate for each soil at different intervals At the end of each incubation period urease activity was estimated by the non-buffer method (Zantua and Bremner, 1975)

3 1 1 Measurement of Eh and pH

The Eh and pH were also measured for all the samples at the end of each submergence period The pH was measured by a combination glass calomel electrode. The Eh was measured by a combination Pt-reference (Silver chloride) electrode connected to a redox meter (TOA Electronics Ltd , Japan

3.1.2 Physico chemical properties of soil

The physico chemical properties of laterite, kari, kayal, kole and black cotton soils are presented in Table 1.

3.2 Pot culture experiment with rice

An alternate stable isotope radio labelling technique was employed in this study.

Three levels of nitrogen (50, 100 and 150 kg ha^{-1}) was applied to the rice grown in pots under flooded and non flooded (60 per cent field capacity) conditions. The three levels of nitrogen were given in two equal splits as basal and one week prior to panicle initiation stage. ^{14}C labelled urea and ^{15}N labelled urea were used alternately to supply the split doses. The detailed programme of the experiment is given below.

Surface samples (0-15 cm) of laterite soil from the Agricultural Research Station, Mannuthy were used for the pot culture experiment. The physico-chemical properties of the soil are presented in Table 3.1 1. Five kilogram of the air dried soil which was ground to pass through a 2 mm sieve was filled in sixty plastic buckets of 7 litre capacity. Half of the filled buckets were kept at field capacity and the remaining at 60 per cent field capacity. Ten pre-soaked seeds of variety Jaya were dibbled, one in each hole at 1-2 cm depth per pot. Later thinning was done to retain

Tabel 1. Physico-chemical properties of laterite, kari, kayal, kole and black soils.

1. Mechanical composition

Soil type	Fraction (per cent composition)			Textural class	Procedure adopted
	Sand	Silt	Clay		
Laterite	85.0	12.5	2.5	Loamy sand	
Kari	62.5	26.25	7.5	Sandy loam	Hydrometer method (Bouyoucos, 1962)
Kayal	75.0	8.75	16.25	Sandy loam	
Kole	55 0	23.75	21.25	Sandy clay loam	
Black	73 75	10.0	16.25	Sandy loam	

2. Physical constants

Soil type	Maximum water holding capacity (%) (Keen Raczhowski Box method Piper, 1950)	Field Capacity (%) (gravimetric method)
Laterite	30.0	20.5
Kari	59.4	25.71
Kayal	74.1	37.77
Kole	67.4	31.3
Black	72.86	35 05

3 Chemical properties

Soil characteristics	Soil type					Method used
	Laterite	Kari	Kayal	Kole	Black	
Organic carbon (per cent)	0.38	3.96	1.86	1.57	0.63	Walkley and Black method (Piper, 1950)
Total N (per cent)	0.154	0.35	0.29	0.26	0.14	Micro Kjeldahl method (Jackson, 1958)
Available P (ppm)	19.0	5.0	1.5	2.5	7.25	Chlorostannous reduced molybdo phosphoric blue colour method (Jackson, 1958)
Available K (ppm)	70.0	25.0	140	140	350	Flame photometric method (Jackson, 1958)
pH	4.7	2.5	4.4	4.7	8.0	Elico' pH meter (Jackson, 1958)
Eh (mV)	+320	+480	+420	+415	+310	Model RM-1K oxidation reduction potential meter of TOA Electronics Ltd., Japan
CEC (me 100 g^{-1} soil)	3.8	33.3	16.4	21.3	52.4	Ammonium acetate method (Jackson, 1958)

four plants at the four corners of the bucket. In pots, kept under flooded condition, initially water was sprinkled for 3-4 days and later a thin film of water was maintained during the initial establishment period of the seedlings During the later stages a constant water level of 4-5 cm height was maintained in the flooded pots In pots which were kept at 60 per cent field capacity, watering was done daily at the rate to account for the evapotranspiration loss

3 2 1 Details of the treatments

Crop - Rice

Variety - Jaya

Moisture regimes - 2 (flooded and 60% field capacity)

Dose of N - 3

1 50 kg N ha^{-1}

2. 100 kg N ha^{-1}

3 150 kg N ha^{-1}

Sources of N - 2

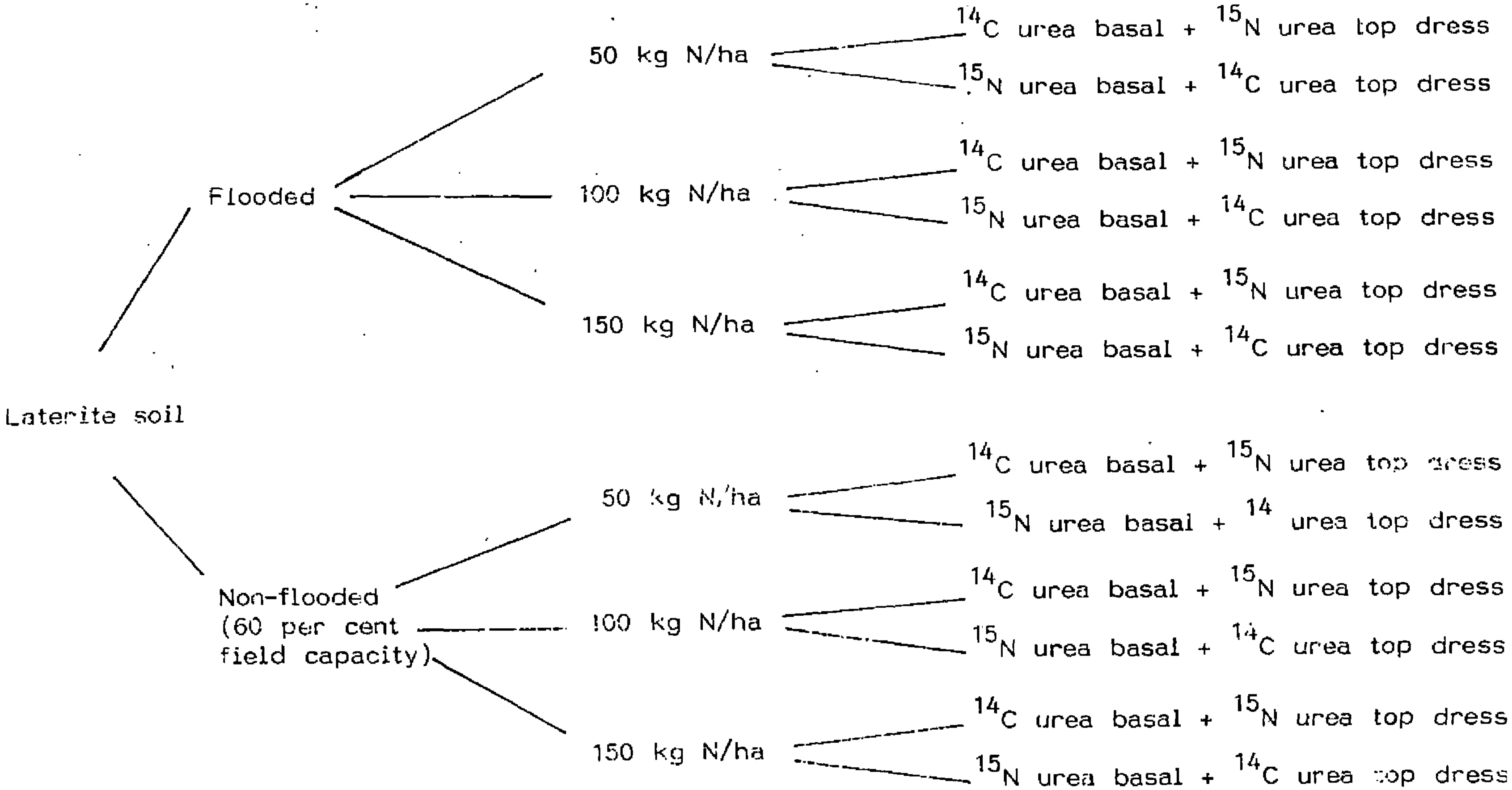
1 ^{14}C labelled urea

2 ^{15}N labelled urea

Method of application

1. ^{14}C urea as basal and ^{15}N urea one week prior to panicle initiation
2. ^{15}N urea as basal and ^{14}C urea one week prior to panicle initiation

3.2.2 Outline of the treatment



- Laterite soil
  - Flooded
    - 50 kg N/ha
      - ^{14}C urea basal + ^{15}N urea top dress
      - ^{15}N urea basal + ^{14}C urea top dress
    - 100 kg N/ha
      - ^{14}C urea basal + ^{15}N urea top dress
      - ^{15}N urea basal + ^{14}C urea top dress
    - 150 kg N/ha
      - ^{14}C urea basal + ^{15}N urea top dress
      - ^{15}N urea basal + ^{14}C urea top dress
  - Non-flooded (60 per cent field capacity)
    - 50 kg N/ha
      - ^{14}C urea basal + ^{15}N urea top dress
      - ^{15}N urea basal + ^{14} urea top dress
    - 100 kg N/ha
      - ^{14}C urea basal + ^{15}N urea top dress
      - ^{15}N urea basal + ^{14}C urea top dress
    - 150 kg N/ha
      - ^{14}C urea basal + ^{15}N urea top dress
      - ^{15}N urea basal + ^{14}C urea top dress

Design - Factorial C R D.

Number of treatment - 12

Replications - 4

In addition to the 48 pots which received the nitrogen treatment, eight control pots were maintained without applying nitrogen for both the flooded and non flooded situations. Thus there were 48 pots, which received the different levels of nitrogen and 16 pots which received no nitrogen.

Out of the 48 treatment pots, basal application of nitrogen was done to 24 pots with ^{14}C urea and to another 24 pots with ^{15}N urea two weeks after sowing. The three basal levels of nitrogen applied was at the rate of 25 kg N ha^{-1}, 50 kg N ha^{-1} and 75 kg N ha^{-1} in the flooded and non flooded conditions. Immediately after applying urea, 4-5 cm level of water was maintained in the pots which were to be kept at flooded situations.

3.2.3 Preparation of ^{14}C urea

^{14}C urea was obtained from Isotope Division, B.A.R.C., Trombay. 500 ml of ^{14}C urea solution was prepared having an activity of 2 µCi/ml and containing 12.5 mg of urea-N ml^{-1}

3.2.4 Preparation of ^{15}N urea

^{15}N enriched urea was obtained from Rashtria Chemicals and Fertilizers, Bombay, having an atom excess of 5% 13 6 g of ^{15}N urea was dissolved in 500 ml to get a concentration of 12 5 mg urea N ml^{-1}

To supply the basal doses of nitrogen, ie 25, 50 and 75 kg N ha^{-1}, 5 ml, 10 ml and 15 ml each of ^{14}C or ^{15}N urea solutions were applied as per the treatment

Phosphorus and potassium were applied at the rate of 45 kg each per hectare in the form of KH_2PO_4 and KCl to all the pots including the control pots

One week prior to panicle initiation ^{14}C urea was top dressed in pots which received ^{15}N urea basaly and ^{15}N urea was top dressed in pots which received ^{14}C urea basaly. N was top dressed at the rate of 25, 50 and 75 kg N ha^{-1} by applying 5 ml, 10 ml and 15 ml each of ^{14}C or ^{15}N urea solutions as per the treatment. After 120 days of maturation, harvesting was done by cutting the stem at 3 cm above the soil surface to avoid any possible contamination from soil or flood water The panicles were harvested first and then the culms were harvested individually, washed the bottom with distilled water, wiped with tissue paper, kept in paper bag and dried in hot air oven at 70°C The grains after detaching from the rachis were dried separately for each pot The rachillae were dried along with the straw After drying, the straw was cut into small pieces and later the dry weights were taken separately for grain and straw.

3.2.5 ^{15}N analysis

The total nitrogen in samples was determined by kjeldahl digestion and distillation. The titrated distillate was made acidic with few drops of 0.05 N HCl and evaporated to dryness by keeping it in a hot air oven at not more than 50°C. The residue was then diluted with distilled water to get a concentration of 1 mg N ml^{-1}. This was then filled in capillary tubes of length 1-2 cm. The capillary tubes containing 9-11 μg N were then oven dried at 50°C. The ^{14}N : ^{15}N ratio estimation was done using Emission Spectrometer available at N.R.L., IARI, New Delhi.

3.2.6 Radio assay of plant samples

The radioactivity of the plant samples was measured following internal standard method to correct the observed countrates for differential quenching in the samples.

The plant samples, both grain and straw were powdered in a mixer-cum-grinder and sub samples of 0.2 g were taken. The weighed sample was added in a liquid scintillation vial, containing 15 ml of dioxan based liquid scintillator and the radio activity was determined within 10 minutes, in the liquid scintillation system. Immediately after taking counts, 0.1 ml of the stock solution of known activity (10 n Ci ml^{-1}) was added and the counts were again taken From this the counting efficiency was calculated for each sample and the dpm for each sample was worked out from the corresponding efficiency values.

The liquid scintillator used in the studies consisted of 4 g PPO 0 2 g POPOP, 60 g naphthalene, 100 ml methanol and 20 ml ethylene glycol diluted to 1000 ml with dioxan. The radio activity was determined in a microcomputer controlled liquid scintillation system (Rackbeta 1215 of Wallac OY, Finland)

3 2 7 Computation of uptake parameters

a) Per cent N derived from fertilizer in plants from both basal and top dressing (% Ndff)

$$= \frac{\left[\begin{array}{l}\text{\% atom excess in the plant}\\ \text{from basal application}\end{array}\right] + \left[\begin{array}{l}\text{\% atom excess in the plant}\\ \text{from top dressing}\end{array}\right] \times 100}{\text{\% atom excess in the fertilizer}}$$

b) % atom excess in whole plant receiving basal application of ^{15}N

$$= \frac{\left[\dfrac{\text{\% } ^{15}\text{N atom excess in straw x mg N in straw}}{100}\right] + \left[\dfrac{\text{\% } ^{15}\text{N atom excess in grain x mg N in grain}}{100}\right] \times 100}{\text{Total N uptake}}$$

c) % atom excess in whole plant receiving top dressing of ^{15}N

$$= \frac{\left[\dfrac{\text{\% } ^{15}\text{N atom excess in straw x mg N in straw}}{100}\right] + \left[\dfrac{\text{\% } ^{15}\text{N atom excess in grain x mg N in grain}}{100}\right] \times 100}{\text{Total N uptake}}$$

d) % Ndfs = 100 - % Ndff

e) Fertilizer N uptake $= \frac{\%\ \text{Ndff} \times \text{total N uptake}}{100}$

f) % N recovery $= \frac{\text{Fertilizer N uptake}}{\text{Fertilizer N applied}} \times 100$

g) Quantity of nitrogen taken up as molecular urea by grain or straw

$$= \frac{\text{Total dpm in the plant part}}{\text{Specific activity of the fertilizer}} \times 0.46$$

h) Quantity of nitrogen taken up as molecular urea by the plant

= Quantity of nitrogen taken up as molecular urea by the grain + Quantity of nitrogen taken up as molecular urea by the straw

i) Quantity of N taken up as molecular urea by the plant from split doses

$$= \frac{\text{total dpm for the grain from split dose} \times 0.46}{\text{specific activity of the fertilizer}} + \frac{\text{total dpm for the stem from split dose} \times 0.46}{\text{specific activity of the fertilizer}}$$

j) % absorption of molecular urea nitrogen by grain, straw and by the plant

$$= \frac{\text{Quantity of nitrogen absorbed as molecular urea}}{\text{Fertilizer N uptake}} \times 100$$

k) Absorption of nitrogen in forms other than molecular urea

= Fertilizer N uptake - quantity of nitrogen absorbed as molecular urea

l) % absorption of nitrogen in forms other than molecular urea

$$\frac{[\text{Fertilizer N uptake} - \text{quantity of nitrogen absorbed as molecular urea}] \times 100}{\text{Fertilizer N uptake.}}$$

m) analysis of the data

To know the direct effect of the nitrogen levels, analysis was done in 2 x 4 factorial C.R.D with 8 replications (2 levels of moisture, ie, flooded and non-flooded, and 4 levels of nitrogen), disregarding the nature of labelling and method of application. To study the effect of moisture regimes and levels of nitrogen on different parameters like fertilizer N uptake, molecular urea N absorption etc. the data of the two pots which received the alternately labelled urea were combined together and the total uptake of ^{14}C labelled urea or ^{15}N labelled urea for a particular level of nitrogen from top or basal dressing was calculated. The data was analysed as a 2 x 3 factorial C.R.D with 4 replications. To study the effect of the split doses of N applied (top and basal dressings) the data were analysed as a 2 x 3 x 2 factorial C.R.D. (2 moisture regimes x 3 levels of nitrogen x 2 methods of application) with 4 replications.

3.3 Development of an isotope method for urease estimation

Laterite, kari, kayal, kole and black cotton soils were included in this study. To five gram portion of the soil sample taken in a 100 ml conical flask was added 5 ml of ^{14}C labelled urea solution, containing urea at the rate of 2 mg ml^{-1} and having a specific activity of 102.08 cpm ^{14}C ug^{-1} urea. The soil was then incubated for 5 h. The experiment was done with four replication for each soil. After 5 h, the soil samples were shaken with 50 ml of 2 M KCl-PMA solution for 60 min and the resulting soil suspension was filtered through Whatman No.42 filter paper. The extract was used to estimate urease activity by nonbuffer method of Zantua and Bremner (1975) and by liquid scintillation counting.

For liquid scintillation counting, an aliquot of the extract was taken in a liquid scintillation vial, containing 15ml of dioxan based liquid scintillation and the radio activity was determined. From the specific activity of ^{14}C urea solution initially added, and the count rates obtained for the KCl-PMA extract, the urea hydrolysis rate was calculated.

To study the influence of varying periods of incubation on hydrolysis of ^{14}C labelled urea, kari and black cotton soils were incubated with urea solution for 24, 48 and 72 h. The urea hydrolysis was estimated by the non-buffer method of Zantua and Bremner (1975) and by liquid scintillation counting.

3.4 Statistical analysis

The statistical analysis were done according the methods suggested by Panse and Sukhatme (1985).

Results

RESULTS

The following experiments were conducted during the course of this investigation and the results of the same are presented in this chapter

1 Effect of soil submergence on urea hydrolysis

2 Absorption of molecular urea and other forms of nitrogen by flooded and non-flooded rice from split doses of nitrogen

3 Development of isotope method for urease estimation

4.1 Effect of soil submergence on urea hydrolysis

The experiment was aimed to find out the effect of submergence of soil for different intervals on urease activity

In this study soil was submerged for 0, 5, 10, 20 and 30 days and at the end of each submergence period, incubation with urea was done for 5, 12, 24, 48 72, 120, 240 480 and 720 h.

4 1 1 Urease activity under non-submergence and submergence for 5 h incubation

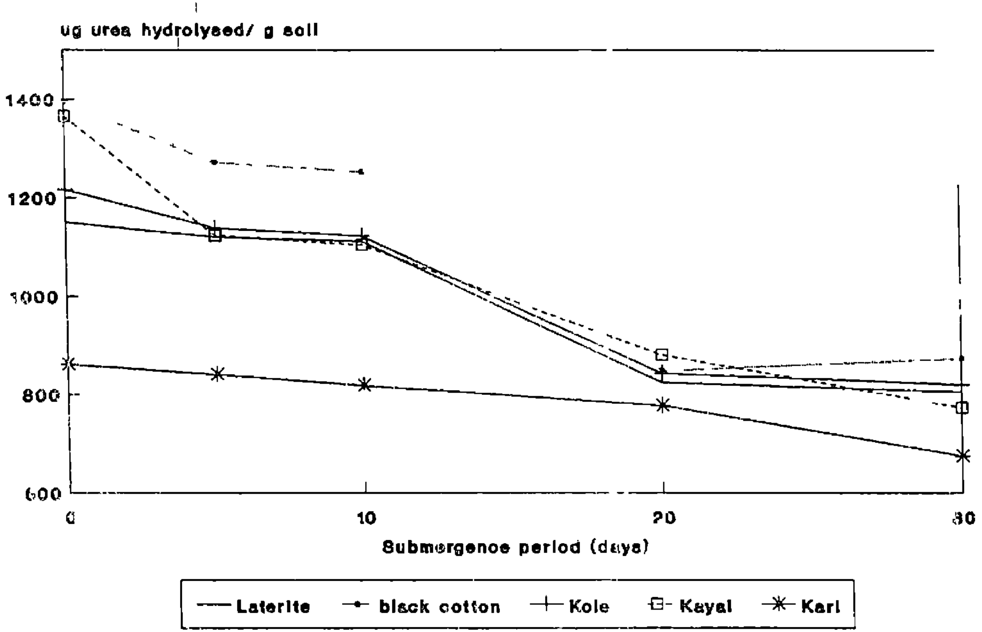
The effect of 5 h incubation with urea under non-submergence and submergence will be presented here (Table 2) as 5 h is the standard incubation time in the non-buffer method of Zantua and Bremner (1975)

Table 2 Urease activity under non-submergence and submergence for 5 h incubation

Soil	Urease activity (*μg urea hydrolysed g^{-1} soil)				
	Non-submergence	Submergence period (days)			
		5	10	20	30
Laterite	1150	1120	1111	824	805
Black cotton	1396	1272	1253	848	873
Kole	1216	1138	1122	843	820
Kayal	1365	1123	1104	880	773
Kari	861	840	818	777	675

* Amount of urea added initially is 2000 μg g^{-1} soil

Fig.1. Soil urease activity under submergence and non-submergence for 5 h incubation with urea.



4.1.1.1 Under non-submergence

Among the different soils studied, Black Cotton Soil registered the highest activity (1396 μg urea g^{-1} soil) and the least activity was recorded by the kari soil (861 μg urea g^{-1} soil). The urease activity of other soils ranged from 1150 to 1365 μg urea g^{-1} soil.

4.1.1.2 Under submergence

A steady decline in urease activity was noticed following submergence of the soil. Though the decline in urease activity was slow upto a submergence period of 10 days, there was almost a 30-40% drop in the activity following submergence periods of 20-30 days for all soils except kari soil In Kari soil submergence had not much effect on urease activity and only a 10-20% decline was noticed even for submergence periods of 20-30 days

4.1.1.3 Influence of submergence under prolonged incubation with urea

The data are presented in Table 3. The decline in the urease activity following submergence was less pronounced when the incubation with urea after the submergence was increased beyond 5 h. Under non-submergence urea hydrolysis was completed within a day in the case of kayal soil and within two days in the case of laterite and kole and within three days in the case of black cotton soil. However, with respect to kari soil, urea hydrolysis

Table 3 Influence of soil submergence on urease activity under prolonged incubation with urea

Soil	Submergence period (days)	*Urea hydrolysed (µg g^{-1} soil)								
		Incubation period (h)								
		5	12	24	48	72	120	240	480	720
Laterite	0	1150	1290	1626	2000	2000	2000	2000	2000	2000
	5	1120	1268	1384	1453	1813	2000	2000	2000	2000
	10	1111	1133	1440	1496	1720	2000	2000	2000	2000
	20	824	1044	1280	1480	1701	2000	2000	2000	2000
	30	805	917	1273	1346	1680	2000	2000	2000	2000
Black cotton	0	1396	1346	1495	1850	2000	2000	2000	2000	2000
	5	1272	1328	1415	1567	1701	2000	2000	2000	2000
	10	1253	1328	1440	1553	1720	2000	2000	2000	2000
	20	848	1123	1423	1482	1684	2000	2000	2000	2000
	30	873	1110	1216	1328	1795	2000	2000	2000	2000
Kole	0	1216	1481	1665	2000	2000	2000	2000	2000	2000
	5	1138	1313	1527	2000	2000	2000	2000	2000	2000
	10	1122	1304	1720	2000	2000	2000	2000	2000	2000
	20	843	1085	1696	2000	2000	2000	2000	2000	2000
	30	820	1412	1717	2000	2000	2000	2000	2000	2000
Kayal	0	1365	1776	2000	2000	2000	2000	2000	2000	2000
	5	1123	1301	1421	2000	2000	2000	2000	2000	2000
	10	1104	129[illegible]	1394	2000	2000	2000	2000	2000	2000
	20	880	97[illegible]	1356	2000	2000	2000	2000	2000	2000
	30	973	1188	1253	2000	2000	2000	2000	2000	2000
Kari	0	861	936	1122	1206	1255	1290	1356	1683	1850
	5	840	916	1100	1229	1260	1257	1309	1346	1355
	10	818	900	1097	1197	1216	1232	1258	1328	1346
	20	777	889	1035	1200	1210	1343	1393	1468	1677
	30	675	820	973	1281	1356	1384	1431	1496	1683

*Amount of urea added initially is 2000 µg g^{-1} soil

Fig.2. Influence of submergence of laterite soil on urea hydrolysis upon prolonged incubation with urea

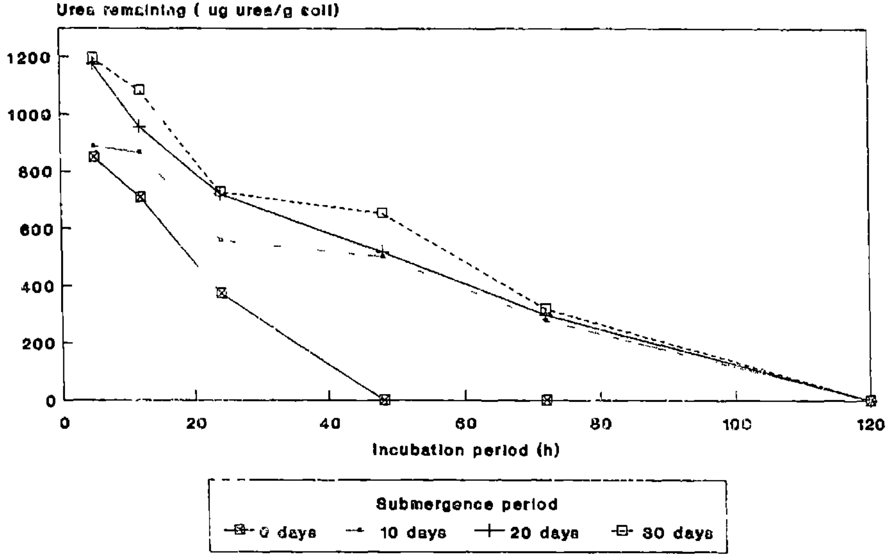


Fig.3. Influence of submergence of black cotton soil on urea hydrolysis upon prolonged incubation with urea

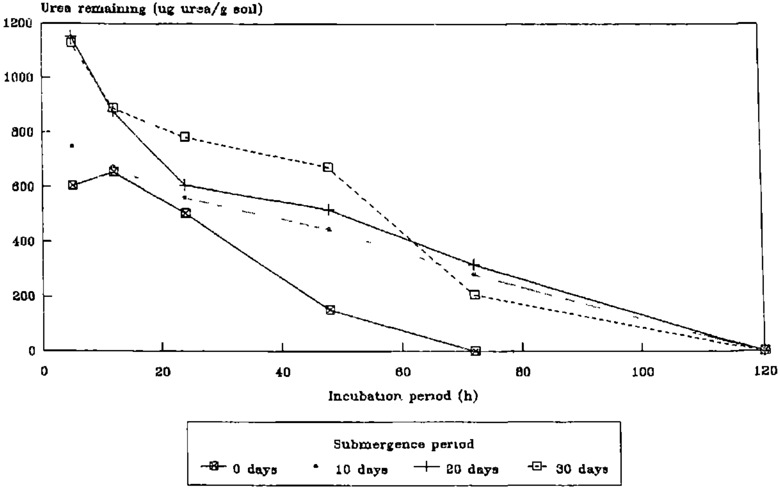


Fig.4. Influence of submergence of kole soil on urea hydrolysis upon prolonged incubation with urea

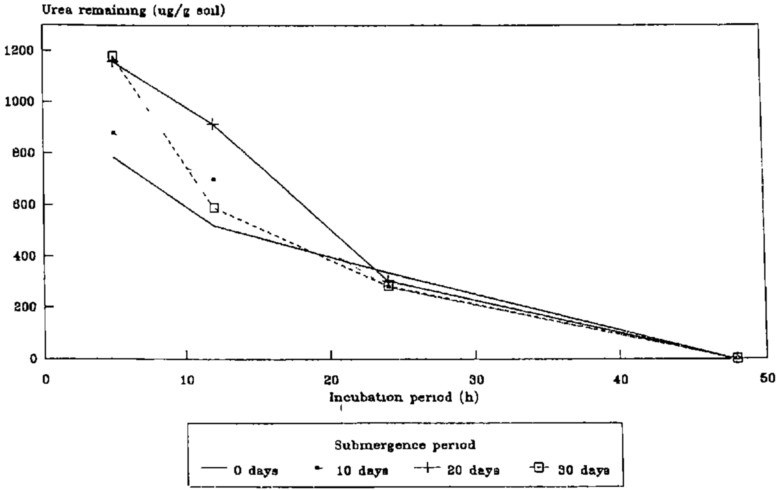


Fig.5. Influence of submergence of kayal soil on urea hydrolysis upon prolonged incubation with urea

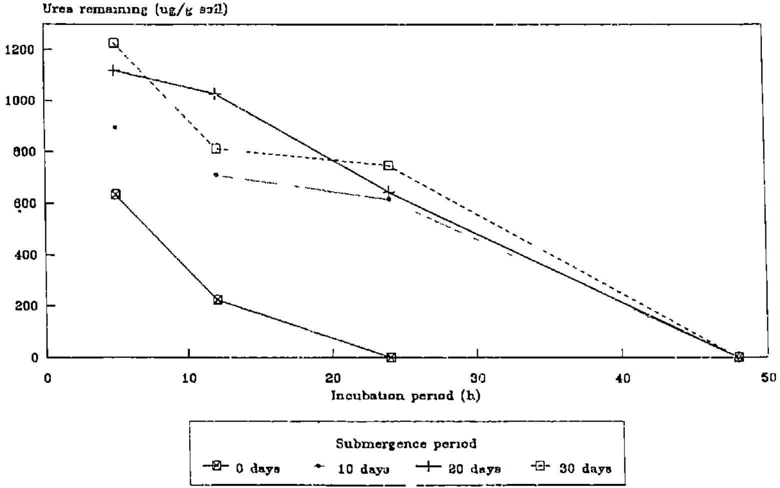
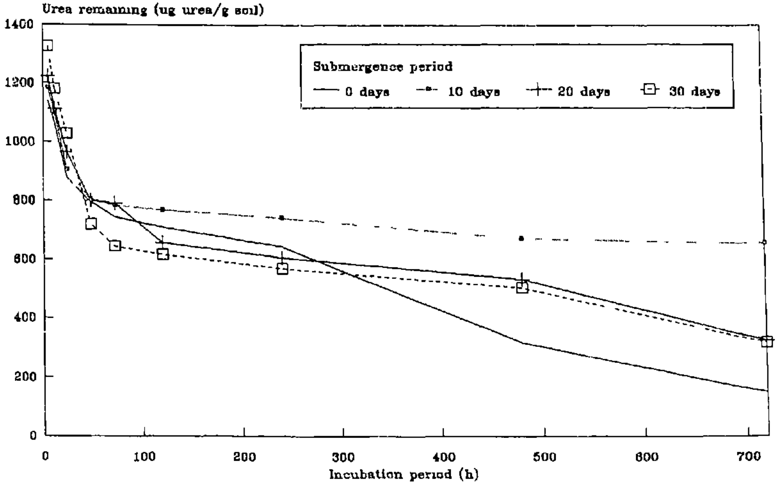


Fig.6. Influence of submergence of kari soil on urea hydrolysis upon prolonged incubation with urea



was not completed even when incubated for 30 days Submergence had not much effect on urea hydrolysis when the incubation with urea was prolonged for more than 5-10 days Even when submerged for as long as 30 days, urea hydrolysis was completed within 3 days in all the soils except Kari In Kari soil, as already mentioned urea hydrolysis was not completed under both non-submerged and submerged conditions even when incubated with urea upto 30 days. However, submergence had a slight retarding effect on urea hydrolysis

4 1 2 Changes in Eh and pH of the different soils under submergence

A slight increase in pH was observed for all soils, except black cotton soil following flooding In laterite, Kayal and Kole the pH increased almost by a unit. In Kari only a slight increase in pH was observed. However, in black, the pH decreased from 7 9 to 7 4 The flooding also had a profound influence on the Eh of the soil While a considerable reduction in the Eh of the laterite, kayal and kole was observed, a similar reduction in the Eh of Kari and Black soil was not observed following flooding of the soil for the different intervals. In the case of the kari soil even after a prolonged submergence for 30 days, there was only a slight reduction in Eh (from +465 to +265)

Table 5 Changes in Eh and pH of soil following flooding

Flooding period (h)	Soil type									
	Laterite		Kari		Kayal		Kole		Black	
	Eh(mV)	pH	Eh(mV)	pH	Eh(mV)	pH	Eh(mV)	pH	Eh(mV)	pH
0	+310	4.75	+485	2.45	+435	4.4	+400	4.6	+320	7.9
120	-60	6.3	+420	2.65	+10	5.4	+90	5.8	+150	7.5
240	-25	7.25	+340	2.90	-10	5.6	-30	5.95	+160	7.7
480	-50	6.6	+360	2.70	+40	5.4	-90	5.9	+130	7.55
720	-35	6.3	+265	2.50	+10	5.4	-60	5.45	+100	7.40

4.2 Absorption of molecular urea and other forms of nitrogen by flooded and non-flooded rice from split doses of nitrogen

In this experiment, three levels of nitrogen ie., 50, 100 and 150 kg N ha^{-1} was applied to flooded and non-flooded rice in the form of ^{14}C and ^{15}N labelled urea

The information generated from this experiment is presented in the following lines.

i) Influence of level of nitrogen and *moisture regimes on yield, dry matter production and nitrogen content (disregarding the source of nitrogen and method of application).

ii) N recovery and forms of absorption of nitrogen as influenced by moisture regimes and levels of nitrogen using the alternately labelled urea (disregarding the split method of application).

iii) Influence of split application on the relative uptake of forms of N (from basal and top dressing).

4 2 1 Influence of levels of nitrogen and moisture regimes on yield, dry matter production and nitrogen content

The objective of this part of the study was to know direct effect of different levels of nitrogen on the different parameters disregarding the source (nature of labelling) and the method of application (split) under flooded and non-flooded situations.

* Moisture regime refers to the flooded and non-flooded treatments and is used in this sense here after

In the original set of treatments there were three levels of nitrogen and two moisture regimes (flooded and non-flooded) and each level of nitrogen was applied in two equal splits, in the form of ^{15}N or ^{14}C labelled urea either as basal or as top dressing While four pots received ^{14}C basal and ^{15}N top, four other pots received ^{15}N basal and ^{14}C top for each level of nitrogen If the nature of labelling is disregarded, each level of nitrogen was applied to eight pots Hence to know the direct effect of different levels of nitrogen, analysis was done in 2 x 4 factorial C R.D with eight replications (2 levels of moisture and 4 levels of nitrogen). In this set of experiment, in addition to the three levels of nitrogen data obtained from control pots (0 kg N ha^{-1}) were also included.

4 2 1 1 Yield of grain (g pot^{-1})

Both moisture regimes (flooded and non-flooded situations) and level of nitrogen (Table 6) had a significant influence on grain yield The flooded rice recorded almost four times higher yield than the non-flooded rice. With respect to the nitrogen levels, 150 kg N ha^{-1} registered the highest yield which was on par with that obtained at 100 kg N ha^{-1} Both these were significantly superior to the yield recorded at zero and 50 kg N ha^{-1} The interaction effect (Table 7) between moisture regimes and levels of nitrogen was also found significant The flooded rice

Table 6. Grain yield as influenced by the moisture regime and level of nitrogen

Treatment	Grain yield (g pot^{-1})
Flooded	5.68^{a}
Non-flooded	1.41^{b}
SEm±	0.15
CD (0.05)	0.506
0 kg N ha^{-1}	1.34^{c}
50 kg N ha^{-1}	3.59^{d}
100 kg N ha^{-1}	3.85^{e}
150 kg N ha^{-1}	4.39^{e}
SEm±	0.21
CD (0.05)	0.598

Table 7. Grain yield (g pot^{-1}) as influenced by the interaction between moisture regime and level of nitrogen

Moisture regime	Level of Nitrogen (kg N ha^{-1})			
	0	50	100	150
Flooded	2.12^{a}	5.41^{b}	7.86^{c}	7.33^{c}
Non-flooded	0.55^{d}	1.78^{e}	1.80^{e}	1.46^{e}

SEm± 0.30
CD (0.05) 0.846

Values followed by the same alphabet are not significantly different

responded better to the nitrogen application than the non-flooded rice Under the non-flooded condition while the 50 kg N ha^{-1} was found at par with 100 and 150 kg N ha^{-1}, under the flooded condition both the 100 and 150 kg N ha^{-1} were significantly superior to zero and 50 kg N ha^{-1}

4 2 1 2 Yield of straw (g pot^{-1})

The flooded rice recorded a significantly higher straw yield than the non-flooded rice (Table 8) Among the nitrogen levels, 100 kg N ha^{-1} registered the highest yield which was significantly superior to the yield recorded at other levels Interaction effect (Table 9) was also found significant. While the flooded rice responded favourably to higher levels of nitrogen, the non-flooded rice recorded a decrease at 150 kg N ha^{-1}

4 2 1 3 Total dry weight (g pot^{-1})

As in the case of grain and straw yield, the flooded rice recorded a significantly higher dry matter production than the non-flooded rice (Table 10). With respect to the nitrogen levels, 100 g N ha^{-1} recorded the highest dry weight. Though this was on par with the straw yield at 150 kg N ha^{-1}, it was significantly superior to that recorded at 0 and 50 kg N ha^{-1} The interaction effect was also observed to be significant (Table 11) while the dry weight of flooded rice increased with increase in nitrogen

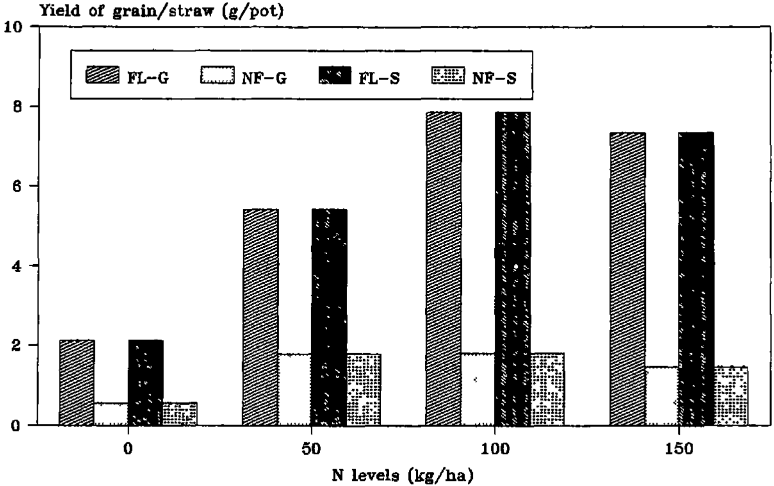


Fig. 7. Yield of grain and straw as influenced by N levels and moisture regimes

FL– flodded; G– grain yield
NF– non–flooded; S– straw yield

Table 8 Straw yield as influenced by moisture regime and level of nitrogen

Treatment	Straw yield (g pot^{-1})
Flooded	6.34^{a}
Non-flooded	5.25^{b}
SEm±	0.19
CD (0.05)	0.545
0 kg N ha^{-1}	2.73^{c}
50 kg N ha^{-1}	5.69^{d}
100 kg N ha^{-1}	7.78^{e}
150 kg N ha^{-1}	6.98^{f}
SEm±	0.28
CD (0.05)	0.770

Table 9 Straw yield (g pot^{-1}) as influenced by the interation between moisture regime and level of nitrogen

Moisture	Levels of nitrogen (kg N ha^{-1})			
	0	50	100	150
Flooded	3.30^{a}	6.03^{b}	7.79^{c}	8.21^{e}
Non-flooded	2.14^{d}	5.35^{b}	7.77^{c}	5.74^{c}

SEm± 0.39
CD (0.05) 1.089

Values followed by the same alphabet are not significantly different

Table 10 Total dry weight as influenced by the moisture regime and level of nitrogen

Treatment	Total dry weight (g pot^{-1})
Flooded	11.51^{a}
Non-flooded	6.67^{b}
SEm±	0.39
CD (0.05)	1.08
0 kg N ha^{-1}	4.07^{c}
50 kg N ha^{-1}	9.29^{d}
100 kg N ha^{-1}	11.62^{e}
150 kg N ha^{-1}	11.37^{e}
SEm±	0.55
CD (0.05)	1.53

Table 11 Total dry weight (g pot^{-1}) as influenced by the interaction between moisture regime and level of nitrogen

Moisture regime	Level of nitrogen (kg N ha^{-1})			
	0	50	100	150
Flooded	5.45^{a}	11.43^{b}	13.62^{c}	15.54^{c}
Non-flooded	2.69^{d}	7.16^{e}	9.61^{f}	7.21^{e}

SEm± 0.78
CD (0.05) 2.165

Values followed by the same alphabet are not significantly different

levels, a decrease in the dry matter yield of non-flooded rice was noticed at 150 kg N ha^{-1}.

4 2 1 4 Nitrogen content in grain (%)

Though the moisture regimes had no significant influence on the nitrogen content in grain, the N levels had a significant influence The highest per cent of N was recorded at 100 kg N ha^{-1} which was significantly superior to that recorded at other levels of nitrogen (Table 12) The interaction effect was also found to be significant Highest N content in grain was recorded by the non-flooded rice at 100 kg N ha^{-1} This was on par with that recorded at 50 kg N ha^{-1} under the same moisture regime and with flooded rice supplied with 100 kg N ha^{-1} (Table 13).

4 2 1 5 Nitrogen content in straw (%)

The non-flooded rice recorded a significantly higher N content in the straw, than the flooded rice Among the nitrogen levels, the highest N per cent was observed at 100 kg N ha^{-1} which was on par with that at 50 and 150 kg N ha^{-1} (Table 14) Interaction effect was also found to be significant The non-flooded rice at 100 kg N ha^{-1} recorded the highest per cent of N. This was on par with that at 50 and 150 kg N ha^{-1} under the same moisture regimes and 100 kg N ha^{-1} under flooded situation (Table 15).

Table 12. Nitrogen content in grain as influenced by the moisture regime and level of nitrogen

Treatment	Nitrogen content (%)
Flooded	1.39
Non-flooded	1 52
SEm±	0 07
CD (0 05)	NS
0 kg N ha^{-1}	0 83^{a}
50 kg N ha^{-1}	1.62^{b}
100 kg N ha^{-1}1	1 97^{c}
150 kg N ha^{-1}	1 38^{d}
SEm±	0 09
CD (0.05)	0 195

NS - Non-significance

Table 13 Nitrogen content in grain (%) as influenced by the interaction between moisture regime and level of nitrogen

Moisture regime	Level of nitrogen (kg N ha^{-1})			
	0	50	100	150
Flooded	1 08	1.41^{b}	1 87^{a}	1.21^{c}
Non-flooded	0.58^{d}	1 84^{a}	2.09^{a}	1.57^{b}

SEm± 0 1[illegible]
CD (0 05) 0 276

Values followed by the same alphabet are not significantly different

Table 14 Nitrogen content in straw as influenced by the moisture regime and level of nitrogen

Treatment	Nitrogen content (%)
Flooded	1.12^{a}
Non-flooded	1.47^{b}
SEm±	0.08
CD (0.05)	0.236
0 kg N ha^{-1}	0.66^{c}
50 kg N ha^{-1}	1.42^{d}
100 kg N ha^{-1}	1.71^{d}
150 kg N ha^{-1}	1.39^{d}
SEm±	0.12
CD (0.05)	0.333

Table 15 Nitrogen content in straw (%) as influenced by the interaction between moisture regime and level of nitrogen

Moisture regime	Level of nitrogen (kg N ha^{-1})			
	0	50	100	150
Flooded	0.79^{a}	0.96^{a}	1.55^{bc}	1.16^{ac}
Non-flooded	0.52^{d}	1.87^{b}	1.87^{b}	1.62^{b}

SEm± 1.17
CD (0.05) 0.472

Values followed by the same alphabet are not significantly different

4.2.1.6 Nitrogen uptake by grain (mg pot^{-1})

The moisture regimes and nitrogen levels (Table 16), had a significant influence on N uptake by grain. The flooded rice recorded a significantly higher N uptake than the non-flooded rice. The highest grain uptake of N was recorded at 100 kg N ha^{-1} which was significantly superior to that observed at other levels of nitrogen. Though the N uptake at 50 and 150 kg N ha^{-1}, was on par, they were significantly superior to that at 0 kg N ha^{-1}.

The interaction effect between moisture regimes and nitrogen levels was also found to be significant (Table 17). The highest N uptake was observed by flooded rice at 100 kg N ha^{-1} which was significantly superior to that recorded at all other levels, under both the moisture regimes. The grain N uptake by the flooded rice at 50 and 150 kg N ha^{-1} though on par was significantly superior to the grain nitrogen uptake of non-flooded rice at all the levels of nitrogen.

4.2.1.7 Nitrogen uptake by straw (mg pot^{-1})

The moisture regimes had no significant influence on the N uptake by straw (Table 18). Among the N level also, 100 kg N ha^{-1} recorded the highest uptake. The interaction effect was not found significant.

Table 16. Nitrogen uptake by grain as influenced by the moisture regime and level of nitrogen

Treatment	Nitrogen uptake (mg pot^{-1})
Flooded	79.90^{a}
Non-flooded	24.29^{b}
SEm±	3.57
CD (0.05)	9.91
0 kg N ha^{-1}	12 41^{c}
50 kg N ha^{-1}	54.67^{d}
100 kg N ha^{-1}	85.54^{e}
150 kg N ha^{-1}	55.77^{d}
SEm±	5.06
CD (0.05)	14.02

Table 17. Nitrogen uptake by grain (mg pot^{-1}) as influenced by by the interaction between moisture regime and level of nitrogen

Moisture regime	Level of nitrogen (kg N ha^{-1})			
	0	50	100	150
Flooded	21 62de	76.92^{b}	132.57^{a}	88.50^{b}
Non-flooded	3.19^{e}	32.42^{cd}	38.50^{c}	23.50^{d}

SEm± 7.15
CD (0.05) 19.82

Values followed by the same alphabet are not significantly different

Table 18 Nitrogen uptake by straw as influenced by the moisture regime and level of nitrogen

Treatment	Nitrogen uptake (mg pot^{-1})
Flooded	73 92
Non-flooded	87 46
SEm±	6 39
CD (0 05)	NS
0 kg N ha^{-1}	18 68^{a}
50 kg N ha^{-1}	79 20^{b}
100 kg N ha^{-1}	130 86^{c}
150 kg N ha^{-1}	94 01^{b}
SEm±	9 05
CD (0 05)	25 08

NS - Non-significance

Values followed by the same alphabet are not significantly different

Table 19. Nitrogen uptake by plant as influenced by the moisture regime and level of nitrogen

Treatment	Nitrogen uptake (mg pot^{-1})
Flooded	153.48^{a}
Non-flooded	111.52^{b}
SEm±	8.15
CD (0.05)	22.59
0 kg N ha^{-1}	30.31^{c}
50 kg N ha^{-1}	133.88^{d}
100 kg N ha^{-1}	216.03^{e}
150 kg N ha^{-1}	149.78^{d}
SEm±	11.52
CD (0.05)	31.93

Values followed by the same alphabet are not significantly different

4 2.1.8 Total N uptake by the plant (mg pot^{-1})

The moisture regimes had a significant influence on total N uptake, the flooded rice recorded a much higher yield than the non-flooded rice (Table 19). Among the N levels 100 kg N ha^{-1} recorded the highest total uptake which was significantly superior to that observed at other levels of nitrogen. The interaction effect was not found significant.

4 2 2 Nitrogen recovery and forms of absorption of nitrogen as influenced by moisture regimes, levels of nitrogen and the split method of application, using the alternately labelled urea

This part of the study includes two sections. In the first section, the influence of moisture regimes and levels of nitrogen on % Ndff, % Ndfs, N recovery and uptake to molecular urea were studied using the alternately labelled urea, disregarding the effect of split application. In the second section the influence of the *timing of N application (split method of application) on the relative uptake of forms of nitrogen was studied.

In the original set of treatments each level of N was applied in two equal splits, when the first split was applied basally as ^{14}C labelled urea, the second split was applied as top dress as ^{15}N labelled urea to the same pot The same level of nitrogen

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* This is also referred to as method of application in some parts of this dissertation.

was also applied as ^{15}N labelled urea basally and ^{14}C labelled urea as top dress

To study the effect of moisture regimes and level of nitrogen on different parameters, the data of the two pots which received the alternately labelled urea were combined together and the total uptake of ^{14}C labelled urea or ^{15}N labelled urea for a particular level of nitrogen from top and basal dressing was calculated. Thus there were 6 treatments (2 moisture regime and 3 levels of nitrogen) and it was analysed in 2 x 3 factorial C.R.D. with 4 replications.

4 2 2 1 Percentage Ndff and percentage Ndfs

Disregarding the split method of application, the influence of moisture regimes and levels of nitrogen on % Ndff and % Ndfs was studied

The moisture regimes had no significant influence on % Ndff or % Ndfs (Table 20, 21). The % Ndff increased with increasing levels of nitrogen application. A reverse trend was observed in the case of % Ndfs. The interaction effect was not found significant.

4.2.2.2 Fertilizer N uptake (mg pot^{-1})

4 2.2.2 1 Effect of moisture regimes and levels of nitrogen

Flooded rice recorded a significantly higher N uptake than the non-flooded rice (Table 22) Among the N levels, higher fertilizer

Table 20. Ndff(%) as influenced by moisture regime and level of nitrogen

Treatment	Ndff (%)
Flooded	43.43
Non-flooded	42.93
SEm±	0.93
Cd (0.05)	NS
50kg N ha^{-1}	33.10^{a}
100 kg N ha^{-1}	44.69^{b}
150 kg N ha^{-1}	51.73^{c}
SEm±	1.14
CD (0.05)	2.775

NS - Non-significance

Values followed by the same alphabet are not significantly different

Table 21. Ndfs (%) as influenced by moisture regime and level of nitrogen

Treatment	Ndfs (%)
Flooded	56.57
Non-flooded	57.07
SEm±	0.93
CD (0.05)	NS
50 kg N ha^{-1}	66.89^{a}
100 kg N ha^{-1}	55.30^{b}
150 kg N ha^{-1}	48.26^{c}
SEm±	1.14
CD (0.05)	2.775

NS - Non-significance

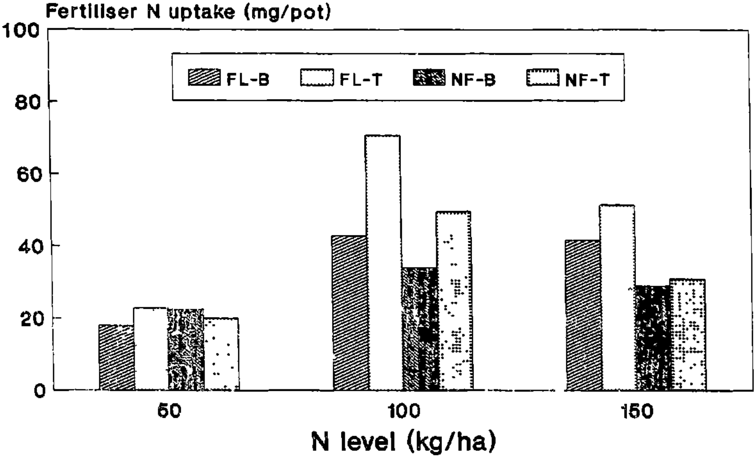
Values followed by the same alphabet are not significantly different

Table 22 Fertilizer N uptake as influenced by moisture regime and level of nitrogen

Treatment	Fertilizer N uptake (mg pot^{-1})
Flooded	82.45^{a}
Non-flooded	61.79^{b}
SEm±	5.51
CD (0.05)	16.37
50 kg N ha^{-1}	41.38^{c}
100 kg N ha^{-1}	98.55^{d}
150 kg N ha^{-1}	76.42^{e}
SEm±	6.74
CD (0.05)	20.05

Values followed by the same alphabet are not significantly different

Fig.8. Fertiliser N uptake by rice as influenced by moisture regime, level of nitrogen and method of application



FL-flooded; NF-non-flooded;
T- half top dress;B- half basal dress

N uptake was noticed at 100 kg N ha^{-1} followed by that at 150 kg N ha^{-1} and 50 kg N ha^{-1}. The interaction affect was found non-significant.

4 2 2.2.2 Effect of split application

It was observed that fertilizer nitrogen uptake from the top dressing was significantly higher than that from the basal dressing. However, its interaction with moisture regime was not found significant (Table 23). However, it was found that though non-significant, the fertilizer nitrogen uptake by the flooded rice from the basal dressing was higher than that by the non-flooded rice from top dressing.

The interaction effect between method ot application and nitrogen levels was also not significant (Table 24). However, the highest fertilizer nitrogen uptake was noticed at 100 kg N ha^{-1} when top dressed and the least uptake was noticed from the basally applied N at the rate of 50 kg ha^{-1}.

The three way interaction effect among the moisture regimes, levels of nitrogen and methods of application was also found non-significant (Table 25). The highest uptake though non-significant was recorded by the flooded rice at 100 kg N ha^{-1} applied as top dressing.

Table 23. Fertilizer N uptake as influenced by the interaction between moisture regime and level of nitrogen

Moisture regime	Fertilizer N uptake (mg pot^{-1})	
	Half basal ^{15}N	Half top-dress ^{15}N
Flooded	34.08	48.33
Non-flooded	28.34	33.44
Mean	31.21^{a}	40.88^{b}

Method of application	SEm±	2.47	CD (0.05)	6.85
Moisture regime x method of application	SEm±	3.49	CD (0.05)	NS

Table 24 Fertilizer N uptake as influenced by the interaction between level of nitrogen and method of application

Level of nitrogen (kg N ha^{-1})	Fertilizer N uptake (mg pot^{-1})	
	Half basal ^{15}N	Half top-dress ^{15}N
50	20.07	21.32
100	38.55	60.19
150	35.22	41.14
Mean	31.21^{a}	40.88^{b}

Method of application	SEm±	2.47	CD (0.05)	6.85
Level of nitrogen x Method of application	SEm±	4.28	CD (0.05)	NS

NS - Non-significance

Values followed by the same alphabet are not significantly different

Table 25. Fertilizer N uptake as influenced by the interaction between moisture regime, level of nitrogen and method of application

Level of nitrogen ($kg\ N\ ha^{-1}$)	Fertilizer N uptake ($mg\ pot^{-1}$)			
	Flooded		Non-flooded	
	Half basal ^{15}N	Half top-dress ^{15}N	Half basal ^{15}N	Half top-dress ^{15}N
50	17.84	22 79	22.29	19.85
100	42.84	70 74	33 87	49.65
150	41.57	51 46	28.87	30 82

SEm± 6.05 CD (0 05) NS

NS - Non-significance

4 2 2 3 Nitrogen recovery (%)

4 2.2 3.1 Effect of moisture regimes and levels of nitrogen

The flooded rice recorded a significantly higher recovery of N than the non-flooded rice (Table 26). The different N levels also had a significant influence on the recovery of N The highest recovery of N observed at 100 kg N ha^{-1} though was on par with 50 kg N ha^{-1} was significantly superior to that recorded at 150 kg N ha^{-1} Interaction between moisture regimes and levels of nitrogen was not found significant.

4 2.2.3 2 Effect of split application

It was observed that the method of application had a significant influence on the % recovery. The top dressing recorded a significantly higher % recovery than the basal application. The interaction between method of application and moisture regimes was not significant (Table 27). Though non-significant, the top dressed flooded rice recorded a higher % recovery (40%) than the basally applied flooded rice (28%)

The interaction between method of application and levels of nitrogen was also not significant (Table 28). However, 100 kg N ha^{-1} when top dressed recorded the highest % recovery of 48%. The interaction between method of application, level of nitrogen and moisture regimes was also not significant (Table 29).

Table 26 Nitrogen recovery as influenced by the moisture regime and level of nitrogen

Treatment	Nitrogen recovery (%)
Flooded	34.26[a]
Non-flooded	27.68[b]
SEm±	2.06
CD (0.05)	6.15
50 kg N ha^{-1}	33.11[d]
100 kg N ha^{-1}	39.42[d]
150 kg N ha^{-1}	20.38[c]
SEm±	2.53
CD (0.05)	7.53

Values followed by the same alphabet are not significantly different

Table 27. Nitrogen recovery (%) as influenced by the interaction between moisture regime and method of application

Moisture regime	Nitrogen recovery (%)	
	Half basal ^{15}N	Half top-dress ^{15}N
Flooded	28.33	40.18
Non-flooded	26.05	29.30
Mean	27.19^{a}	34.74^{b}

Method of application	SEm±	2.10	CD (0.05)	5.82
Moisture regime x Method of application	SEm±	2.96	CD (0.05)	NS

Table 28. Nitrogen recovery (%) as influenced by the interaction between level of nitrogen and method of application

Level of nitrogen (kg N ha^{-1})	Nitrogen recovery (%)	
	Half basal ^{15}N	Half top-dress ^{15}N
50	32.11	34.11
100	30.69	48.15
150	18.73	21.98
Mean	27.19^{a}	34.74^{b}

Method of application	SEm±	2.10	CD (0.05)	5.82
Level of nitrogen x Method of application	SEm±	3.63	CD (0.05)	NS

NS - Non-significance

Values followed by the same alphabet are not significantly different

Table 29. Nitrogen recovery as influenced by the interaction between moisture regime, level of nitrogen and method of application

Level of nitrogen (kg N ha^{-1})	Nitrogen recovery (%)			
	Flooded		Non-flooded	
	Half basal ^{15}N	Half top-dress ^{15}N	Half basal ^{15}N	Half top-dress ^{15}N
50	28.55	36.45	35.66	31.76
100	34.27	56.59	27.10	33.70
150	22.17	27.51	15.40	16.44

SEm± 5.14 CD (0.05) NS

NS - Non-significance

4.2.2.4 Molecular absorption of urea N (ug pot^{-1})

4.2.2.4.1 Effect of moisture regimes and levels of nitrogen

It was observed that there was many fold increase in the uptake of N as molecular urea by the flooded rice, compared to non-flooded rice (Table 30). Among the different levels of nitrogen, the molecular absorption of urea increased significantly with increase in levels of nitrogen. The interaction effect (Table 31) between moisture regimes and levels of nitrogen was also significant. While there was a steady and significant increase in molecular absorption with increase in the levels of nitrogen by the flooded rice, the different levels of nitrogen had no such effect on non-flooded rice.

4.2.2.4.2 Effect of split application

The method of application had a significant influence on the absorption of nitrogen as molecular (Table 32). Molecular absorption from the top dressed urea was found significantly higher than that from the basal dressed urea. Its interaction with moisture regimes was also found significant (Table 32). The highest molecular urea nitrogen uptake (368 $\mu g\ pot^{-1}$) was recorded by the top dressed flooded rice and the least was exhibited by the basal dressed non-flooded rice (5 $\mu g\ pot^{-1}$).

Table 30. Absorption of molecular urea N as influenced by the moisture regime and level of nitrogen

Treatment	Molecular urea N (μg pot^{-1})
Flooded	581.60^{a}
Non-flooded	13.51^{b}
SEm±	14.40
CD (0.050	42.79
50 kg N ha^{-1}	122.39^{c}
100 kg N ha^{-1}	323.35^{d}
150 kg N ha^{-1}	446.92^{e}
SEm±	17.64
CD (0.05)	52.41

Table 31. Absorption of molecular urea N (μg pot^{-1}) as influenced by the interaction between moisture regime and level of nitrogen

Moisture regime	Level of nitrogen (kg N ha^{-1})		
	50	100	150
Flooded	233.68^{a}	633.18^{b}	877.94^{c}
Non-flooded	11.10^{d}	13.52^{d}	15.90^{d}

SEm± 24.94 CD (0.05) 74.12

Values followed by the same alphabet are not significantly different

Table 32. Absorption of molecular urea N as influenced by the interaction between moisture regime and method of application

Moisture regime	Molecular urea N ($\mu g\ pot^{-1}$)			
	Half basal ^{14}C			Half top-dress ^{14}C
Flooded	213.97^{a}			368.46^{b}
Non-flooded	4.60^{c}			8.91^{c}
Mean	109.29^{d}			188.69^{e}
Method of application	SEm±	7.94	CD (0.05)	22.0
Moisture regime x Method of application	SEm±	11.23	CD (0.05)	31.12

Table 33. Absorption of molecular urea N as influenced by the interaction between level of nitrogen and method of application

Level of nitrogen (kg N ha^{-1})	Molecular urea N ($\mu g\ pot^{-1}$)			
	Half basal ^{14}C			Half top-dress ^{14}C
50	54.80^{a}			$57\ 59^{a}$
100	122.66^{b}			200.68^{c}
150	150.39^{b}			297.78^{d}
Mean	$109\ 29^{e}$			188.69^{f}
Method of application	SEm±	7.94	CD (0.05)	22.0
Level of nitrogen x Method of application	SEm±	13.75	CD (0.05)	38 12

Values followed by the same alphabet are not significantly different

Table 34 Absorption of molecular urea N (µg pot^{-1}) as influenced by the interaction between moisture regime, level of nitrogen and method of application

Level of nitrogen (kg N ha^{-1})	Molecular urea N (ug pot^{-1})			
	Flooded		Non-flooded	
	Half basal ^{14}C	Half top-dress ^{14}C	Half basal ^{14}C	Half top-dress ^{14}C
50	104.81^{a}	128.87^{a}	4.79^{f}	6.31^{f}
100	239.71^{b}	393.47^{c}	5.62^{f}	7 90^{f}
150	297.40^{d}	583.04^{e}	3.38^{f}	12.52^{f}

SEm± 19.44 CD (0.05) 53.90

Values followed by the same alphabet are not significantly different

The interaction effect between the methods of application and the levels of nitrogen was also found significant (Table 33). The highest molecular urea N uptake was observed at 150 kg N ha^{-1}, when applied as a top dressing This was significantly higher than that recorded at all other treatments. The second highest uptake was noticed at 100 g N ha^{-1}, applied as top dressing. At 50 kg N ha^{-1}, the split application did not show any significant influence on the molecular urea N absorption.

The combined effect of method of application, moisture regimes and levels of nitrogen was also found significant (Table 34). The highest uptake of molecular urea N was recorded by the flooded rice top dressed with 150 kg N ha^{-1}. Though both at 150 and 100 kg N ha^{-1} flooded rice registered a significantly higher molecular urea nitrogen uptake from the top dressing, no such significant influence was observed at 50 kg N ha^{-1} Quite unlike the flooded situation, the non-flooded rice did not show any significant difference for the different levels of nitrogen with respect to urea nitrogen uptake

4 2 2 5 Percentage absorption of molecular urea N

4 2.2.5 1 Effect of moisture regimes and levels of nitrogen

It was observed that the flooded rice absorbed a significantly higher % N as molecular urea than the non-flooded rice (Table 35). Among the different levels of nitrogen, the highest

Table 35 Percentage absorption of molecular urea N as influenced by the moisture regime and level of nitrogen

Treatment	Molecular urea N (%)
Flooded	0.73^{a}
Non-flooded	0.02^{b}
SEm±	0.03
CD (0.05)	0.114
50 kg N ha^{-1}	0.32^{c}
100 kg N ha^{-1}	0.29^{c}
150 kg N ha^{-1}	0.52^{d}
SEm±	0.04
CD (0.05)	0.14

Table 36. Percentage absorption of molecular urea N as influenced by the interaction between moisture regime and level of nitrogen

Moisture regime	Level of nitrogen (kg N ha^{-1})		
	50	100	150
Flooded	0.62^{a}	0.56^{a}	1.00^{b}
Non-flooded	0.03^{c}	0.02^{c}	0.03^{c}

SEm± 0.06 CD (0.05) 0.198

Values followed by the same alphabet are not significantly different

% absorption of molecular urea N was noticed with 150 kg N ha^{-1} followed by 50 kg N ha^{-1}, which was on par with that at 100 kg N ha^{-1}. The interaction effect (Table 36) of levels of nitrogen with flooded and non-flooded systems was also significant. Flooded rice with 150 kg N ha^{-1} recorded the highest % absorption of molecular urea N. The percentage absorption of molecular urea by the flooded rice at 50 and 100 kg N ha^{-1} were at par. With increase in levels of nitrogen, there was no significant influence on % absorption of molecular urea N by non-flooded rice.

4.2.2.5.2 Effect of split application

It was observed that the % absorption of molecular urea N from the top dressing of ^{14}C urea was significantly higher than that from basal dressing. It's interaction with moisture regimes was also found significant (Table 37). The highest % uptake was noticed by the top dressed flooded rice and the lease by basally dressed non-flooded rice.

The interaction between the method of application and the levels of nitrogen was also found significant (Table 38) The highest % molecular urea absorption was registered at 150 kg N ha^{-1}, when applied as a top dressing This was significantly higher than that recorded at all other treatments. The % uptake registered at basally applied 150 kg N ha^{-1} was found comparable with that

Table 37. Percentage absorption of molecular urea N as influenced by the interaction between moisture regime and method of application

Moisture regime	Molecular urea N (%)			
	Half basal ^{14}C		Half top dress ^{14}C	
Flooded	0.49^{a}		1.09^{b}	
Non-flooded	0.02^{c}		0.04^{c}	
	0.25^{d}		0.56^{e}	
Method of application	SEm±	0.04	CD (0.05)	0.111
Moisture regime x Method of application	SEm±	0.05	CD (0.05)	0.157

Table 38. Percentage of absorption of molecular urea N as influenced by the interaction between level of nitrogen and method of application

Level of nitrogen (kg N ha^{-1})	Molecular urea N (%)			
	Half basal ^{14}C		Half top dress ^{14}C	
50	0.28^{a}		0.39^{bc}	
100	0.18^{a}		0.51^{b}	
150	0.30^{ac}		0.77^{d}	
	0.25^{e}		0.56^{f}	
Method of application	SEm±	0.04	CD (0.05)	0.114
Level of nitrogen x Method of application	SEm±	0.06	CD (0.05)	0.193

Values followed by the same alphabet are not significantly different

Table 39. Percentage absorption of molecular urea N as influenced by the interaction between moisture regime, level of nitrogen and method of application

Level of nitrogen (kg N ha^{-1})	Molecular urea nitrogen (%)			
	Flooded		Non-flooded	
	Half basal ^{14}C	Half top dress ^{14}C	Half basal ^{14}C	Half top dress ^{14}C
50	0.53	0.75	0.03	0.03
100	0.34	0 09	0.09	0.03
150	0.60	1.52	0.01	0 05

SEm± 0 09 CD (0.05) NS

recorded at 50 kg N ha^{-1} as top dressing The 3 way interaction was found non-signifcant (Table 39).

4.2 2.6 Absorption of N in forms other than molecular urea(mg pot^{-1})

4.2 2.6.1 Effect of moisture regimes and levels of nitrogen

The flooded situation recorded a higher absorption of N in forms other than molecular urea (Table 40). Among the different N levels, 100 kg N ha^{-1} recorded the highest. It was observed that there was a significant decrease in the absorption of nitrogen in forms other than molecular urea as the level of N was increased to 150 kg ha^{-1} from 100 kg ha^{-1}. The interaction effect was not significant.

4 2 2 6 2 Effect of split application

It was observed that absorption of nitrogen in forms other than molecular urea from the basal application was significantly higher than that from the top dressing (Table 41). However, its interaction with moisture regimes was not significant (Table 41). Though non-significant, the basaly dressed flooded rice registered the highest absorption of N in forms other than molecular urea.

The interaction between method of application and nitrogen levels was also not significant (Table 42). The highest uptake in forms other than molecular urea was registered at 100 kg N ha^{-1} when basal dressed Though non significant, at 150 kg N ha^{-1},

Table 40. Absorption of N in forms other than molecular urea as influenced by the moisture regime and level of nitrogen

Treatment	N in forms other than molecular urea (mg pot^{-1})
Flooded	81.87^{a}
Non-flooded	61.77^{b}
SEm±	5.50
CD (0.05)	11.34
50 kg N ha^{-1}	41.26^{c}
100 kg N ha^{-1}	98.23^{d}
150 kg N ha^{-1}	75.96^{e}
SEm±	6.74
CD (0 05)	20.02

Values followed by the same alphabet are not significantly different

Table 41 Absorption of N in forms other than molecular urea, as influenced by the interaction between moisture regime and method of application

Moisture regime	Nitrogen in forms other than molecular urea N (mg pot^{-1})	
	Half basal ^{14}C	Half top dress ^{14}C
Flooded	48.15	38.71
Non-flooded	33.44	28.34
Mean	40.80^a	31.02^b

Method of application	SEm±	2.47	CD (0.05)	6.85
Moisture regime x Method of application	SEm±	3.49	CD (0 05)	NS

NS - Non-significance

Table 42. Absorption of N in forms other than molecular urea, as influenced by the interaction between level of nitrogen and method of application

Level of nitrogen (kg N ha^{-1})	Nitrogen in forms other than molecular urea N (mg pot^{-1})	
	Half basal ^{14}C	Half top dress ^{14}C
50	21.26	20.00
100	60.07	38.16
150	41.05	34.92
Mean	40.80^a	31.02^b

Method of application	SEm+	2 47	CD (0.05)	6.85
Level of nitrogen x Method of application	SEm±	4 28	CD (0.05)	NS

NS - Non-significance

Values followed by the same alphabet are not significantly different

Table 43. Absorption of nitrogen in forms other than molecular urea as influenced by the interaction between moisture regime, level of nitrogen and method of application

Level of nitrogen (kg N ha^{-1})	Nitrogen in forms other than molecular urea N (mg pot^{-1})			
	Flooded		Non-flooded	
	Half basal ^{14}C	Half top dress ^{14}C	Half basal ^{14}C	Half top dress ^{14}C
50	22.68	17 71	19.65	22.29
100	70.50	42.45	49.65	33.86
150	51.29	40 99	30.82	28.86

SEm± 6 05 CD (0.05) NS

NS - Non-significance

the absorption of N in forms other than molecular urea was comparatively lesser than that at 100 kg N ha^{-1} under both the methods of application. The three way interaction effect among the moisture regimes, levels of nitrogen and methods of application was also found non-significant (Table 43).

4 2.2 7 Percentage absorption of N in forms other than molecular urea

4.2 2 7 1 Effect of moisture regimes and levels of nitrogen

The % absorption of N absorbed in forms other than molecular urea was significantly higher in non-flooded situation (Table 44). Among the N levels, 50 kg N ha^{-1} recorded the highest absorption, which was on par with that recorded at 100 kg N ha^{-1}. The interaction was also significant (Table 45). In the flooded situation, the % absorption at 100 kg N ha^{-1} was on par with that at 50 kg and the least % absorption was noticed at 150 kg N ha^{-1}. Under the non-flooded situation there was no significant difference among the N levels

4.2 2.7.2 Effect of split application

The method of application had a significant influence on the % absorption of N in forms other than molecular urea (ie., NH_4^+ and NO_3^- forms) (Table 46) The basal application registered the highest % absorption in other forms. The interaction of moisture regimes and method of application (Table 46) was found significant.

Table 44 Percentage absorption of N in forms other than molecular urea as influenced by the moisture regime and level of nitrogen

Treatment	Nitrogen in forms other than molecular urea (%)
Flooded	99.27^{a}
Non-flooded	99.92^{b}
SEm±	0 04
CD (0 05)	0.137
50 kg N ha^{-1}	99.68^{c}
100 kg N ha^{-1}	99.67^{c}
150 kg N ha^{-1}	99.45^{d}
SEm±	0.05
CD (0.05)	0.168

Table 45. Percentage absorption of N in forms other than molecular urea as influenced by the interaction between moisture regime and level of nitrogen

Moisture regime	Nitrogen in forms other than molecular urea (%)		
	50	100	150
Flooded	99.38^{a}	99.43^{a}	99.00^{b}
Non-flooded	99.97^{c}	99.91^{c}	99.89^{c}

SEm± 0.08 CD (0 05) 0 162

Values followed by the same alphabet are not significantly different

Table 46 Percentage absorption of N in forms other than molecular urea as influenced by the interaction between moisture regime and method of application

Moisture regime	Nitrogen in forms other than molecular urea (%)			
	Half basal ^{14}C		Half top dress ^{14}C	
Flooded	99.50^{a}		98.91^{b}	
Non-flooded	99.98^{c}		99.96^{c}	
Mean	99.75^{d}		99.44^{e}	
Method of application	SEm±	0.04	CD (0.05)	0.111
Moisture regime x Method of application	SEm±	0.05	CD (0.05)	0.15

Table 47. Percentage absorption of N in forms other than molecular urea as influenced by the interaction between level of nitrogen and method of application

Level of nitrogen (kg N ha^{-1})	Nitrogen in forms other than molecular urea (%)			
	Half basal ^{14}C		Half top dress ^{14}C	
50	99.72^{ab}		99.61^{ac}	
100	99.82^{b}		99.49^{c}	
150	99.70^{ab}		99.21^{d}	
Mean	99.75		99.44	
Method of application	SEm±	0.04	CD (0.05)	0.111
Level of nitrogen x Method of application	SEm±	0.06	CD (0.05)	0.193

Values followed by the same alphabet are not significantly differen

Table 48. Percentage absorption of N in forms other than molecular urea as influenced by the interaction between moisture regime, level of nitrogen and method of application

Level of nitrogen (kg N ha^{-1})	Nitrogen in forms other than molecular urea			
	Flooded		Non-flooded	
	Half basal ^{14}C	Half top dress ^{14}C	Half basal ^{14}C	Half top dress ^{14}C
50	99.46	99.25	99.97	99.97
100	99.66	99.01	99.99	99.97
150	99.40	98.48	99.99	99.95

SEm± 0.09 CD (0.05) NS

NS - Non-significance

The highest % absorption was noticed by the basaly applied non-flooded rice and it was at par with that observed at the top-dressed non-flooded situation. The interaction effect of method of application and level of nitrogen was also found significant (Table 47) The least % absorption of nitrogen in other forms was recorded at 150 kg N ha^{-1} when top-dressed. The highest % uptake in other forms was noticed at basaly applied 100 kg N ha^{-1} and it was found on par with that recorded at all other N levels under the same method of application The three way interaction was not significant (Table 48)

4.3 Development of isotope method of urease estimation

The isotope method reported for urease activity determination involves estimation of $^{14}CO_2$ evolved during the hydrolysis of ^{14}C labelled urea. In one of the earlier studies conducted in this lab, this method failed to give comparable urease activity due to the poor tapping of $^{14}CO_2$ Hence it was decided to estimate the urease activity from the residual ^{14}C labelled urea remaining after hydrolysis With this objective, a comparison of this method with the standard non-buffer method was made using six soils. The results of the same are presented in the Table 49. It was observed that while the urease activity by the non-buffer method ranged from 900-1200 μg urea g^{-1} of soil for all the soils, the urease activity by isotope method ranged from 200-400 μg urea g^{-1}

Table 49. Urea hydrolysis as estimated by isotope method and non-buffer method following 5 h incubation

Soil	Replication	Urea hydrolysed (µg g^{-1} soil)			
		Isotope method	Mean	Non-buffer method	Mean
Laterite	1	0	0	903	880.25
	2	0		863	
	3	0		900	
	4			855	
Kole	1	221	231.5	955	961.25
	2	211		955	
	3	244		973	
	4	250		962	
Kari	1	226	265.75	1046	1049.75
	2	302		1046	
	3	277		1083	
	4	258		1024	
Kayal	1	454	432 25	1193	1201.00
	2	431		1230	
	3	416		1193	
	4	428		1188	
Karappadam	1	189	182.5	1266	1240.00
	2	162		1248	
	3	197		1230	
	4	182		1216	
Black cotton	1	290	266.75	1193	1143.50
	2	216		1101	
	3	293		1156	
	4	268		1124	

Table 50. Urea hydrolysis as estimated by isotope method and non-buffer method following prolonged incubation with ^{14}C urea

Incubation (h)	Replication	Urea hydrolysis (µg urea hydrolysed g^{-1} soil)							
		Kari				Black soil			
		Isotope method	Mean	Non-buffer	Mean	Isotope method	Mean	Non-buffer	Mean
24	1	346	374	772	777	741	722	1120	1118
	2	396		790		708		1120	
	3	388		778		726		1[illegible]34	
	4	36[illegible]		768		714		1096	
48	1	392	360	308	808	1111	1060	1486	1445
	2	390		808		1052		1450	
	3	339		812		1049		1423	
	4	320		805		1028		1420	
72	1	496	461	826	830	1610	1603	1780	1773
	2	411		826		1556		1761	
	3	470		840		1670		1796	
	4	468		829		1596		1756	

of soil for different soils. In the laterite soil the isotope method failed to register any urease activity.

It was then decided to prolong the incubation of soil with labelled urea to get a complete hydrolysis of urea, using the soils Kari and Black The data are presented in Table 50. Still the urea hydrolysis as evidenced by the isotope method was much lower than that registered by the non-buffer method.

Discussion

DISCUSSION

5.1 Effect of soil submergence on urea hydrolysis

In this experiment, the effect of soil submergence for different intervals on soil urease activity was studied. The soil was submerged for 0, 5, 10, 20 and 30 days and at the end of each submergence period, incubation with urea was done for 5, 12, 24, 48, 72, 120, 240, 480 and 720 h.

Under non-submergence and for 5 h incubation though the black cotton soil registered the highest urease activity. it did not differ much from the urease activity exhibited by laterite, kole and kayal soils (Table 2 and Fig. 1). The kari soil recorded the least urease activity. This probably was due to the fact that the kari soil was strongly acidic (pH 2.5) and acidity is known to inhibit urea hydrolysis (Bremner and Mulvaney, 1978). When the soil was submerged a decline in the urease activity was noticed in all soils. Though the decline in urease activity was slow upto a submergence period of 10 days, there was almost a 30-40% drop in activity, following submergence period of 20-30 days in all soils except kari soil (Fig 1). The immediate consequence of soil submergence is the rapid depletion of oxygen, and a reduction in the Eh which inturn affects the urease activity According to Pulford and Tabatabai (1988) a strong correlation exists between Eh and urease activity and urease activity was decreased following the decrease in Eh. In the present study, upon submergence of the soil there was a decline in the Eh in all soils except kari (Table 4). This could be the reason for the negligible decrease in urease activity of kari soil upon flooding for as long as 30 days. However, when the submerged soils were incubated with urea for periods longer than 5 hours, complete hydrolysis was noticed within a period of 2-3 days in the case of all soils except kari (Table 3 and Fig 2 to 6) This means that the decline in urease activity following flooding is noticed only initially and has no longstanding effect. In most of the investigations conducted to study the effect of soil submergence, soil urease activity was measured for a 5 h incubation period. From our study it was evident that though the decline in urease activity was there for 5 h incubation period, on prolonged incubation no such decline was observed. Several workers have also reported that the moisture regime do not have much influence on soil urease activity (Skujins and Mc Laren, 1969: Delaune and Patrick, 1970; Gould et al., 1973).

5.2 Influence of moisture regimes and levels of nitrogen on yield dry matter production and nitrogen content

In this section the direct effect of nitrogen and moisture levels regardless of the source and method of application is discussed. It was observed that the yield of grain and straw, total dry weight, N uptake by grain and the total N uptake by the plant were significantly higher in flooded rice than in non-flooded rice (Table 6, 8, 10, 16, 19 and Fig. 7). This favourable effect of flooding may be due to the increased solubilization and availability of nutrients particularly P, K, Ca, Si and Fe of nutrients (De Datta, 1981), better root uptake of nutrients by mass flow and diffusion, better reflectance of the solar radiation to the lower leaves of the rice by the flood water etc Similar favourable effects for flooding have been reported by several workers (Raymond and Shapiro, 1981; Meera, 1986) Though moisture levels did not register any significant effect on grain nitrogen content and N uptake by straw, the straw N content was significantly higher in the non-flooded rice (Table 1[illegible]) This probably was due to the low dry matter accumulation by the non-flooded rice and the N taken up by the plant got accumulated in the limited portion of the vegetative tissues resulting in a high concentration of N. For the same reasons, the straw N uptake by the non-flooded rice though non-significant was slightly higher than that of flooded rice

Among the N levels, though the grain yield at 100 kg N ha^{-1} and 150 kg N ha^{-1} were found at par (Table 6), plant dry weight, straw yield, % of N in grain and straw, the total N uptake by the plant were found significantly higher at 100 kg N ha^{-1} than that at all other levels (Table [illegible], 10, 12, 14 and 19). In several other studies also the optimum dose of N to rice was found to range from 90-110 kg ha^{-1} (Meera, 1986; Surendran, 1985; Vaijayanthi, 1986). In a study conducted by Singh et al (1978), it was observed that for the rice variety Jaya, the optimum level of N was 100 kg N ha^{-1} Application of N at levels higher than 90 kg ha^{-1} was found to induce more vegetative growth and there by lower grain yield, in a study conducted at Agricultural Research Station, Mannuthy (Latir, 1982). The interaction effect between moisture regimes and N levels was also found significant. It was observed that the flooded rice responded better to N application than the non-flooded rice (Table 7) The favourable effect produced on flooding the soil has already been discussed

5.3 Influence of moisture regimes, levels of nitrogen and split method of application on N recovery and forms of N absorption using the alternately labelled urea

5 3.1 Percentage absorption of fertilizer nitrogen and soil nitrogen (% Ndff and % Ndfs)

Though the two different moisture regimes did not show any significant difference with respect to the relative contribution of soil and fertilizer nitrogen, the different N levels had a significant influence (Table 20, 21). When the N levels were increased (from 50-150 kg N ha^{-1}) the contribution from the fertilizer towards total N uptake increased correspondingly from 33 1 to 51 7 % Ndff This would mean that the dependence of plant on native nitrogen decreased considerably (from 66.9 to 48.2%). Azam et al. (1991) reported a contribution of 25.1 - 28.6% by fertilizer N to the total plant nitrogen under flooded field conditions. In the present study, it was found that even at the highest level of 150 kg N ha^{-1} the soil contribution was considerably high (48.2%) Broadbent (1979) reported a range of 50-80% of plant nitrogen, originated from soil. The reason for increased dependence on soil nitrogen as explained by Shiga annd Ventura (1976) is that, fertilizer is a short lived source of N (unless it is conserved through microbial immobilization), whereas the soil supplies N continuously and thus soil N plays a crucial role in the growth and yield of crops.

5.3.2 Fertilizer N uptake

5.3.2.1 Effect of moisture regimes and N levels

The fertilizer N uptake was significantly higher by the flooded rice than by the non-flooded rice (Table 22 and Fig. 8). The fertilizer N uptake is computed from % Ndff and total N uptake. As the N uptake was much higher in the flooded rice (Table 19) the same trend was obtained with respect to fertilizer N uptake also

Though the % Ndff was higher at 150 kg N ha^{-1}, the highest fertilizer N uptake was recorded at 100 kg N ha^{-1}. As in the case of the effect of moisture regimes the highest total N uptake was recorded at 100 kg N ha^{-1} (Table 19) which in turn led to a high fertilizer N uptake. The interaction effect between moisture regimes and N levels were found non-significant.

5 3.2.2 Effect of split application

As in the case of moisture regimes and N levels, the method of application exhibited a significant influence on fertilizer N uptake. The fertilizer N uptake was significantly higher from the top dressing than from the basal dressing (Table 23). This is mainly due to the fact that the basal dressing was done to the rice at its seedling stage when the root system was not well established. At the time of the top dressing rice plants would be having a well developed root system, which could be the reason for the higher fertilizer N uptake. The interaction of method of application with moisture regimes and levels of nitrogen was found non-significant (Tables 23, 24).

5.3.3 Nitrogen recovery (%)

5 3 3 1 Effect of moisture regimes and N levels

The flooded rice recorded a significantly higher recovery of nitrogen (34%) than the non-flooded rice (28%) (Table 26). It is but natural that the flooded rice recorded a higher % of N recovery as the flooded rice had recorded a significantly higher fertilizer N uptake and dry matter production (Tables 10, 22). However, many studies conducted on the losses of nitrogen and its recovery by rice plant have indicated that both denitrification and ammonia volatilization are substantial from flooded rice soils (De Datta, 1981) Inspite of this fact in the present study N recovery is much higher from the flooded rice than from the non-flooded rice. This indicates that the denitrification and volatilization losses under the upland conditions (non-flooded) are comparable with that under flooded conditions. There is a slight possibility for a higher volatilization loss of ammonia from the non-flooded soil if the NH_3 loss is in any way linked with the evaporation loss of moisture. However, in one of the earlier studies conducted by Chao and Kroontje (1964) it was observed that the rate of ammonia volatilization and of water vapour from some soils followed different functions. As already mentioned, the main reason for better recovery of applied N by the flooded rice is its robust growth noticed throughout the various growth stages.

Among the N levels, the highest recovery was noticed at 100 kg N ha^{-1} and there is a considerable reduction in the recovery when the N level was increased to 150 kg N ha^{-1} (Table 26). It has already been observed in the present study that the rice growth and yield is at its best at 100 kg N ha^{-1}. Also it is at this level that the highest fertilizer N uptake was noticed. However, at the highest level of nitrogen ie., 150 kg ha^{-1}, a significant decrease in the N recovery was noticed. Similar decrease in N recovery following increase in N levels had been reported by Rajale and Prasad (1973) also.

5 3 3.2 Effect of split application

The recovery of the applied N from the top dressing was significantly higher than that from basal dressing (Table 27). In this experiment, the top dressing was done at the early panicle initiation stage. One of the main reasons for high recovery at this stage is the better developed root system, especially the superficial secondary root system. Another reason probably is the high N requirement by the rice plant at this stage In a study conducted by Yanagisawa et al (1967) in Japan, it was observed that as much as 55% of the N applied 15 days before heading was used by the plant and only 7%, when N was applied as basal dressing. In studies conducted at IRRI also the highest efficiency of N use for a medium duration rice variety was observed when fertilizer N was applied, at half of the optimum rate (60 kg N ha^{-1}), just before panicle initiation (De Datta et al., 1969). In the present study the interaction of method of application with moisture regimes and N levels was not found significant (Tables 27, 28).

5.3.4 Molecular absorption of urea

5 3 4 1 Effect of moisture regimes and N levels

The flooded rice recorded a significantly higher molecular absorption of urea than the non-flooded rice as evidenced from the urea N uptake (Table 30) and % absorption of molecular urea N (Table 35). With respect to the % absorption of N in forms other than molecular urea, the non-flooded rice had recorded a higher value than the flooded rice (Table 44). The higher absorption of molecular urea under the flooded condition could be due to the decline in the urease activity upon flooding the soil. There are divergent findings on the effect of water level on the urease activity. While some workers have reported that urease activity is unaffected by water level (Skujins and Mc Laren, 1969, Delaune and Patrick, 1970, Gould _et al._, 1973). Several other workers have reported a decrease in urease activity upon flooding the soil. Pulford and Tabatabai (1988) observed that urease activity decreased in water logged soil and was significantly correlated with Eh. Saraswathi _et al._ (1991) in their study on the effect of moisture regimes on urease activity observed a complete cessation of urea hydrolysis following flooding. The results obtained in this investigation (Table 2) also indicate that there is atleast a decline in urea hydrolysis in flooded soils which must have resulted in the molecular absorption of urea. The fact that rice plants are capable of absorbing N in the molecular form has been well established (Mitsui and Kurihara, 1962, Saraswathi _et al._, 1991). According to Mitsui and Kurihara, urea taken up through the rice roots is converted into ammonium carbonate by urease at a slower rate than it is absorbed. This may result in the accumulation of urea in the rice roots prohibiting further urea absorption.

It is assumed that the quantity of the radiolabel recovered in the plant is entirely due to the absorption of urea in the molecular form The reactions involved in the hydrolysis of applied urea in the soil lead to the evolution of CO_2 as an end product besides the ammonium. When ^{14}C urea is applied to the soil, it is therefore likely that $^{14}CO_2$ will be evolved which can be absorbed by the foliage or can be converted into other forms in the soil which may be absorbed by the plant. Since it has already been shown that the rice plant can absorb molecular urea through roots (Saraswathi et al., 1991) and that the quantity of N so absorbed is very little (Tables 35, 36, 37, 38 and 39), it may be safely concluded that the quantity of ^{14}C label observed in the plant in the present study represents absorption of molecular urea. The molecular absorption of urea has also been reported in wheat by Bradley et al. (1989). In this study they have found that the capacity of the plant for the absorption of molecular urea was substantially less than the ammonium uptake

Among the N levels, the highest absorption of urea was observed at 150 kg N ha^{-1} (Table 30), despite the fact that the N uptake and recovery was maximum at 100 kg N ha^{-1} This is mainly due to the fact that at 150 kg N ha^{-1}, higher amount of urea was present in the soil. As the rate of hydrolysis is slow in the flooded soil, the amount of unhydrolysed urea present would be naturally more at 150 kg N ha^{-1} Because of the higher substrate concentration, there was naturally a higher molecular absorption This is further evident from the fact that the interaction between moisture regime and level of nitrogen was significant (Table 31) and the highest molecular absorption was noticed by the flooded rice supplied with 150 kg N ha^{-1}

5 3.4.2 Effect of split application

The molecular absorption of urea was significantly higher from the top dressing than from the basal dressing. While the basal dressing was done two weeks after sowing, the top dressing was done just before panicle initiation. Not only molecular absorption, but the nitrogen recovery as such was higher from the top dressing and the reasons for the same have already been discussed. From the interaction (Table 32) it could be seen that the highest molecular absorption was by the flooded rice from the top dressing. At the time of top dressing the flooded rice was having a standing water level of 4-5 cm continuously from the seedling stage Hence there is a possibility for more urea to remain unhydrolysed in this condition when applied at the rate of 150 kg N ha^{-1}.

5.4 Development of isotope method for urease estimation

In this study, an attempt was made to develop an isotope method for urease estimation from the residual ^{14}C labelled urea remaining after hydrolysis. This method had the advantage that it was less combursome and do not involve the development of colour The standard non-buffer method involves the development of colour which is rather unstable and depended on the quality of the reagents used (Mulvaney and Bremner, 1979). In this respect, the isotope method had a distinct advantage over the non-buffer method However, in the present study, the isotope method resulted in a gross underestimation of urease activity While the urease activity by the non-buffer method ranged from 900-1200 μg urea g^{-1} soil, that by the isotope method ranged from 200-400 μg urea g^{-1} soil (Table 49). This could be due to the isotope effect in the sense that the urease shows a discrimination between labelled and unlabelled urea. This discrimination was seen even when the incubation with urea was prolonged upto 3 days (Table 50) Such a discrimination was also noticed by Rabinowitz _et_ _al_. (1956) who found that jackbean urease hydrolysed ^{12}C urea about 10% faster than ^{14}C urea

Conclusion

The present investigation included 3 sets of experiments intended to study the effect of soil submergence on soil urease activity, to develop an isotope method for urease estimation and to know the molecular absorption of urea by the flooded rice. The effect of soil submergence was pronounced only during the initial stages of incubation with urea. Upon prolonged incubation with urea, soil submergence had not much effect. With respect to the isotope method, it was observed that it resulted in a gross under estimation of urease activity due to the isotope effect.

The results of the pot culture experiment to study the molecular absorption of urea clearly indicated that the molecular absorption does take place and it is much higher from the flooded soil than from the non-flooded soil. However, there exists a possibility for an underestimation of the quantity of urea absorbed due to the possible loss of $^{14}CO_2$ though respiration. However the present study does not throw any light on the mode of absorption of urea and its fate inside the plant.

Regarding the mode of absorption, detailed studies have been conducted using several algae. Healey (1977) studied the uptake of urea and ammonium by the green alga Scenedesmus quadricauda and blue green alga Pseudoanabaena catenata He observed that urea and ammonium uptake resemble each other and the uptake of urea was depressed by ammonium. These results suggest that both uptake reactions might occur at the same site However, it was observed that ammonium and urea uptake by both algae have different pH dependences. Also in a variety of experiments with the algae the ratio of ammonium uptake to urea uptake did not vary systematically with the ratio of the external concentration of ammonium and urea indicating that ammonium and urea did not seem to compete for the same site. In summary these results suggest that ammonium and urea uptake occur at different sites. However, the fact that ammonium and urea uptake mutually depend on one another shows that these sites are not entirely independent. Presumably some internal mechanisms governs the rate of uptake at one site, depending on what is occuring on the other site. Another strong theory for the urea uptake is that it is a carrier-mediated process and the amino acid arginine and the urea are taken up by the same carrier (William and Hodson, 1977). However, in a detailed study using multicellular eukaryote Volvox carteri F nagariensis. Kirk and Kirk (1978) observed that arginine and urea carriers are distinct and different. According to Mitsui and Kurihara (1962) the absorbed urea initially get accumulated in the root and then is translocated into the grain part with the transportation stream and is decomposed more rapidly in the shoot than in the root as the urease activity is greater in the shoot With respect to the fate of the urea in the plants detailed studies have been conducted following foliar application of urea. Dilley and Walker (1961) observed that apple and peach leaves supplied with ^{14}C labelled urea assimilated this in the amino acids, amides, proteins and other soluble compounds within 20 hours Only small portion of the urea absorbed by the peach remained in the leaves as unhydrolysed urea

Summary

SUMMARY

An investigation was conducted at the Radio Tracer Laboratory, College of Horticulture, Vellanikkara during the period 1990-92 to study the molecular absorption of urea by flooded rice. The following experiments were undertaken during the course of this investigation

1 Effect of soil submergence on urea hydrolysis
2. Pot culture experiment to know the absorption of molecular urea and other forms of nitrogen by flooded and non-flooded rice from split doses of nitrogen
3 Development of isotope method for urease estimation

The results of the investigation are summarised below.

Under non-submergence and for 5 h incubation though the black cotton soil registered the highest soil urease activity, it did not differ much from the urease activity exhibited by laterite, kole and kayal soils. However, the kari soil recorded the least urease activity When the soil was submerged, a decline in the urease activity was noticed in all soils. Though the decline in urease activity was slow up to a submergence period of 10 days, there was almost a 30-40% drop in activity following submergence period of 20-30 days in all soils, except kari soil. When the submerged soils were incubated with urea for periods longer than

5 hours, a complete hydrolysis was noticed within a period of 2-3 days in the case of all soils except kari

In the pot culture experiment, it was observed that flooded rice recorded significantly higher yield of grain and straw, total dry weight and N uptake than the non-flooded rice. Among the N levels (disregarding the source and method of application) though the grain yield at 100 kg N ha^{-1} and 150 kg N ha^{-1} were found at par, plant dry weight, straw yield, % of N in grain and straw, the total N uptake by the plant were found significantly higher at 100 kg N ha^{-1} than at all other levels of N.

The % Ndff increased with increasing levels of nitrogen application. A reverse trend was observed in the case of % Ndfs. The moisture regimes and its interaction with nitrogen levels had no significant influence on % Ndff and % Ndfs.

Flooded rice recorded a significantly higher N uptake than non-flooded rice and among the N levels, the highest fertilizer N uptake was noticed at 100 kg N ha^{-1} and the least at 50 kg N ha^{-1} The interaction effect was found non-significant. The fertilizer nitrogen uptake from the top dressing was significantly higher than that from the basal dressing. The interaction of method of application with moisture regimes, levels of nitrogen and with both (three way interaction) were not significant

The % recovery of fertilizer N by the flooded rice was significantly superior to that by non-flooded rice The highest recovery was noticed at 100 kg N ha^{-1}. The interaction of moisture regimes and levels of nitrogen was not significant The top dressing recorded a higher % recovery than basal dressing The interaction of method of application with moisture regimes levels of nitrogen and the three way interactions were not significant.

There was many fold increase in the uptake of N as molecular urea by the flooded rice compared to non-flooded rice Among the different levels of nitrogen, the molecular absorption of urea increased significantly with increase in levels of nitrogen. The interaction effect between moisture regimes and levels of nitrogen was also found significant. Under the flooded situation, while there was a steady and significant increase in the molecular absorption of urea with increase in the levels of nitrogen, under the non-flooded situation, the different levels of nitrogen had no effect. With respect to the percentage absorption of N in forms other than molecular urea. the non-flooded rice had recorded a higher value than the flooded rice. The molecular absorption of urea was significantly higher from the top dressing than from the basal dressing. The highest molecular absorption was recorded by the flooded rice from the top dressing.

The isotope method of urease estimation was found not comparable with the non-buffer method as the isotope method recorded very low values of urease activity. While the urease activity by the non-buffer method ranged from 900-1200 µg urea g^{-1} soil for all the soils, the urease activity by isotope method ranged from 200-400 µg urea g^{-1} of soil

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* Original not seen

Appendices

APPENDIX-1

Weather data (weekly average) for the cropping period
(from 9-8-1991 to 8-12-1991)

Standard Week No	Month and date	Total rainfall (mm)	Temperature (°C)		Relative humidity (%)		Sunshine hours
			Maximum	Minimum	Forenoon	Afternoon	
32	Aug 6-12	65.2	29.5	23.1	95	79	2.3
33	Aug 13-19	13.9	27.8	22.0	95	84	1.8
34	Aug 20-26	53.0	29.1	22.3	96	79	3.5
35	Aug 27-Sept 2	12.5	30.4	23.3	94	66	6.5
36	Sept 3-9	6.0	31.4	23.2	90	59	9.1
37	Sept 10-16	6.4	31.6	24.6	90	65	6.1
38	Sept 17-23	18.3	31.5	22.4	92	59	6.8
39	Sept 24-30	36.8	31.7	24.0	90	70	6.5
40	Oct 1-7	11.4	31.2	23.5	92	77	4.1
41	Oct 8-14	97.3	30.6	23.0	91	75	4.3
42	Oct 15-21	57.6	30.8	23.1	87	66	6.4
43	Oct 22-28	40.0	29.8	23.0	87	72	[illegible]
44	Oct 29-Nov 4	75.4	32.1	22.5	96	76	3.0
45	Nov 5-11	105.0	31.4	22.8	89	62	7.4
46	Nov 12-18	53.4	31.0	20.9	[illegible]	69	5.0
47	Nov. 19-25	0.5	31.3	21.4	76	58	7.7
48	Nov. 26-Dec 2	0.0	30.9	23.5	78	45	8.6
49	Dec 3-9	0.0	31.9	23.2	69	56	9.7

Source Meteorological Observatory, Vellanikkara

Appendix 2 Abstract of Anova

Effect of moisture regime and level of nitrogen on yield, dry matter production and nitrogen content

Source	df	Mean square 1 Grain yield (g pot^{-1})	2 Straw yield (g pot^{-1})	3 Total dry weight (g pot^{-1})	4 N in grain (%)	5 N in straw (%)	6 N uptake by grain (mg pot^{-1})	7 N uptake by straw (mg pot^{-1})	8 N uptake by plant (mg pot^{-1})
Moisture regime	1	292 03**	18 98**	375.44**	0 262^{NS}	1.99**	49482 9**	2931 3^{NS}	28162.3**
Level of nitrogen	3	38.95**	78 63**	196.55**	3 71**	3.22**	14473.9**	34891.5**	94506 4**
Interaction	3	17.69**	.3**	23 44**	0 74**	0 95*	4109 7**	2570 1^{NS}	3814 7**
Error	56	0 75	1 24	4 89	0 159	0 23	409.3	1310.53	2126 8

** Significant at 1%

* Significant at 5%

NS - Non-significance

Appendix 3. Abstract of Anova

Effect of moisture regime and level of nitrogen on fertilizer and soil N uptake, N recovery and forms of absorption of nitrogen

Source	df	Mean square 1	2	3	4	5	6	7	9
		% Ndff	% Ndfs	Fert N uptake	% recovery	Molecular urea N uptake (ug plot^{-1})	%^{14}C urea N uptake	N absorbed in forms other than molecular urea (mg pot^{-1})	% absorption of N in forms other molecular urea
Moisture regime	1	1.494^{NS}	1.495^{NS}	2561*	259.8*	1936380.3**	2.98**	2425.4*	2.55**
Level of nitrogen	2	707.56**	707.6**	6647.5**	752.4**	214628.2**	0.119**	6592.4**	0.135*
Interaction	2	8.13^{NS}	8.13^{NS}	743.8^{NS}	95.9^{NS}	208447.2**	0.11**	721.[illegible]^{NS}	0.097*
Error	18	10.47	10.47	364.4	51.4	2489.7	0.02	363.5	0.026

** Significant at 1%
* Significant at 5%
NS -- Non-significance

Appendix 4. Abstract of Anova

Effect of moisture regimes, levels of nitrogen and split method of application on fertilizer N uptake, N recovery and forms of absorption of nitrogen

Source	df	Mean square 1 Fertlizer N uptake	2 Recovery N (%)	3 Molecular urea N (ug pot^{-1})	4 Urea N uptake (%)	5 N absorbed in forms other than molecular urea (ug pot^{-1})	6 Absorption of N in forms other than molecular urea (%)
Moisture regime	1	1275 87**	519 54*	971032**	6.99**	1211**	6 95**
Levels of nitrogen	2	3322.04**	1504.40**	108063**	0.23**	6593**	0.23**
Moisture regimes x Levels of nitrogen	2	370.30^{NS}	191 90^{NS}	104960**	0.21**	720^{NS}	0.21**
Method of application	1	1122 39**	684.30*	75653**	1 15**	1145**	1.14**
Moisture regime x Method of application	1	250.94^{NS}	222.20^{NS}	67662**	0.98**	261^{NS}	0 98**
Levels of nitrogen x Method of application	2	465.87^{NS}	295.95^{NS}	18122**	0.14*	932^{NS}	0 14*
Moisture regime x Level of nitrogen x Method of application	2	6.71^{NS}	15.02^{NS}	16126**	0.11^{NS}	13^{NS}	0.11^{NS}
Error	36	146.7	105.8	1513	0.04	5278	0.04

** Significant at 1%   * Significant at 5%

NS - Non-significance

Plates

Plate 1a Rice variet, Java under flooded and non flooded conditions, supplied with 50 kg N ha^{-1}


VARIETY JAYA
75 DAS
FLJ
50 Kg / ha
V

Plate II. Rice variety Jaya under flooded and non-flooded conditions, supplied with 100 kg N ha^{-1}


VAR
YA
FLOODED
100 Kg/ha

Plate 1c. Rice variety Jaya under flooded and non-flooded conditions, supplied with 150 kg N ha^{-1}

A
Kg ha
VAR 1
N

Plate 1d Rice variety Jaya under flooded condition supplied with 50, 100 and 150 kg N ha^{-1}

Plate 1e. Rice variety Jaya under non-flooded condition supplied with 50, 100 and 150 kg N ha^{-1}


VARIETY JAYA
75 DAS
NON FLOODED
50 Kg N ha⁻¹
VARIETY JAYA
75 DAS
NON FLOODED
100 Kg N ha⁻¹
VARIETY JAYA
75 DAS
NON FLOODED
150 Kg N ha⁻¹

MOLECULAR ABSORPTION OF UREA BY FLOODED RICE

By

SAFEENA A. N.

ABSTRACT OF A THESIS

Submitted in partial fulfilment of the
requirement for the degree of

Master of Science in Agriculture

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ABSTRACT

An investigation on molecular absorption of urea by flooded rice was conducted at the Radio Tracer Laboratory, College of Horticulture, Vellanikkara, during the period 1990-1992. The effect of soil submergence on soil urease activity was also studied on five different soils of Kerala namely, laterite, kole, kari, kayal and black soils An attempt was also made to develop an isotope method for urease estimation using ^{14}C labelled urea. From the specific activity of ^{14}C urea solution initially added and the count rates obtained for the KCl-PMA extract, the urea hydrolysis rate was calculated.

To study the molecular absorption of urea, a pot culture experiment was done employing an alternate stable isotope radio-labelling technique. In this experiment rice was grown in pots under flooded and non-flooded conditions and supplied with ^{14}C and ^{15}N labelled urea basaly and or as top-dress alternately. This study clearly revealed that absorption of nitrogen as molecular urea does takeplace and it is much higher from the flooded rice than from the non-flooded rice. Also, the molecular absorption was higher from the top-dressing than from the basal dressing. With respect to the influence of soil submergence on urease activity it was observed that a slight decline in activity occured following flooding.

The isotope method of urease estimation did not yield values comparable with the non-buffer method. The isotope method resulted in an under estimation of urease activity probably due to the isotope effect