

TRANSFORMATIONS OF APPLIED UREA IN SOILS OF TEA GROWING AREAS AS AFFECTED BY AN UREASE INHIBITOR (AGROTAIN)

P.L.K. Tennakoon, L.S.K. Hettiarachchi, A.K.N Zoysa, and G.P. Gunaratne
(Tea Research Institute of Sri Lanka,, Talawakelle, Sri Lanka)

Ammonia volatilization from urea is an important mechanism of nitrogen loss that may reduce nitrogen fertilizer use efficiency. It has been reported that urease inhibitors reduce the rate of hydrolyses of urea and thereby helps efficient utilization of applied nitrogen fertilizer. However, very little published information is available on the response of urea in the presence of urease inhibitor in soils of tea growing areas of Sri Lanka. Therefore, a glasshouse study was conducted to evaluate the effect of an urease inhibitor (Agrotain) on the changes of applied urea fertilizer in three great soil groups (RYP, RBL, and IBL) over a period of 06 weeks. Urea at 03 levels of N (50, 75 and 100 kg N ha⁻¹) with and without Agrotain, were used as treatments in a randomized complete block design. Altogether, there were 18 treatments with 03 replicates. The treated soils were incubated under glasshouse conditions at 60% field capacity.

During experimentation, periodic changes in soil available NH₄⁺-N, NO₃⁻-N, pH (water and 0.01M CaCl₂) and volatilized ammonia were measured. Significant differences were observed in plant available NH₄⁺-N and NO₃⁻-N concentrations due to differences in soil great groups. The application of Agrotain did not show any effect on NH₄⁺-N concentration in the soil. However, Agrotain had a significant effect on NO₃⁻-N concentration in the soil till the end of second week. Agrotain did not show any significant effect on reducing volatilization losses of ammonia.

In all three great soil groups, there was a pH decrease at the end of experimentation. The higher activity of nitrification is confirmed by the consequent reduction in soil pH. There were significant differences among soil pH s in the soil great groups and it was due to nitrification of NH₄⁺-N in soil. The level of nitrogen application had a significant effect on soil pH but only until the second week from treatment application. Thereafter, the level of nitrogen showed no effect on soil pH.

INTRODUCTION

Nitrogen is the most important fertilizer nutrient for tea. Tender vegetative shoots, which constitute the harvestable yield of tea, contain as much as 3-5 percent nitrogen. To sustain soil fertility and obtain higher yields, currently 270 to 400 kg N ha⁻¹ yr⁻¹ is recommended for clonal vegetatively propagated tea. At present Tea Research Institute (TRI) recommends urea as the sole source of nitrogen for mature tea, because its application gives rise to same yield of tea when compared to sulphate of ammonia. Besides, it acidifies the soil to a lesser extent than sulphate of ammonia and is cheaper per unit of nitrogen as well (Wickremasinghe, 1986). However, practically nitrogen has been applied to tea in the form of both urea and / or sulphate of ammonia.

The fate of urea applied to the soils in tea growing areas was studied extensively by several workers (Bhavananthan, 1970; Bhavananthan and Fernando, 1970; Sivasubramaniam, 1980; and Wickremasinghe *et al.*, 1981). It releases less bases into the soil solution from cation exchange sites when compared to sulphate of ammonia (Wickremasinghe *et al.*, 1985). Urea when applied to soil gets hydrolyzed to the unstable compound ammonium carbamate, which is then immediately converted to ammonium carbonate by urease activity. The production of ammonium carbonate, on hydrolysis causes a temporary increase in the soil pH, when it exceeds seven, may cause the release of one molecule of ammonia from ammonium carbonate along with the formation of the more stable ammonium bicarbonate. This loss of ammonia, results in loss of nitrogen and reduces fertilizer use efficiency (Bhavananthan, 1970).

It has been reported that Agrotain ([N – (n-butyl) Thiophosphoric Triamide]), a new generation nitrogen fertilizer additive, inhibits the enzymatic degradation of urea by free urease, an enzyme produced by soil microorganisms which lives in crop residues and soil organic matter. It works by inactivating the urease enzyme and does not affect soil microorganisms, earthworms or other soil organisms (Anon, 1997). Thereby, Agrotain inhibits the surface applied urea and urea containing fertilizer from the rate of breakdown and reduces loss through volatilization. However, very little published information is available on the response of urea in the presence of urease inhibitors in soils of tea growing areas in Sri Lanka. Therefore, objective of this preliminary study was to investigate the role of urease inhibitor (Agrotain) on the transformations of applied urea to soils of three great groups of tea growing areas, adopting a glasshouse technique.

MATERIALS AND METHODS

Soils and Locations

Soils used for this study were Red Yellow Podzolic (RYP), Reddish Brown Latasolic (RBL) and Immature Brown Loam (IBL) which belongs to Ultisols, Oxisols and Inceptisols, respectively according to USDA soil taxonomy (De Alwis and Panabokke, 1972).

Composite samples of RBL and IBL soils were collected from Wathtegama area in Kandy district (472 m AMSL) and RYP were collected from St. Coombs estate, Talawakelle (1382 m AMSL). The experiment was conducted in the greenhouse of Tea Research Institute (TRI) of Sri Lanka, Talawakelle.

Treatments and Methodology

After removing the surface litter, the soils were collected from a depth of 0-15 cm, air dried, passed through a 2 mm sieve and used for the study. Plastic pots (of 20 cm diameter) were filled with 4 kg of soil and urea as well as Agrotain were applied by broadcasting on the soil surface. Urea at three levels of nitrogen (50, 75 and 100 kg of N ha⁻¹) with and without Agrotain, were used as treatments in a randomized complete block design. Altogether there were 18 treatments (3 soils x 3 N levels x with and

without Agrotain) with three replicates. The pots were covered with polyethylene bags and kept under glasshouse conditions at 60% field capacity.

A method described by Fenn and Kissel (1973) was used to estimate the volatilized ammonia. A plastic vial (25ml) containing 5 ml of 0.5 N H_2SO_4 solution and 2 drops of methyl red indicator was placed (to absorb volatilized ammonia) inside the bag. At each sampling time (initially at 04-day intervals and later at 07-day intervals), the acid solution inside the vial was transferred to a conical flask and then titrated against 0.01N NaOH solution until the colour changes from pink to yellow. The amount of nitrogen released from the applied urea fertilizer was calculated. Representative random samples of fresh soils were also drawn from the pots at the same time of sampling, using a cork bore (2 cm diameter). These were then used for determining pH, available ammonium and nitrate nitrogen.

pH in soil was measured at each sampling, using water and 0.01M CaCl_2 solution (1:2.5). Soil available NH_4^+ -N and NO_3^- -N contents were determined by Kjeldahl method (Hesse, 1971).

Borax (pH 1.5) extractable phosphorus was determined by Vandamolybdate method (Jackson, 1958), 1N NH_4Cl (pH 7) extractable, exchangeable potassium and magnesium were determined by Flame Photometer and Atomic Absorption Spectrophotometer, respectively. Organic Carbon in soil was determined by Walkley Black method and Cation Exchange Capacity (CEC) by leaching with 1 N NH_4Cl , 1N Na_2SO_4 and ammonia distillation with NaOH.

RESULTS AND DISCUSSION

Some of the important properties of the soils used are presented in Table 1.

Table 1 - Some of the important properties of the soils used

Character	Unit	RYP	RBL	IBL
pH(water)	1: 2.5, soil : water	4.75	6.53	6.39
pH(0.01M CaCl_2)	1 : 2.5, soil : suspension	4.11	5.73	5.36
Total Phosphorus	mg^{-1} kg soil	8.14	16.4	106
Ex. Potassium	mg^{-1} kg soil	175	140	45.0
Ex. Magnesium	mg^{-1} kg soil	43.2	54.1	17.2
Ava. NH_4^+ - N	mg^{-1} kg soil	50.4	92.4	44.8
Ava. NO_3^- - N	mg^{-1} kg soil	50.4	42.0	36.4
CEC	meq per 100g soil	15.2	15.9	15.0
O.C	%	3.29	3.56	2.70

In RYP great soil group pH levels were comparatively lower than the other two great soil groups. Except for pH and total phosphorus, all the other properties were somewhat lower in IBL great soil group than the RYP and RBL great soil groups. According to Lamb (1953), fertility status of a soil may influence nitrification.

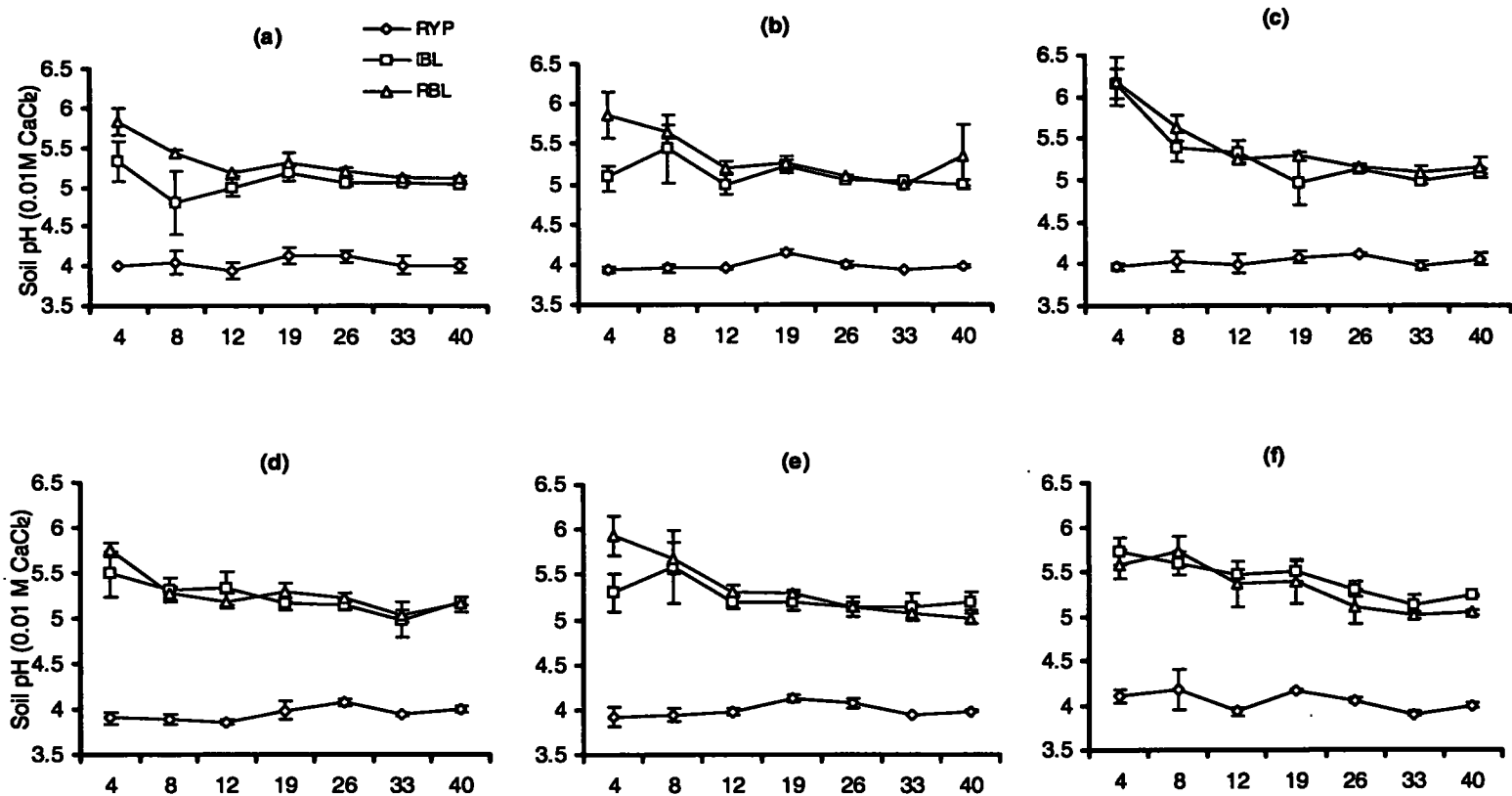


Fig 1: Effect of soil group on soil pH at increasing rates of N (50, 75 and 100 kg ha⁻¹) with Agrotain (a,b and c respectively) and without Agrotain (d, e and f respectively). Vertical bars represent standard errors of the means.

Changes in soil pH

Table 2 - Levels of significance for soil pH (0.01 M CaCl₂) due to soil group (SG), nitrogen levels (LN), and Agrotain levels (AG)

Source	Intervals (days)						
	4	8	12	19	26	33	40
SG	***	***	***	***	***	***	***
LN	**	**	**	NS	NS	NS	NS
AG	NS	NS	*	NS	NS	NS	NS
SG*LN	**	NS	*	NS	NS	NS	NS
SG*AG	NS	NS	*	NS	NS	NS	NS
LN*AG	NS	NS	NS	**	NS	NS	NS
SG*LN*AG	NS	NS	NS	NS	NS	NS	NS

* P ≤ 0.05, ** P ≤ 0.01, *** P ≤ 0.001, NS – Not Significant

Significant ($P \leq 0.001$) differences could be observed in soil pH levels among the soil great groups (Table 2). Any nitrogen fertilizer (Urea or $(\text{NH}_4)_2\text{SO}_4$) when nitrified in the soil by nitrifying bacteria, releases hydrogen ions to the soil solution and this acidifies the soil. Sahrawat (1982) and Wickremasinghe *et al.* (1985) reported that different soil types differ in their ability to nitrify ammonium even under similar conditions of temperature, moisture, and level of added ammonium. Since nitrification is a microbiological process, one factor that may be responsible is the variation in the population of nitrifying organisms present in different soils (Tisdale *et al.*, 1985). Due to such differences in the nitrification of NH_4^+ - N in soil, significant differences can be observed in soil pH levels among the soil great groups (Table 2). Soil pH is gradually decreasing as the experiment progress (Fig. 1). However, in the RYP soil great group pH levels were fairly constant with time and low compared to other two soil great groups. The resistance of the soil pH to change gives rise to the buffering phenomenon in soils (Foth, 1972). Therefore this may be due to differences in soils buffering capacities. RYP may be having higher buffering capacity than other two soil great groups (Sandanam *et al.*, 1978), although no marked differences were found between RYP and RBL great group.

The level of nitrogen application had a significant ($P \leq 0.01$) effect on soil pH but only up to the 12th day following the treatment application (Table 2). Thereafter, the level of nitrogen showed no significant effect on soil pH. At higher rates of nitrogen application, pH levels were higher compared to lower levels of nitrogen application. Wickremasinghe *et al.* (1985) studied transformation of nitrogen fertilizers in soil and found that when urea is applied to soil it is firstly hydrolyzed within about seven days, in the presence of enzyme urease, which is then followed by the nitrification. The

production of ammonium carbonate on hydrolysis causes a temporary increase in the soil pH (Bhavanathan, 1970). Bremner and Mulvaney (1978) reported that the rate of urea hydrolysis increased as a first order reaction with respect to the amount of urea applied to the surface. This would have increased the soil pH levels during the initial period with respect to higher rates of urea added. Thereafter, although it was not significant soil pH levels have decreased with respect to higher rates of urea added, due to release of protons/H⁺ through nitrification reaction as previously described (Fig. 1). Application of Agrotain did not cause any significant impact on soil pH levels (Table 2).

Changes in exchangeable NH₄⁺ - N and NO₃⁻ - N in soil

Table 3 - Levels of significance for soil available NH₄⁺ -N due to soil groups (SG), nitrogen levels (LN), and Agrotain levels (AG)

Source	Intervals (days)						
	4	8	12	19	26	33	40
SG	***	***	***	***	NS	*	***
LN	NS	**	NS	*	NS	NS	*
AG	NS	NS	NS	NS	NS	NS	NS
SG*LN	NS	NS	NS	NS	**	NS	*
SG*AG	*	NS	NS	NS	NS	NS	NS
LN*AG	NS	*	NS	NS	*	NS	*
SG*LN*AG	NS	NS	NS	NS	NS	NS	NS

* P ≤ 0.05, ** P ≤ 0.01, *** P ≤ 0.001, NS – Not Significant

Table 4 - Levels of significance for soil available NO₃⁻ -N due to soil groups (SG), nitrogen levels (LN), and Agrotain levels (AG)

Source	Intervals (days)						
	4	8	12	19	26	33	40
SG	NS	NS	***	***	***	***	***
LN	NS	NS	NS	NS	NS	NS	NS
AG	***	**	NS	NS	NS	NS	NS
SG*LN	*	*	NS	NS	NS	NS	NS
SG*AG	NS	NS	NS	NS	*	NS	***
LN*AG	NS	NS	NS	NS	NS	NS	NS
SG*LN*AG	***	NS	NS	NS	NS	NS	NS

* P ≤ 0.05, ** P ≤ 0.01, *** P ≤ 0.001, NS – Not Significant

Significant ($P \leq 0.001$) differences were observed in soil available NH_4^+ - N concentration due to differences in soil great groups (Table 3). As explained earlier this is due to differences in nitrification of NH_4^+ -N in soil (Sahrawat, 1982; and Wickremasinghe *et al.*, 1985). As can be seen from Fig. 2, during the initial period NH_4^+ - N contents in soils were higher at all levels of nitrogen treatments irrespective of the presence or absence of Agrotain. This may be due to rapid conversion of urea to NH_4^+ by hydrolysis. However the behavior of IBL was somewhat different in relation to available NH_4^+ - N content, having lower levels during the initial period. This may be due to low rate of hydrolysis of urea in IBL great soil group. Urease activity, which governs the rate and extent of hydrolysis of urea, depends on the microbial activity of the soil and organic matter content (Bhavanandan and Fernando, 1970). It appears that the fertility status of IBL great soil group is lower compared to RYP and RBL particularly in terms of organic matter (Table 1). This may have caused the rate of hydrolysis to be lower resulting in lesser NH_4^+ - N concentration in soil.

Wickremasinghe *et al.* (1981) studied urea hydrolysis in some tea soils under similar conditions and found that the urea added to soil was hydrolyzed within about 07 days and subsequently NH_4^+ - N is to be derived. However after 02 weeks, the concentration of NH_4^+ -N in all treatments decreased indicating that most of the NH_4^+ - N formed, converted to NO_3^- -N. This is confirmed in Fig. 3 indicating an increase in available NO_3^- - N concentration with time. Higher activity of nitrification is confirmed by the consequent reduction in soil pH (Bhavananthan, 1970). The results in this study are in conformity with the above findings indicating a reduction in pH levels with time.

However in IBL great soil group, towards the end of the trial there was a slight increase in NH_4^+ -N content corresponding to the slight decrease in NO_3^- -N content, in general. Sandanam *et al.* (1978) have reported that, apart from soil pH and the nature of the nitrifying organisms, fertility of the soil may also influence nitrification. Therefore this may be partly due to decreased rate of nitrification towards the latter part of experiment.

Levels of nitrogen application or Agrotain treatment had no significant effect on available NH_4^+ - N concentration in soils (Table 3). Although it was not significant, increasing rates of nitrogen, increased the amount of available NH_4^+ - N concentration in soils. This is due to provision of more NH_4^+ - N caused by the level effect whether it is treated with Agrotain or not. However, lower ammonium concentrations have been reported where the urease inhibitor, Agrotain had been applied than where it had not, due to low rate of breakdown of urea, from a similar experiment carried out in a vertisol soil of the Queensland, Australia by Dowling (1999), but the rates of application of urea were unable to extract from the article for comparison.

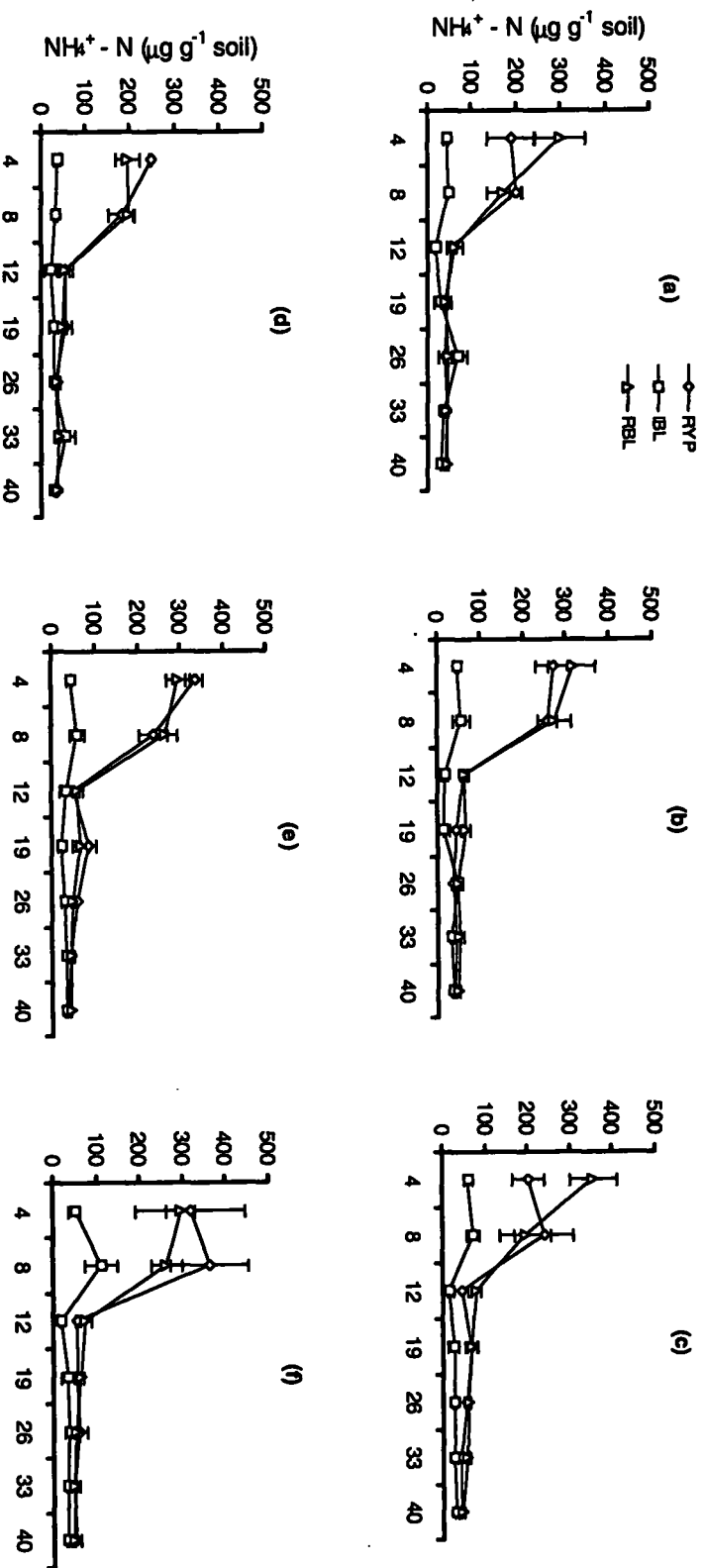


Fig 2: Effect of soil group on $\text{NH}_4^+ - \text{N}$ in soil at increasing rates of N (50, 75 and 100 kg ha⁻¹) with Agrotain (a,b and c respectively) and without Agrotain (d, e and f respectively). Vertical bars represent standard errors of the means.

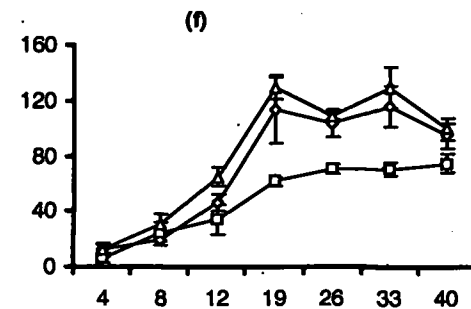
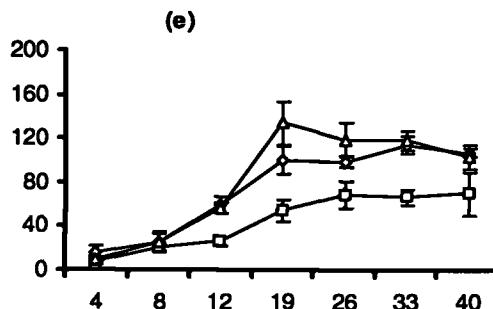
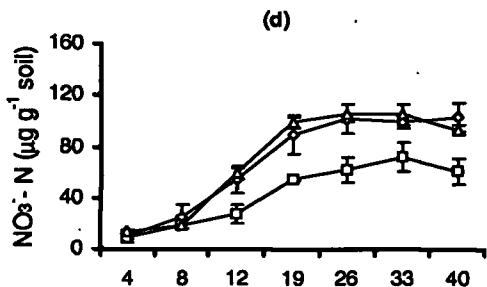
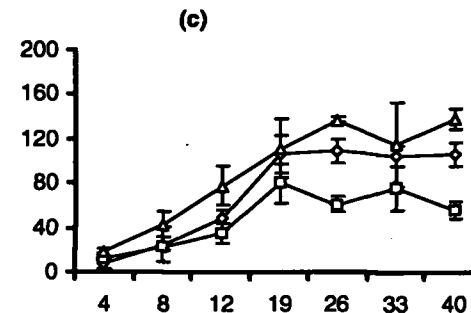
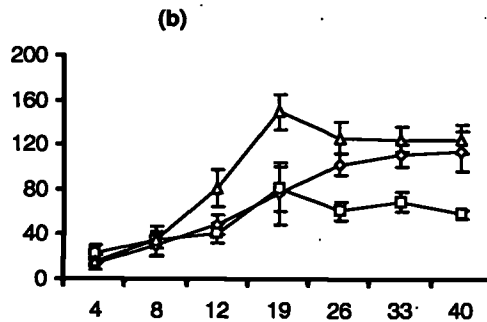
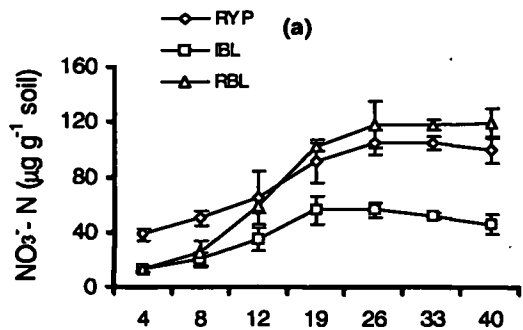


Fig 3: Effect of soil group on NO_3^- -N in soil at increasing rates of N (50, 75 and 100 kg ha^{-1}) with Agrotain (a,b and c respectively) and without Agrotain (d, e and f respectively). Vertical bars represent standard errors of the means.

Treatment of urea with and without Agrotain had a significant ($P \leq 0.01$) effect on soil available NO_3^- -N concentration but only up to the 8th day following the treatment application (Table 4). Thereafter Agrotain had no significant effect on it. Soils treated with Agrotain showed higher contents of NO_3^- -N compared to soils without Agrotain. However, this finding was not in agreement with the Agrotain activity as it should inhibit the rate of nitrification and reduce NO_3^- -N content in the treated soils. Only in the initial 8th day period, NO_3^- -N contents in RYP soil treated with Agrotain at 75 and 100 kg N ha⁻¹ rates, were higher than the non treated soils (Fig. 3). This could be due to the interaction effect, which was significant ($P \leq 0.05$) between soil great group and nitrogen levels during this period. Also soil great groups, nitrogen levels and Agrotain interaction was significant ($P \leq 0.001$) only until fourth day after treatment application. All other interaction effects were found non- significant.

Changes in volatilized NH_3 from soil

Table 5 - Levels of significance for volatilized % of added N as NH_3 due to soil groups (SG), nitrogen levels (LN), and Agrotain levels (AG)

Source	Intervals (days)						
	4	8	12	19	26	33	40
SG	NS	NS	NS	NS	NS	NS	NS
LN	***	*	***	**	NS	*	***
AG	NS	NS	*	NS	NS	NS	NS
SG*LN	NS	NS	NS	*	NS	NS	NS
SG*AG	NS	NS	NS	NS	NS	NS	NS
LN*AG	NS	NS	NS	**	*	NS	NS
SG*LN*AG	NS	NS	NS	**	NS	NS	NS

* $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, NS – Not Significant

No significant differences can be observed in volatilized % of added N as NH_3 due to soil great groups (Table 5). But there was a significant ($P \leq 0.05$) difference due to levels of nitrogen added to soil. However at lowest rate of N (50 kg ha⁻¹) the volatilized % of added N as NH_3 was higher irrespective of Agrotain treatment (Fig. 4). With increasing level of nitrogen, the volatilized N% reduced considerably. Fernando and Bhavanathan (1971) reported that losses of ammonia were proportional to the rate of application of urea from an experiment carried out, using four rates of urea (56, 84, 112 and 140 kg ha⁻¹), to determine the volatilization losses of ammonia from urea when applied to tea soils under field conditions. In the present study this finding was not in agreement with respect to the changes of pH, soil available NH_4^+ and NO_3^- -N levels with time. However this could be due to limitations on the periodic

removal of volatilized ammonia from the reaction site, as in the case of high nitrogen levels, the volume of the polythene chamber to hold volatilized ammonia is low because the amount of nitrogen evolved is considerably high. So that at higher rates of nitrogen, this mechanism appears to inhibit further volatilization of ammonia (Duminda, 2000). Treating of urea with Agrotain did not show any significant effect on reducing volatilization losses of nitrogen as ammonia.

Further, similar results, indicating no significant effect on reducing volatilization losses of NH_3 and the changes of soil pH, soil available NH_4^+ -N and NO_3^- - N have been reported from a another greenhouse and a field experiment carried out in TRI, to ascertain the role of Agrotain on N nutrition using same levels of N (Anon, 2000). Most of the experiments carried out testing the efficacy of Agrotain under field conditions in temperate region, in silt loam or sandy loam soils using corn, cotton, sorghum, wheat like short term crops, have shown increased yields and reduced N losses measured as volatilized NH_3 . However the research data, based on tropical soils are scanty.

It has been reported that, Agrotain urease inhibitor will exhibit some tendency to decompose at elevated temperatures and also lack of performance may result due to unfavorable temperatures and soil conditions (Anon, 1997). This suggests that tea soils incubated under tropical greenhouse conditions could have imposed somewhat different environment from that of temperate conditions soil surface, which wouldn't have given an environment conducive for the Agrotain activity.

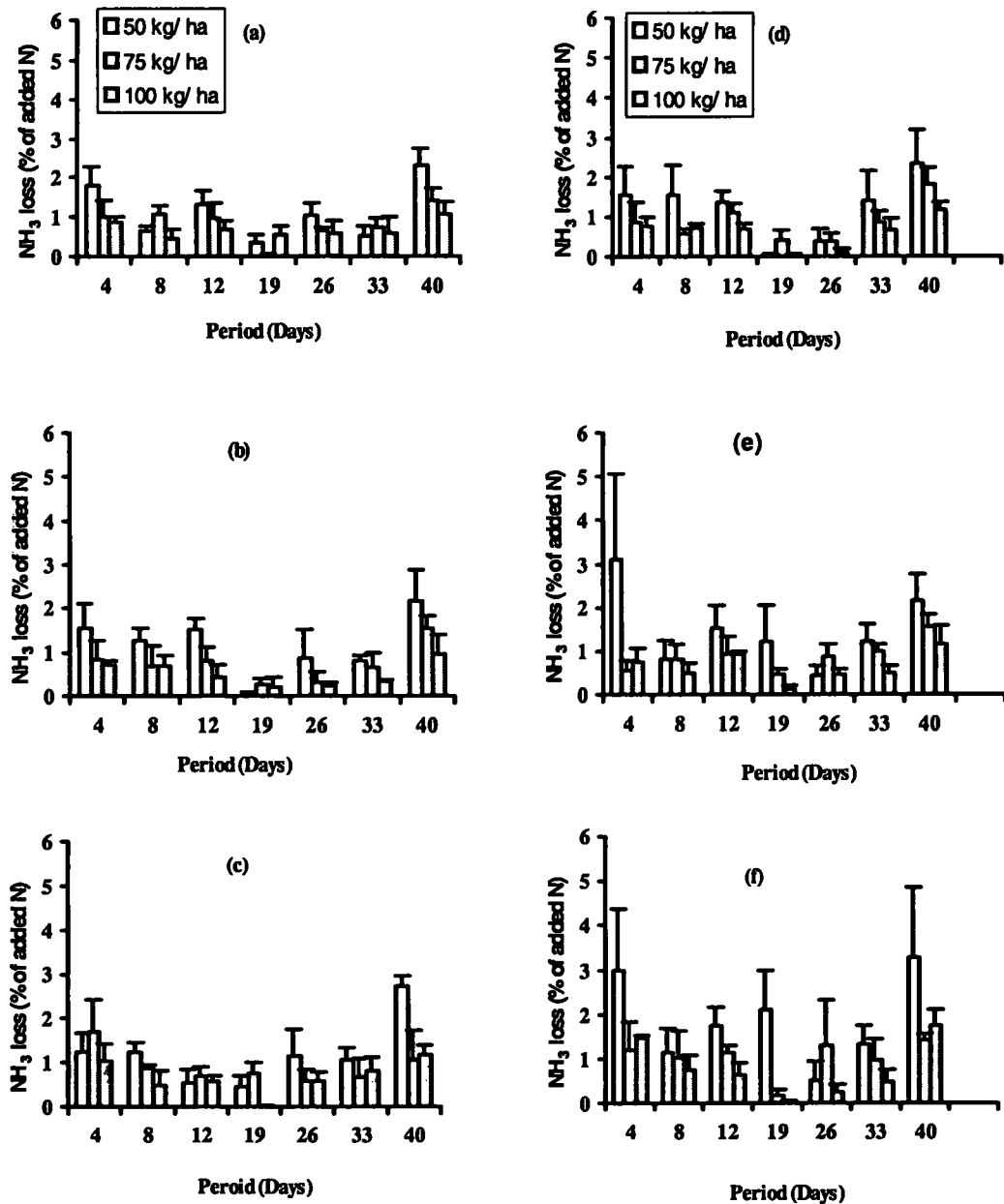


Fig 4 Volatilization losses of NH₃ from urea applied at increasing rates of N for RYP (a,d), IBL (b,e), and RBL (c,f) soil great groups with Agrotain (a,b,c) and without Agrotain (d,e,f). Vertical bars represent standard errors of the means.

CONCLUSION

Agrotain did not show any significant effect on reducing volatilization losses of ammonia, the changes of soil pH (water and CaCl_2) and soil available NH_4^+ - N contents for the applied rates of nitrogen in this study. Moreover significant differences can be observed in soil pH, soil available NH_4^+ - N and NO_3^- -N concentrations due to differences in soil great groups.

ACKNOWLEDGMENTS

The support provided by Mr. T.C.N. Peries and Mr. W.M.S.Wijayatunga is gratefully acknowledged.

REFERENCES

- Anon. 1997 Agrotain Product Information Guidebook IMC - Agrico Company, 2345, Waukegan Road, Suite E-200, Bannockburn, Illinois.
- Anon. 2000 Report of the Actg. Head of Soils and Plant Nutrition Division, Annual Report, Tea Res. Inst. Sri Lanka, 225-226.
- Bhavananthan V P 1970 Studies on the use of urea as a fertilizer for tea in Ceylon 1 Introduction and preliminary observations. Tea Q. 41, 87-91.
- Bhavananthan V P and Fernando V 1970 Studies on the use of urea as a fertilizer for tea in Ceylon 2 - Urease activity in tea soils. Tea Q. 41, 94-98.
- Bremner J M and Mulvaney R L 1978 Urease activity in soils *In* Soil enzymes Ed. R. G Burns pp 149-196., Academic Press, New York.
- De Alwis K A and Panabokke C R 1972 Handbook of soils of Sri Lanka (Ceylon). J. Soil Sci. Soc. Sri Lanka 2, 85
- Dowling C 1999 Modifying the production of ammonium and nitrate from urea bands with urease inhibitor Agrotain (NBPT) in a vertisol soil of the Darling Downs, Queensland: Commun. Soil Sci. Plant Anal. 31, No 11 – 14, pp 1445
- Duminda D M S 2000 The fate of applied nitrogen fertilizer in an acid tea soil. Undergraduate project report. University of Ruhuna, Matara. pp 61-63.
- Fenn L B and Kissel D E 1973 Ammonia volatilization from surface applications of ammonium compounds on calcareous soils: I.General theory. Soil Sci. Am. Proc.37, 55- 59.
- Fernando V Bhavananthan V P 1971 Volatilization losses of ammonia from urea applied to the soil. Tea Q. 42, 48-56.
- Foth H D 1972 Fundamentals of soil science. 5th Ed. John Wiley and Sons, Inc. New York. 177p
- Hesse P R 1971 A Textbook of Soil Chemical Analysis. John Murray, London.
- Jackson M L 1958 Soil Chemical Analysis. Englewood Cliff, Prentice Hall.

- Lamb J 1953 Chemical and Biochemical investigations in Ceylon tea soils. *Tea Q.* 26, 31-36.
- Sahrawat K L 1982 Nitrification in some tropical soils. *Plant and Soil* 65, 281-286.
- Sandanam S, Krishnapillai S and Sabaratnam J 1978. Nitrification of Ammonium Sulphate and Urea in an acid Red Yellow Podzolic tea soil in Sri Lanka in relation to soil fertility. *Plant and Soil* 49, 9-22.
- Sivasubramaniam S 1980 The use of fertilizers for tea in Sri Lanka. *Tea Q.* 49, 137-142.
- Tisdale S L Nelson WE and Bealon J D 1985 *Soil fertility and fertilizers*. 4th Ed. Macmillan publishing company, Inc. New York. pp. 754.
- Wickremasinghe K N, Rodgers G A and Jenkinson D S 1985 Transformation of nitrogen fertilizers in soil. *Soil Biol. Biochem.* 17 (5), 625-630.
- Wickremasinghe K N 1986 The current thinking on the substitution of urea for sulphate of ammonia in tea plantations. *Tea Bull.* 6, 20-35.
- Wickremasinghe K N Sivasubramaniam S and Nalliah P 1981 Urea hydrolysis in some tea soils. *Plant and Soil* 62, 473-477.