Title of experiment: Twin N as a source of nitrogen for tea

Introduction

The industry spends considerable amounts of money on fertilizers each year because high costs. Transportation and application costs further increase the costs of utilising fertilisers. Nitrogen is the main nutrient of all major elements required for tea production. It is usually applied three times per season and at higher rates than any other nutrients.

In Malawi and Zimbabwe, the cost of fertilizer has been increasing over the years. As prices of fertilizers continue to increase along with costs of transportation and application, growers may be faced with a hard choice of reducing the amount of fertilizer they buy and apply per unit area of a tea crop. This would reduce, in the short and long term, the health and productivity of tea bushes, which will threaten the viability of the tea industry and sustainability of tea production.

TwinN has been reported to be a potentially less expensive source of nitrogen compared to the traditional sources of nitrogen. It supplies crop plants with natural form of nitrogen through the activity of endophytic and soil nitrogen fixing microorganisms. These interact with the plant metabolism system to fix atmospheric nitrogen in a form available to plants. TwinN is also reported to increase root growth, assist in solubilisation of phosphorus, increases production of photosynthetic sugar and improves efficiency of plant water use. TwinN is further reported to increase the number of pseudomonad bacteria and manganese-reducing bacteria which are important in increasing plant available manganese and other micronutrients (Mapleton International Pty ltd; 2008). TwinN can easily be applied through fertigation, overhead irrigation or as a foliar spray. When sprayed onto the leaf surface, or root zone, TwinN microbes are effectively absorbed by the plant via leaf stomata, leaf abrasions and lateral root cracks, diluting and multiplying through the plant via the vascular system.

The objectives of this trial are to establish yield and quality response of mature clonal tea to application of TwinN and urea as sources of nitrogen and to determine the combination of TwinN and urea that would produce optimum yield and quality of tea.

Materials and Methods

Experiments on TwinN as a source of nitrogen in tea continued to be monitored by TRFCA at Nsuwadzi and Makandi. The design was RCBD with seven treatments replicated four times. Details of treatments are provided in Table 1. The experimental plot size was 7.1m x 6.3m.

The treatments of the experiment were as described in Table 1.

Table 1: Fertilizer treatments at Makandi and Nsuwadzi

Treatment	Description
T 1	1 application of TwinN + no Urea
T 2	1 application of TwinN + 25 % of recommended N applied as Urea i.e. 68.75 kg/ha N as Urea
T 3	1 application of TwinN + 50% of recommended N applied as Urea i.e.137.5kg/ha N as Urea
T 4	1 application of TwinN + 75% of recommended N applied as Urea i.e. 206.25 kg/ha N as urea
T 5	1 application of TwinN + 100% of recommended N applied as Urea i.e. 275kg/ha N as Urea
T 6	No TwinN applied + zero urea i.e. no nitrogen
T 7	No TwinN + 100% of recommended N as urea i.e 275kg N /ha

Preparation and application of Twin N Mixture

One TwinN package, which contained 1 vial of freeze dried microbes and a rehydration container of microbe food, was used at each site. The rehydration container was filled with 50ml of clean unchlorinated water from a borehole and shaken to ensure a thorough mix of the microbes and water. This mixture was thoroughly mixed with the freeze dried microbes in the vial, allowed to stand for 4 hours at room temperature and added to 200litres of unchlorinated water in a drum and was applied on the plant foliage using a knapsack sprayer on 27 and 28 January, 2011 at Makandi and Nsuwadzi respectively.

Application of inorganic fertilisers

Required quantities of urea for each treatment were calculated and applied. Phosphorus and potassium were applied at recommended rates of 55kg ha^{-1} and 92kg ha^{-1} respectively using Mono ammonium phosphate and Muriate of potash as sources of P and K, respectively. Application rates for P and K were the same for all treatments. These were applied to the plots on 27 and 28 January 2011 at Makandi and Nsuwadzi respectively.

Data on yield, shoot density, fresh and dry masses of different categories of shoots from both sites (Makandi and Nsuwadzi) were recorded. Green leaf plucked from each plot was weighed and converted to made tea yield by multiplying by a factor of 0.22.

Made tea yield data for both sites was summarised and subjected to analysis of variance using GENSTAT computer package. Treatment means were compared using the Duncan's Multiple Range Test (DMRT) at $P<0.05$ in order to find significant differences between treatments.

Results and Discussion

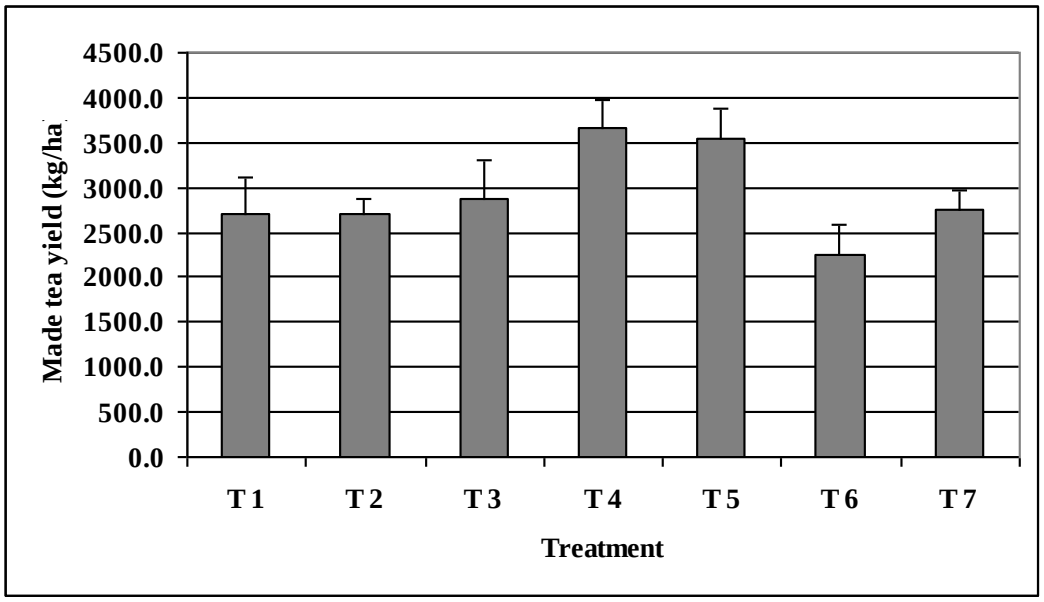
Made tea yield, average shoot dry mass and shoot density

Yield data at Makandi and Nsuwadzi are presented in Figures 1 and 2, respectively.

Made tea yield

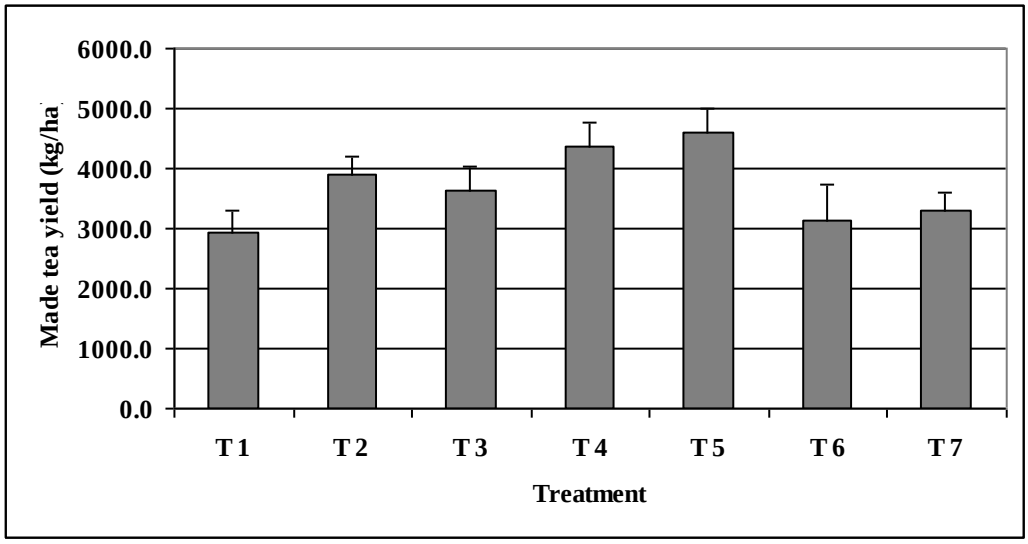
Yield was significantly different among combinations of TwinN and nitrogen from urea at Makandi ($p<0.001$). Highest yield of 3653 kg ha^{-1} was obtained by combining TwinN with 75% of nitrogen from Urea, representing an increase of 32.7% over the standard N application rate of 275kg N ha^{-1} using urea as a source. Combining 100% of N from urea with TwinN significantly increased yield by 28.5% while as combining TwinN with 50% N from urea non-significantly increased yield by 4.7%. A combination of 25% of N from urea with TwinN non-significantly reduced yield to 1.3% while applying TwinN alone led to a non-significant yield reduction of 1.9%. Yield was significantly reduced by 18% in plots where no any nitrogen was applied.

At Nsuwadzi, yield was highly significant among treatments ($p=0.007$). One application of TwinN combined with 100% gave the highest yield of 4609kg ha^{-1} representing a yield increase of 38.9% over the standard application rate of 275kg ha^{-1} using urea as a source of nitrogen. TwinN combined with 75% of recommended N from urea gave a yield of 4375 kg ha^{-1} representing a yield increase of 31.9%. The lowest yield of 2947kg ha^{-1} was obtained when TwinN was applied alone representing a significant yield decline of 11.1%. Combining TwinN and 25% N from urea and TwinN with 50% N from urea significantly increased yield by 17.3% and 9.9% respectively while no application of nitrogen non-significantly reduced yield by 5%.



***Note**: Vertical bars represent standard error of the means*

Figure 1: Made tea yield kg ha^{-1} at different combinations of TwinN and urea as sources of nitrogen at Makandi estate



***Note**: Vertical bars represent standard error of the means*

Figure 2: Made tea yield kg ha^{-1} at different combinations of Twin N and urea as sources of nitrogen at Nsuwadzi estate

Key for Figures 1 and 2:

T 1: 1 application of Twin N + no Urea
T 2: 1 application of Twin N + 25 % of recommended N applied as Urea i.e. 68.75 kg/ha N as Urea
T 3: 1 application of Twin N + 50% of recommended N applied as Urea i.e.137.5kg/ha N as Urea
T 4: 1 application of Twin N + 75% of recommended N applied as Urea i.e. 206.25 kg/ha N as urea
T 5: 1 application of Twin N + 100% of recommended N applied as Urea i.e. 275kg/ha N as Urea
T 6: No Twin N applied + zero urea i.e. no nitrogen
T 7: No Twin N + 100% of recommended N as urea i.e 275kg N /ha (recommended rate)

Shoot dry mass

Dry mass of 2+b, 3+b and 4+b shoots was not affected by application of TwinN and urea applied either separately or in different combinations at Makandi (Table 2). At Nsuwadzi, average mass of 2+b shoots was affected by application of TwinN and urea. All combinations of TwinN and urea produced heavier 2+b shoots than the standard nitrogen application. TwinN applied alone produced shoots that had a similar dry mass to those that received a standard application. Average masses of 1+b, 3+b and 4+b shoots were not affected by the fertilizer treatments. Results on average shoot dry mass for 2+b, 3+b and 4+b shoots at Makandi and Nsuwadzi are presented in Table 2.

Table 2: Shoot dry mass for 1+b, 2+b, 3+b, 4+b shoots at Makandi and Nsuwadzi

	Makandi				Nsuwadzi			
	1+b	**2+b**	**3+b**	**4+b**	**1+b**	**2+b**	**3+b**	**4+b**
1 application of Twin N + no Urea	0.07	0.13	0.20	0.29	0.08	0.16a	0.28	0.44
1 application of Twin N + 25 % of recommended N applied as Urea i.e. 68.75 kg/ha N as Urea	0.06	0.13	0.21	0.32	0.09	**0.19b**	0.34	0.49
1 application of Twin N + 50% of recommended N applied as Urea i.e.137.5kg/ha N as Urea	0.07	0.13	0.20	0.31	0.09	**0.19b**	0.32	0.51
1 application of Twin N + 75% of recommended N applied as Urea i.e. 206.25 kg/ha N as urea	0.06	0.13	0.21	0.34	0.12	**0.19b**	0.33	0.56
1 application of Twin N + 100% of recommended N applied as Urea i.e. 275kg/ha N as Urea	0.07	0.12	0.22	0.32	0.09	**0.18b**	0.32	0.53
No Twin N applied + zero urea i.e. no nitrogen	0.07	0.12	0.20	0.27	0.09	**0.19b**	0.31	0.43
No Twin N + 100% of recommended N as urea i.e. 275kg N /ha (standard)	0.06	0.12	0.19	0.29	0.09	0.16a	0.31	0.46
Mean	0.066	0.125	0.205	0.307	0.092	0.181	0.315	0.488
P Value	0.057	0.149	0.153	0.138	0.054	0.006	0.121	0.233
LSD Value	0.010	0.012	0.019	0.055	0.014	0.019	0.036	0.118
CV (%)	10.5	6.6	6.0	12.0	9.9	6.9	7.8	16.3

Note: Shoot masses followed by the same letters within a column are not significantly different at P<0.05

Shoot density

Shoot density (number of shoots per square meter) significantly varied between TwinN and urea combinations treatments at both sites (Tables 3 and 4). Combinations of 1 application of TwinN and 100% of recommended N applied as Urea i.e. 275kg/ha N as Urea; 1 application of TwinN + 75% of recommended N i.e. 206.25 kg/ha N as urea; 1 application of TwinN + 50% of recommended N applied as Urea i.e.137.5kg/ha N as Urea significantly increased shoot density at Makandi.

Tea plants that received TwinN and 75% of nitrogen from urea had the highest shoot density. The least shoot density was recorded in the standard application of nitrogen (100% nitrogen from urea) and TwinN alone treatments.

Table 3: Shoot density per m^2 at different treatments at Makandi

	banjhi	**1+b**	**2+b**	**3+b**	**4+b**	**Total**
1 application of Twin N + no Urea	1.8c	4.2a	6.8abc	3.1	1.2	17.0a
1 application of Twin N + 25 % of recommended N applied as Urea i.e. 68.75 kg/ha N as Urea	1.8c	4.9b	7.0abc	3.1	1.1	17.9abc
1 application of Twin N + 50% of recommended N applied as Urea i.e.137.5kg/ha N as Urea	1.7c	5.4b	7.6bc	3.3	1.4	19.3bc
1 application of Twin N + 75% of recommended N applied as Urea i.e. 206.25 kg/ha N as urea	1.5ab	5.5b	7.9c	3.4	1.4	19.8c
1 application of Twin N + 100% of recommended N applied as Urea i.e. 275kg/ha N as Urea	1.6ab	5.4b	7.9c	3.4	1.3	19.5c
No Twin N applied + zero urea i.e. no nitrogen	1.7c	4.9b	6.6ab	2.9	1.1	17.2ab
No Twin N + 100% of recommended N as urea i.e 275kg N /ha	1.3a	4.0a	6.1a	3.1	1.2	15.7a
Mean	1.622	4.889	7.120	3.193	1.229	18.05
P value	0.033	<0.001	0.011	0.577	0.418	0.003
LSD Value	0.315	0.617	1.034	0.661	0.349	2.009
CV (%)	13.1	8.5	9.8	13.9	19.1	7.5

Note: Values with the same letters within a column are not significantly different at P<0.05

Table 3: Shoot density at different treatments at per m^{-2} at Nsuwadzi

	banjhi	**1+b**	**2+b**	**3+b**	**4+b**	**Total**
1 application of Twin N + no Urea	1.8c	4.9ab	7.1ab	3.4a	1.6	18.9ab
1 application of Twin N + 25 % of recommended N applied as Urea i.e. 68.75 kg/ha N as Urea	1.5bc	5.3bc	7.8ab	3.6ab	1.5	19.8b
1 application of Twin N + 50% of recommended N applied as Urea i.e.137.5kg/ha N as Urea	1.8c	5.5bc	8.1b	3.9abc	1.8	21.2bc
1 application of Twin N + 75% of recommended N applied as Urea i.e. 206.25 kg/ha N as urea	1.5bc	5.8cd	9.7c	4.3bc	1.7	23.0c
1 application of Twin N + 100% of recommended N applied as Urea i.e. 275kg/ha N as Urea	1.4ab	6.2d	9.2c	4.8c	2.1	23.6c
No Twin N applied + zero urea i.e. no nitrogen	1.7bc	4.7a	6.8a	3.7ab	1.6	18.6ab
No Twin N + 100% of	1.0a	4.6a	6.8a	3.1a	1.5	17.0a

recommended N as urea i.e 275kg N /ha						
Mean	1.528	5.289	7.940	3.835	1.696	20.29
P value	0.004	<0.001	<0.001	0.06	0.115	<0.001
LSD Values	0.396	0.573	1.011	0.785	0.435	2.536
CV (%)	17.5	7.3	8.6	13.8	17.3	8.4

Note: Shoot density numbers followed by the same letters within a column are not significantly different at P<0.05

Made tea quality

Results received from TBCA as of June 2011 indicated that made tea quality, as determined by professional tea tasters from TBCA, did not significantly vary between treatments at Nsuwadzi but at Makandi. At this site, total score and valuation were higher in treatments where TwinN alone, a combination of TwinN with 25%, 50%, 75% and 100% N from urea were applied than in the standard application of nitrogen (Table 4).

Table 4. Total score and valuation at different treatments at Makandi and Nsuwadzi

Trt. No	**Description**	**Makandi**		**Nsuwadzi**	
		Total score	**Valuation (USc kg^{-1})**	**Total score**	**Valuation (USc kg^{-1})**
T 1	1 application of Twin N + no Urea	19.88c	188.5 b	20.74a	189.0ab
T 2	1 application of Twin N + 25 % of recommended N applied as Urea i.e. 68.75 kg/ha N as Urea	19.92c	190.3 b	20.56a	190.0ab
T 3	1 application of Twin N + 50% of recommended N applied as Urea i.e.137.5kg/ha 188.2N as Urea	19.16bc	182.4 b	20.48a	193.5ab
T 4	1 application of Twin N + 75% of recommended N applied as Urea i.e. 206.25 kg/ha N as urea	19.56bc	188.4b	19.75a	188.2a
T 5	1 application of Twin N + 100% of recommended N applied as Urea i.e. 275kg/ha N as Urea	19.28bc	186.5b	19.80a	190.5ab
T 6	No Twin N applied + zero urea i.e. no nitrogen	18.80b	181.6b	20.25a	194.0ab
T 7	No Twin N + 100% of recommended N as urea i.e 275kg N /ha	17.50a	168.8a	20.67a	199.6b
Mean		**19.16**	**183.8**	**20.32**	**192.1**
CV%		**2.9**	**3.6**	**4.9**	**3.4**
P value		**<0.001**	**0.003**	**0.69**	**0.233**
LSD Value		**0.838**	**9.71**	**1.493**	**9.57**

Note: Numbers followed by the same letters within a column are not significantly different at P<0.05

Soil and leaf samples: 112 soil samples and 56 leaf samples collected at both sites, two samples per plot in October 2010, were divided into two lots for analysis. One lot of soil samples was sent to the Agricultural Research and Extension Trust (ARET) and have not been analysed because the analytical laboratory is being renovated. The other lot together with the 56 leaf samples was destined for BEMLAB in RSA but were not sent because payment was not done for analysis of previous samples. The purpose of sending the samples to two laboratories was to compare results between the two laboratories and decide if future samples could be sent to ARET only.

Cost/Benefit Analysis of using TwinN and Urea as nitrogen sources in tea

The income and expenditure analysis of using TwinN as a source of nitrogen on tea in combination with urea shows that TwinN combined with 25%, 50%, 75% N is cheaper than using urea alone (Table 5). Combinations of TwinN and urea at 50% and 75% N produced higher yields than urea applied alone at both Nsuwadzi and Makandi implying that growers could cut costs associated with use of urea while improving on yield as well as increasing returns. However, cutting costs by applying TwinN alone may not be advisable because of the reduction in yield.

Table 5: Expenditure and income analysis of using Twin N and urea as nitrogen sources at Makandi and Nsuwadzi

Trt No	Description	Expenditure on fertilizers (USD/ha)	Total income (USD/ha) (Nsuwadzi	Total Income (USD/ha) (Makandi)
T 1	1 application of Twin N + no Urea	58.00	5570.59	5089.50
T 2	1 application of Twin N + 25 % of recommended N applied as Urea i.e. 68.75 kg/ha N as Urea	153.80	7388.34	5168.55
T 3	1 application of Twin N + 50% of recommended N applied as Urea i.e.137.5kg/ha 188.2N as Urea	243.80	7049.79	5252.57
T 4	1 application of Twin N + 75% of recommended N applied as Urea i.e. 206.25 kg/ha N as urea	**333.80**	**8233.56**	**6881.31**
T 5	1 application of Twin N + 100% of recommended N applied as Urea i.e. 275kg/ha N as Urea	**423.80**	**8779.19**	**6596.32**
T 6	No Twin N applied + zero urea i.e. no nitrogen	-	6197.52	4096.35
T 7	No Twin N + 100% of recommended N as urea i.e. 275kg N /ha	365.80	6619.14	4646.22

Summary
Results in 2010_11 season showed that there was no yield benefit in using TwinN as a sole source of nitrogen in mature clonal tea. Applying TwinN in combination with urea as sources of nitrogen in tea, however, showed a yield benefit, the maximum benefit was realized when applying TwinN together with 75% of nitrogen from urea and when applying TwinN together with 100% nitrogen from urea.

Quality response to TwinN and urea applications significantly varied at Makandi. At this site, valuation of made tea was significantly improved when TwinN and urea were applied in combination and when TwinN was applied alone.

TwinN is a cheaper source of nitrogen than urea but use of TwinN alone in tea decreases yield leading to decreased returns. Application of Twin N in combination with urea at 50% and 75% N will help in reducing costs associated with use of urea as the only source of nitrogen while improving yield as well as increasing returns.