Season, nitrogen rate, and plant type affect nitrogen uptake and nitrogen use efficiency in rice

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Abstract. Studies were undertaken in the Burdekin River Irrigation Area of northern Australia to improve the efficiency of nitrogen (N) use for rice (*Oryza sativa* L.) production. The aim was to maximise grain yield by optimising its functional components: N uptake, efficiency of N use for dry matter production (NUE_{dm}), and harvest index (HI). The effects of season (wet and dry), N rate (0, 70, 140, 210, and 280 kg/ha), and plant type (maturity and stature) on N uptake, NUE_{dm}, and HI were examined in 2 wet and 2 dry seasons.

Leaf area development was closely related to N uptake. In the wet season, genotypes had similar rates of increase in leaf area index (LAI) with N uptake but differed in the level of LAI (curves were parallel). In the dry season, the relationship between N uptake and LAI was different for each genotype (curves not parallel). In both seasons cv. Newbonnet generally had a lower LAI per unit N uptake (i.e. leaf area production was not excessive) than cvv. Lemont and Starbonnet.

Dry matter production and grain yield were also closely related to N uptake. At low levels of N availability (N uptake <100 kg/ha) tissue N concentrations were low and both total above-ground dry mass (AGDM) and grain yield were linearly related to N uptake. As N availability increased, N uptake and tissue N concentration increased, resulting in a deviation of the AGDM and grain yield curves from the linear at about 100 kg/ha N uptake. This finally resulted in AGDM and grain yield plateauing at around 200 kg/ha N uptake. Above this level some factors other than N availability limited yield.

Seasonal differences in N uptake, NUE_{dm}, and HI were observed. Seasonal variation in the response of grain yield to N uptake was found. There was a trend for higher N uptake in the absence of fertiliser application in the wet than the dry season, and the recovery fraction was less for N rates >140 kg/ha in all seasons, i.e. fertiliser N uptake efficiency declined with increasing N rate. Nitrogen was used more effectively by the rice crop to produce grain compared with non-grain parts when average daily mean temperatures were lower during the period between panicle initiation and anthesis.

Genotypic variation was found in N uptake, NUE_{dm}, and HI. The ability to capture these components in crop improvement programs depends on the extent to which genetic linkages between N uptake and both NUE_{dm} and HI can be broken. While our data suggest that N uptake is generally negatively correlated with both NUE_{dm} and HI, there is some evidence that these linkages can be broken. For example, the fact that HI did not change with increasing N uptake in Lemont and, to a lesser extent, in Newbonnet suggests that HI does not always decline with increasing N uptake. The example of Newbonnet suggests that, to some extent, it is possible to increase yield by increasing each of the functional components independently within a specific genotype.

Additional keywords: Oryza sativa, nitrogen uptake, nitrogen use efficiency, harvest index, genotypes, semi-arid tropics.

Introduction

Nitrogen (N) requirements for crop growth are often described by response curves. However, recommendations based on applied fertiliser alone provide limited information on the source of nutrients (fertiliser or soil-derived sources) or the fate of applied fertilisers, and are characterised by considerable seasonal and genotypic variation. An alternative method of analysis, as suggested by van Keulen and Stol (1991), involves a distinction being made between the response of the crop to increased N application and the response of the crop to increased N uptake. Using this approach, seasonal variation in the N applied–grain yield response curves can be explained by differences in the relations between N applied and N uptake. These relations account for differences in N supply from soil-derived sources (the uptake at zero N application) and differences in the recovery of applied N (the slope of the N applied–N uptake regression). Hence, a single relation between N uptake and grain yield may be developed for a given genotype across sites and seasons if the difference is only due to the N applied–N uptake relation. Variation in the slope of the N uptake–grain yield curve among genotypes indicates genetic differences in the efficiency of N use for grain production. It is well documented that genotypes within species differ markedly in ability to take up and use mineral elements (Vose 1987; Clark 1990).

Research on genotypes differing in plant type often shows variation in dry matter production, grain yield, and N uptake. Norman *et al.* (1992) found that 2 plant types differing in height and canopy architecture (Lemont, short-statured and erect leaves; Lebonnet, tall-statured and droopy leaves) were similar in their total N uptake, although Lebonnet produced more dry matter at heading and 21 days after heading. In a related study, Guindo *et al.* (1994) reported that the differential N rate response between Lebonnet and Lemont was associated more with excessive plant growth of Lebonnet at high N rates rather than uptake or translocation of fertiliser N.

Borrell *et al.* (1998) reported how season and plant type affect the response of rice yield to N fertilisation in semi-arid tropical Australia. Grain yield varied among seasons and was negatively correlated with average daily mean temperature during the 30 days before anthesis. Of the 3 genotypes examined, Newbonnet (early-maturing, medium-statured) attained the highest yields by combining high total dry matter production with high harvest index (HI). This paper explores the physiological basis of these responses.

The objective of the research outlined in this paper was to study the effect of season (i.e. wet or dry), N rate, and plant type on N uptake, NUE_{dm} (kg above-ground dry mass/kg N uptake), and HI in flooded direct-seeded rice in a semi-arid tropical environment in northern Australia.

Materials and methods

Experimental details

Experimental details are described in Borrell *et al.* (1998). Briefly, 4 experiments were conducted in 4 consecutive seasons at Millaroo Research Station ($20°03'$ S, $147°16'$ E): Expt 1 (1986 dry season), Expt 2 (1987 wet season), Expt 3 (1987 dry season), and Expt 4 (1988 wet season). In all experiments, 5 rates of N fertiliser (0, 70, 140, 210, and 280 kg/ha) were split for 3 genotypes (cv. Lemont, early-maturing short-statured; cv. Newbonnet, early-maturing medium-statured; and cv. Starbonnet, late-maturing tall-statured). There were 4 replications. While all experiments were harvested at maturity, additional harvests were taken near panicle initiation and anthesis in Expts 2 and 3.

Chemical analyses of plant tissue for N concentration

All harvested dry matter samples were analysed for concentration of N and, in conjunction with dry matter yields, were used to calculate uptake of N. Nitrogen in the senesced tissue was included in the total above-ground N content at the maturity harvest. A subsample (about 0·25 g) of finely ground (1 mm) tissue was digested in sulfuric acid/sodium sulfate mixture using a selenium catalyst in a semi-micro kjeldahl apparatus. The digestate was diluted prior to automated colorimetric determination of N using the indophenol reaction with salicyclate and a nitroprusside catalyst after modification of the Bertholot reaction.

Specific leaf nitrogen

Specific leaf nitrogen (SLN) is positively correlated with the photosynthetic capacity of the rice plant (Sinclair and Horie 1989), and was used in this study as such an indicator. SLN is defined as leaf N uptake per unit leaf area (g N/m^2).

Determination of soil-derived N contribution

The contribution of soil-derived N to crop growth was determined by N uptake in non-fertilised plots. Mini-bays (as described by Borrell *et al.* 1998) prevented the movement of fertiliser N in the floodwater between treatments, ensuring that only soil N was utilised by plants in non-fertilised plots. While the amount of N in the irrigation water was not determined, it was considered to be negligible.

Statistical analysis

N uptake in the straw and grain at maturity, total above-ground N uptake (AGNU) at maturity, and N harvest index (kg N uptake in grain/ kg AGNU) were analysed by standard analysis of variance. Concentration of N in the various organs of the rice plant at each of panicle initiation, anthesis, and maturity for Expts 2 and 3 were also analysed by standard analysis of variance. Pairwise comparisons of means were performed using the protected l.s.d. procedure at $P = 0·05$. Pooled analyses were performed across experiments for AGNU, efficiency of N use for dry matter production (NUE_{dm}), efficiency of N use for grain production (kg grain dry mass/kg N uptake, NUE_g), fertiliser N uptake efficiency, and N harvest index. Fertiliser N uptake efficiency was derived from the proportional relationship of total above-ground N uptake (kg/ha) to fertiliser N applied (kg/ha). Correlations were calculated between average daily

mean temperature during the 30 days before anthesis and AGNU, NUE_{dm}, NUE_g, and N HI.

Curve fitting

Relations between N applied and AGNU and between AGNU and leaf area index (LAI), above-ground dry mass (AGDM), NUE_{dm}, HI, and grain dry mass were investigated.

Preliminary analyses revealed that data for most of the above relations could not be combined over season (i.e. experiments) or phenological event (i.e. panicle initiation, anthesis, and maturity) so, for consistency, seasons and phenological events were always treated separately.

For each of the above relations, genotypes were compared by performing a parallel curve analysis within each season or phenological event. That is, individual curves were fitted for each genotype. Tests were then performed to determine parameters that were not significantly different ($P > 0.05$) among genotypes based on a pooled residual variance from the regressions of the 3 genotypes. Parameters that were not different were constrained to be common. The procedure was 'step-down' with non-linear parameters tested first, linear parameters next, and finally the constants.

Exponential curves were fitted to the relation between N applied and AGNU for each of the 4 seasons (Expts 1–4). Nitrogen uptake data were meaned over replicates for each genotype at each level of N applied.

Exponential curves were also fitted to the relations between AGNU and (*i*) AGDM at each of panicle initiation, anthesis and maturity for Expt 2 (wet season) and Expt 3 (dry season); (*ii*) LAI at each of panicle initiation and anthesis for Expt 2 (wet season) and Expt 3 (dry season); and (*iii*) grain yield for the 4 seasons (Expts 1–4). Individual plot data were used for these regressions.

The relation between AGNU and NUE_{dm} at each of panicle initiation, anthesis, and maturity for Expt 2 (wet season) and Expt 3 (dry season) and between AGNU and HI for the 4 seasons (Expts 1–4) were described by linear regressions. Individual plot data were used for these regressions. Within season and phenological event, genotypes were compared by an analysis of parallelism. For the relation between N uptake and HI, preliminary analyses revealed that the linear regression of HI on N uptake for Lemont was not significant (i.e. the slope was not significantly different from zero; $P > 0.05$) in each of the 4 seasons. Consequently, Lemont was removed from the pooled regression analysis and tests of parallelism were only performed for Newbonnet and Starbonnet. The computer program GENSTAT 5 (Genstat 5 Committee 1993) was used to fit the exponential and linear regressions.

Results

Nitrogen uptake

Seasonal variation in N uptake was observed. AGNU at maturity was higher ($P < 0.01$) in Expt 2 (195 kg/ha) than Expts 1, 3, and 4 (mean of 163 kg/ha). N uptake was not significantly correlated with ADMT during the 30 days before anthesis.

N uptake in the straw and grain increased ($P < 0.05$) with N rate in all seasons (Table 1). Pooled analysis over seasons and genotypes revealed that AGNU increased ($P < 0.01$) from 84 kg N/ha (0 kg N/ha applied) to 237 kg N/ha (280 kg N/ha applied).

Genotypic variation in N uptake at maturity was found in the dry season only (Table 1). AGNU in Newbonnet was higher ($P < 0.05$) than in Lemont for both dry seasons, although Newbonnet exceeded Starbonnet in Expt 1 only. Interestingly, N uptake in the straw was higher ($P < 0.05$) in Starbonnet than in Newbonnet in 2 of 4 experiments, yet N uptake in the grain was higher ($P < 0.05$) in Newbonnet in all experiments. Hence, N harvest index was higher ($P < 0.05$) in Newbonnet than Starbonnet in all experiments.

Exponential curves were fitted to the relation between N applied and N uptake, although the slope was close to linear in 3 of 4 experiments (Fig. 1). A significant ($P < 0.05$) genotype effect was evident in the relation between N applied and N uptake in both dry seasons with genotype curves being parallel (Fig. 1*a*, *b*). The level of N uptake for Lemont was lower than Starbonnet, which was lower than Newbonnet for any given level of applied N. There was no significant difference ($P > 0.05$) among genotypes in either wet season, so the relationship was described by a common curve in each season (Fig. 1*c*, *d*). In the absence of N fertilisation, all genotypes extracted about 90 kg N/ha from the soil in the wet season, yet in the dry season Newbonnet, Starbonnet, and Lemont extracted 93, 74, and 60 kg N/ha, respectively, from the soil.

Fertiliser nitrogen uptake efficiency

The proportional relationship of total N uptake (kg/ha) to fertiliser N applied (kg/ha) varied ($P < 0.01$) among seasons: Expt 2 (1.51) > Expts 1, 3, and 4 (mean of 1.21). These relationships declined ($P < 0.01$) with increasing N rate such that the ratio for 70 kg N/ha applied (1.97) > 140 kg N/ha (1.27) > 210 kg N/ha (1.04) > 280 kg N/ha (0.85). Genotypic variation ($P < 0.01$) in fertiliser N uptake efficiency was also observed such that Newbonnet (1.35) > Lemont (1.22). Starbonnet was intermediate (1.28) in this parameter. In the case of Lemont when 70, 140, 210, and 280 kg N/ha was applied, the respective fertiliser N uptake efficiencies were 1.94, 1.23, 0.93, and 0.78.

Nitrogen use efficiency

Unless otherwise stated, the wet and dry season data presented in the following sections relate to Expts 2 and 3, respectively.

Efficiency of N use for leaf area production

Leaf area development was closely related to N uptake. There was considerable genotypic variation in the relationship between N uptake and LAI (Fig. 2). Genotypes had similar rates of increase in LAI with

Table 1. Nitrogen uptake in straw and in grain and total N uptake (kg/ha), and N harvest index (N HI) for five rates of applied N and three genotypes across four experiments

Means within a column, experiment, and treatment followed by a common letter are not significantly different at $P = 0.05$

Treatment	N uptake in straw	N uptake in grain	Total N uptake	N HI
Expt 1 (dry season)				
Rate of N (kg/ha)				
0	21a	48a	71a	0·70bc
70	28a	84b	112b	0·75c
140	52b	119c	170c	0·69b
210	70c	139d	211d	0·67ab
280	86c	138d	220d	0·62a
Genotype				
Lemont	44a	99a	145a	0·69b
Newbonnet	48a	122b	172b	0·74c
Starbonnet	62b	95a	153a	0·62a
Expt 2 (wet season)				
Rate of N (kg/ha)				
0	38a	58a	96a	0·60c
70	70b	109b	178b	0·61c
140	86b	115b	203c	0·57bc
210	125c	116b	241d	0·49ab
280	142d	116b	257d	0·44a
Genotype				
Lemont	89a	97a	186a	0·52a
Newbonnet	89a	115b	204a	0·59b
Starbonnet	98a	97a	195a	0·51a
Expt 3 (dry season)				
Rate of N (kg/ha)				
0	40a	44a	84a	0·53a
70	52a	79b	131ab	0·62b
140	77ab	90b	170bc	0·54ab
210	108bc	110c	219cd	0·51a
280	135c	106c	241d	0·47a
Genotype				
Lemont	63a	84ab	149a	0·58b
Newbonnet	92b	97b	189b	0·55b
Starbonnet	93b	76a	169ab	0·47a
Expt 4 (wet season)				
Rate of N (kg/ha)				
0	29a	56a	85a	0·66b
70	46b	83ab	131b	0·65b
140	75c	93b	169c	0·56a
210	102d	105bc	207d	0·51a
280	100d	118c	220d	0·53a
Genotype				
Lemont	63a	96ab	159a	0·61b
Newbonnet	67a	98b	166a	0·61b
Starbonnet	82b	80a	161a	0·52a

N uptake at panicle initiation and anthesis in the wet season (i.e. curves were parallel, Fig. 2*a*, *c*) but differed in the level of LAI. At panicle initiation, Lemont and Starbonnet had significantly ($P < 0.05$) higher LAI than Newbonnet and at anthesis, Starbonnet had significantly ($P < 0.05$) higher LAI than Lemont and Newbonnet. Leaf area index approached a plateau for N uptakes greater than 130 kg N/ha at panicle initiation and for N uptakes >400 kg N/ha at anthesis.

In the dry season, the relationship between N uptake and LAI was different for each genotype at both panicle initiation and anthesis (Fig. 2*b*, *d*, respectively) with curves having significantly different ($P < 0.05$) non-linear components of curvature. At both panicle initiation and anthesis, Lemont and Starbonnet tended to respond similarly while Newbonnet responded quite differently: at panicle initiation, Lemont and Starbonnet began to plateau while Newbonnet continued to
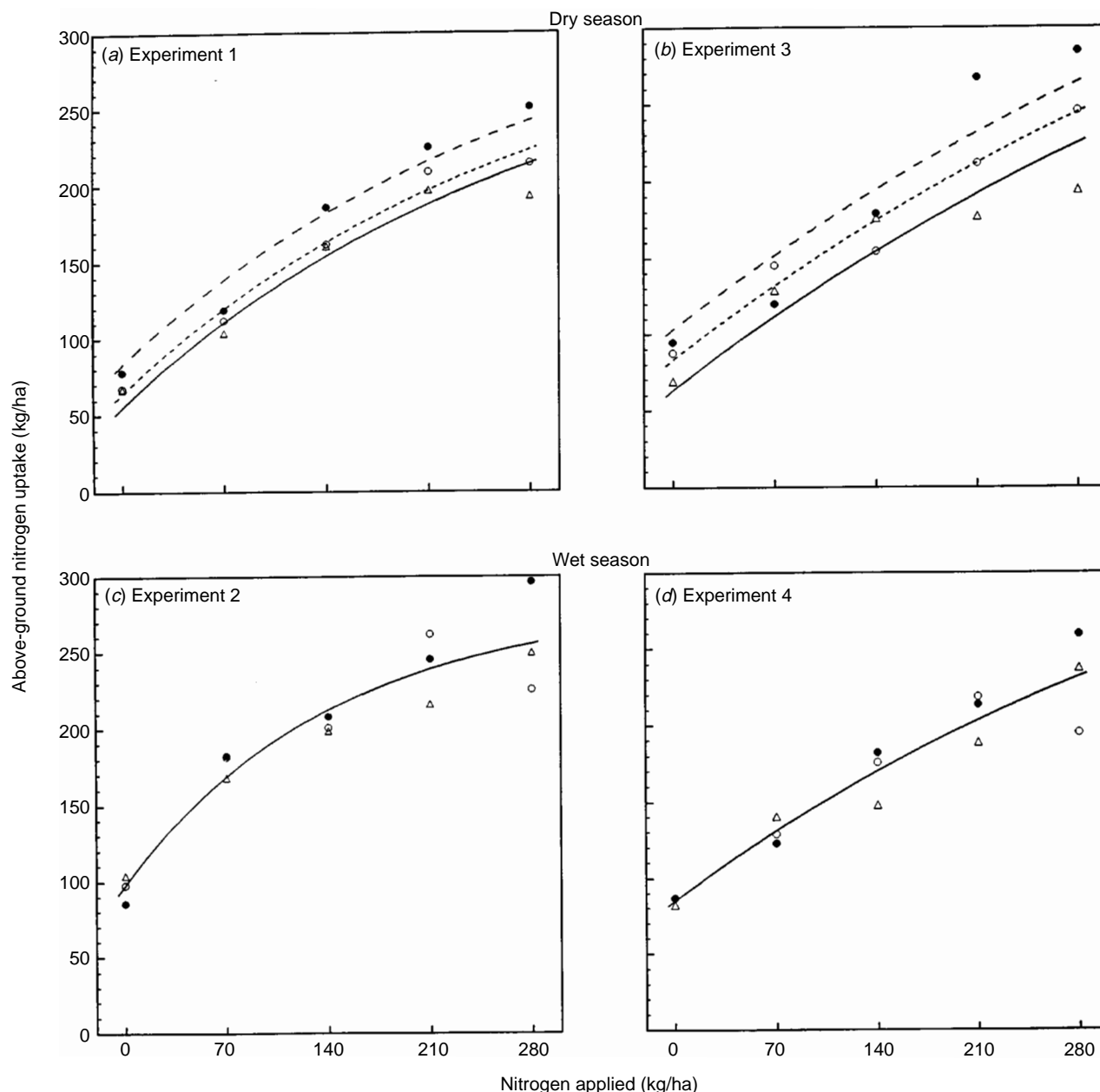
Fig. 1. Relation between nitrogen applied and above-ground nitrogen uptake in two dry seasons (a, $r^2 = 0.955$; b, $r^2 = 0.868$) and in two wet seasons (c, $r^2 = 0.899$; d, $r^2 = 0.907$) for three rice genotypes: Lemont ($\triangle$), Newbonnet ($\bullet$), Starbonnet ($\bigcirc$). Exponential curves were fitted to the data. Line styles for genotypes in (a) and (b): Lemont (—), Newbonnet (– –), and Starbonnet (- - -).

increase in LAI (almost linearly) over the range of N uptakes; at anthesis, Lemont and Starbonnet continued to increase in LAI (almost linearly) while Newbonnet plateaued for N uptake greater than 200 kg N/ha.

Efficiency of N use for dry matter production

NUE_{dm} (kg AGDM/kg AGNU) varied ($P < 0.01$) among seasons: Expt 3 (117 kg/kg) > Expts 1 and 4 (109 kg/kg) > Expt 2 (102 kg/kg). NUE_{dm} was not significantly correlated with ADMT (°C) during the 30 days before anthesis.

A pooled analysis across seasons and genotypes found that NUE_{dm} declined ($P < 0.01$) with increasing N rate from 120 to 88 kg/kg as N rate increased from 0 to 280 kg/ha. Nitrogen harvest index also declined ($P < 0.01$) with increasing N rate, regardless of genotype and season, indicating the proportion of N partitioned to the grain compared with non-grain parts was lower for higher N rates.
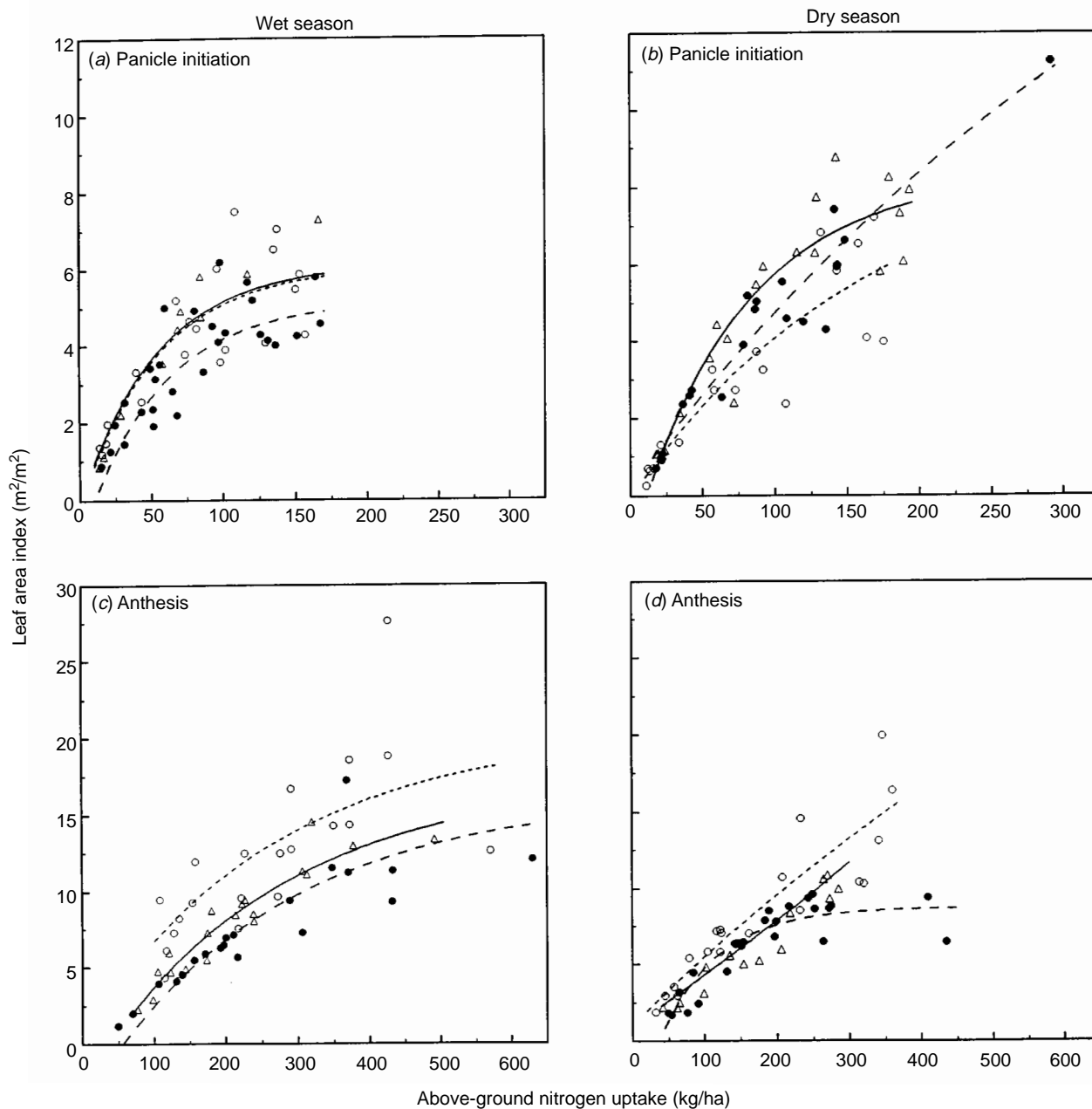
Fig. 2. Relation between above-ground nitrogen uptake and leaf area index at panicle initiation during the (*a*) wet season ($r^2 = 0.762$) and (*b*) dry season ($r^2 = 0.860$), and at anthesis during the (*c*) wet season ($r^2 = 0.734$) and (*d*) dry season ($r^2 = 0.832$) for three rice genotypes: Lemont ($\triangle$—$\triangle$), Newbonnet ($\bullet$--$\bullet$), Starbonnet ($\circ$---$\circ$). Data from Expts 2 and 3 only were used. Exponential curves were fitted to the data.

At panicle initiation there was no genotypic difference in the relation between N uptake and AGDM in the wet season (Fig. 3*a*), but there was in the dry season (Fig. 3*b*), with Lemont and Newbonnet having consistently greater AGDM than Starbonnet. The reverse was the case at anthesis, with a genotype effect (Lemont with lower AGDM than Starbonnet, and Newbonnet intermediate; curves being parallel) in the wet season (Fig. 3*c*) and no genotype effect in the dry season (Fig. 3*d*). Also at panicle initiation, AGDM tended to be greater in the dry season than in the wet but at anthesis the reverse was true.

At panicle initiation, the slope of the N uptake–AGDM regression increased rapidly to uptakes of about 130 and 160 kg N/ha (based on 90% of estimated asymptote) for the wet and dry seasons respectively, indicating
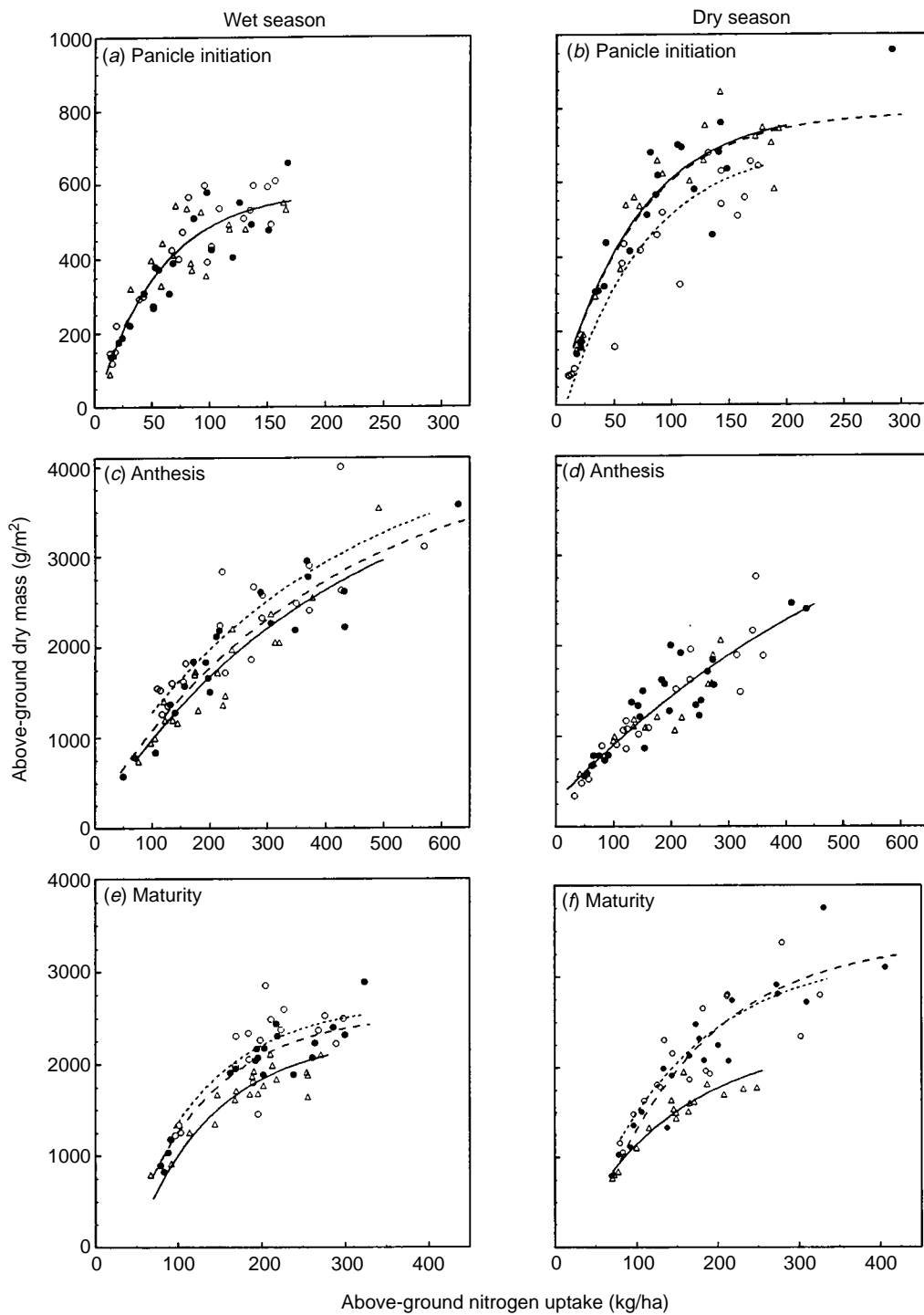
Fig. 3. Relation between above-ground nitrogen uptake and above-ground dry mass at panicle initiation in the (a) wet season ($r^2 = 0 \cdot 829$) and (b) dry season ($r^2 = 0 \cdot 870$), and at anthesis in the (c) wet season ($r^2 = 0 \cdot 826$) and (d) dry season ($r^2 = 0 \cdot 822$), and at maturity in the (e) wet season ($r^2 = 0 \cdot 913$) and (f) dry season ($r^2 = 0 \cdot 855$) for three rice genotypes: Lemont ($\triangle$), Newbonnet ($\bullet$), Starbonnet ($\bigcirc$). Line styles for genotypes in (b), (c), (e), and (f): Lemont (—), Newbonnet (– –), Starbonnet (- - -). Data from Expts 2 and 3 only were used. Exponential curves were fitted to the data.

Table 2. Concentration of nitrogen (%) in selected organs of rice at three times of harvest for two seasons and five rates of applied nitrogen

Means within a row followed by a common letter are not significantly different at $P = 0.05$; DAS, days after sowing

	Rate of applied nitrogen (kg/ha)				
	0	70	140	210	280
Wet season (Expt 2)					
Panicle initiation (48 DAS)					
Stem	0·87a	0·92ab	1·12b	1·49c	1·66c
Leaf	1·69a	2·06b	2·65c	3·2d	3·50d
Anthesis (84 DAS)					
Stem	0·54a	0·53a	0·67ab	0·78b	0·95c
Leaf	1·82a	1·93a	2·14a	2·57b	2·96c
Panicle	1·46a	1·35a	1·36a	1·22a	1·15a
Maturity (129 DAS)					
Straw	0·61a	0·61a	0·67a	0·87b	1·02c
Grain	1·23a	1·33a	1·45b	1·61c	1·77d
Dry season (Expt 3)					
Panicle initiation (66 DAS)					
Stem	0·82a	0·77a	1·00ab	1·30bc	1·52c
Leaf	2·12a	2·19a	2·67b	3·30c	3·77d
Anthesis (94 DAS)					
Stem	0·57a	0·64ab	0·70b	0·87c	1·10d
Leaf	1·81a	2·07b	2·36c	2·67d	3·07e
Panicle	0·90a	0·84a	0·94ab	0·98ab	1·09b
Maturity (130 DAS)					
Straw	0·65ab	0·48a	0·60ab	0·76b	0·87c
Grain	1·09a	1·15a	1·28b	1·40c	1·45c

limiting N concentrations in the leaf and stem at low N availability (Table 2). At anthesis, the rate of dry matter production in both seasons did not decrease substantially (i.e. the curves did not plateau) over the range of uptakes considered (Fig. 3c, d). This may indicate limiting N concentrations in the tissue over the range of available N (Table 2).

At maturity, significant genotypic variation was present in both seasons (Fig. 3e, f). For the wet season, curves were parallel with genotypes attaining 90% of the estimated maximum AGDM production at N uptakes of 254–268 kg N/ha (Fig. 3e). In the dry season, genotypes had a similar nonlinear component of curvature but a significantly different ($P < 0.05$) linear component of curvature with genotypes reaching 90% of the estimated maximum AGDM production at N uptakes of 315–340 kg N/ha (Fig. 3f). For Lemont, this asymptotic value is outside the range of data and is therefore a poor estimate of N uptake required for 90% of the estimated maximum AGDM production. In both seasons, the medium- and tall-statured genotypes (Newbonnet and Starbonnet) had consistently greater AGDM than the short-statured genotype (Lemont), particularly in the dry season.

NUE$_{dm}$ declined linearly with increasing N uptake at panicle initiation, anthesis, and maturity in both seasons (Fig. 4). There were no significant differences ($P > 0.05$) in the rate of decline among genotypes within seasons and phenological events (i.e. lines for genotypes were parallel), and in some cases (at panicle initiation for the wet season and at anthesis for the dry season), there were no differences in intercepts among genotypes (i.e. a common line was fitted for all genotypes).

The rate of decline in NUE$_{dm}$ was greatest at panicle initiation (0·38 and 0·25 units/kg N·ha for wet and dry seasons, respectively) and least at anthesis (0·12 and 0·17 units/kg N·ha for wet and dry seasons, respectively). Newbonnet and Starbonnet exceeded ($P < 0.05$) Lemont in NUE$_{dm}$ at maturity in both seasons (Fig. 4e, f). Less consistent genotypic variation was evident at panicle initiation and anthesis.

Efficiency of N use for grain production (NUE$_g$)

NUE$_g$ (kg grain/kg AGNU) varied ($P < 0.01$) among seasons such that Expt 1 (54·7 kg/kg) > Expt 4 (47·5 kg/kg) > Expt 3 (42·7 kg/kg) > Expt 2 (37·9 kg/kg). Borrell *et al.* (1998) found rice grain yield in northern Australia to be negatively correlated ($r^2 = 0.93^*$) with average daily mean temperature (°C, ADMT) during the 30 days before anthesis. Similarly, NUE$_g$ (kg/kg) was highly negatively correlated ($r^2 = 0.99^{**}$) with ADMT (°C) over the same period in the current study ($y = 220 - 6.39x$).
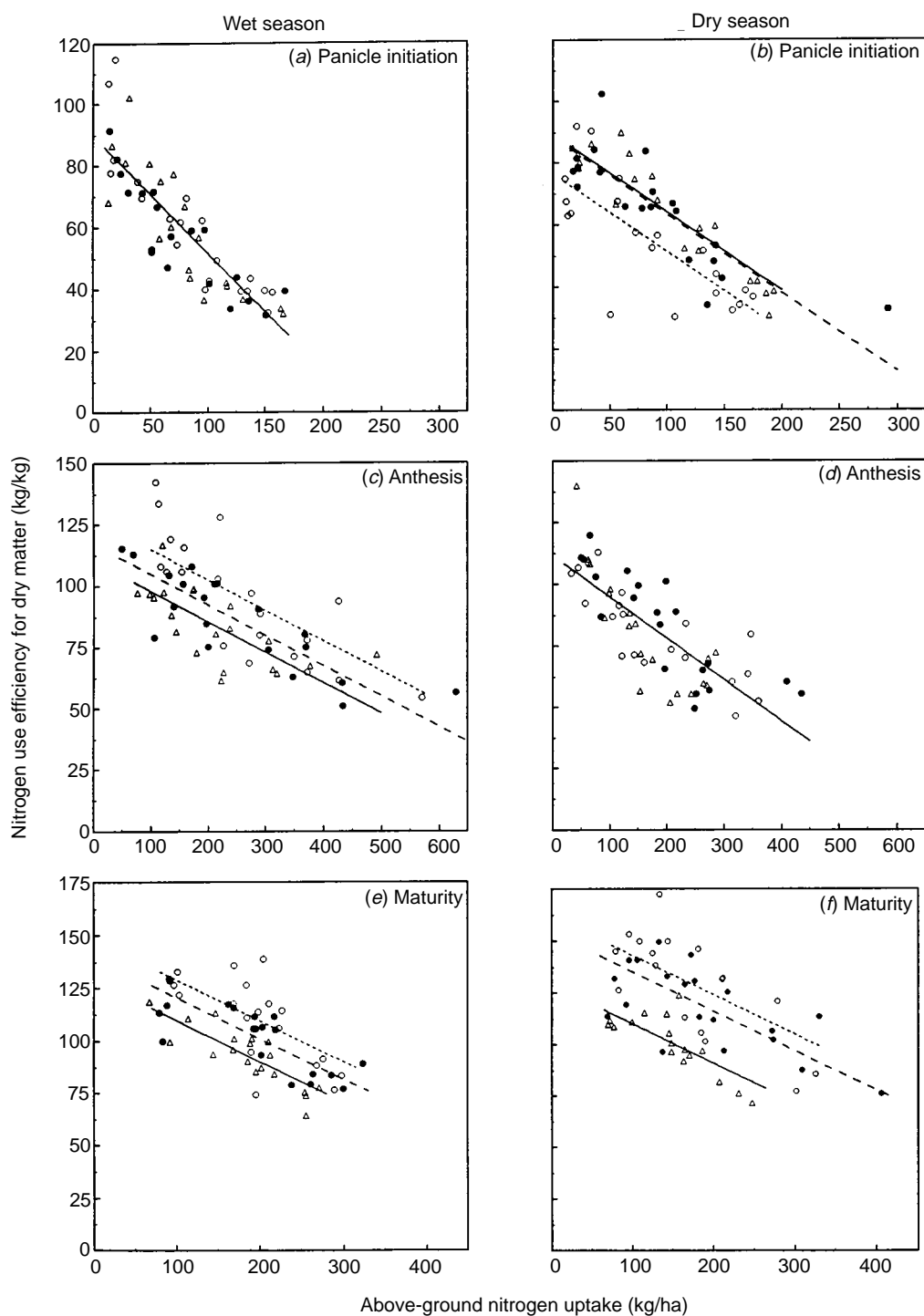
Fig. 4. Relation between above-ground nitrogen uptake and nitrogen use efficiency for dry mater (NUE$_{dm}$) at panicle initiation in the (a) wet season ($r^2 = 0\cdot741$) and (b) dry season ($r^2 = 0\cdot708$), and at anthesis in the (c) wet season ($r^2 = 0\cdot603$) and (d) dry season ($r^2 = 0\cdot610$), and at maturity in the (e) wet season ($r^2 = 0\cdot610$) and (f) dry season ($r^2 = 0\cdot599$) for three rice genotypes: Lemont ($\triangle$), Newbonnet ($\bullet$), Starbonnet ($\bigcirc$). Line styles for genotypes in (b), (c), (e), and (f): Lemont (—), Newbonnet (– –), Starbonnet (- - -). Data from Expts 2 and 3 only were used. Linear regressions were fitted to the data.
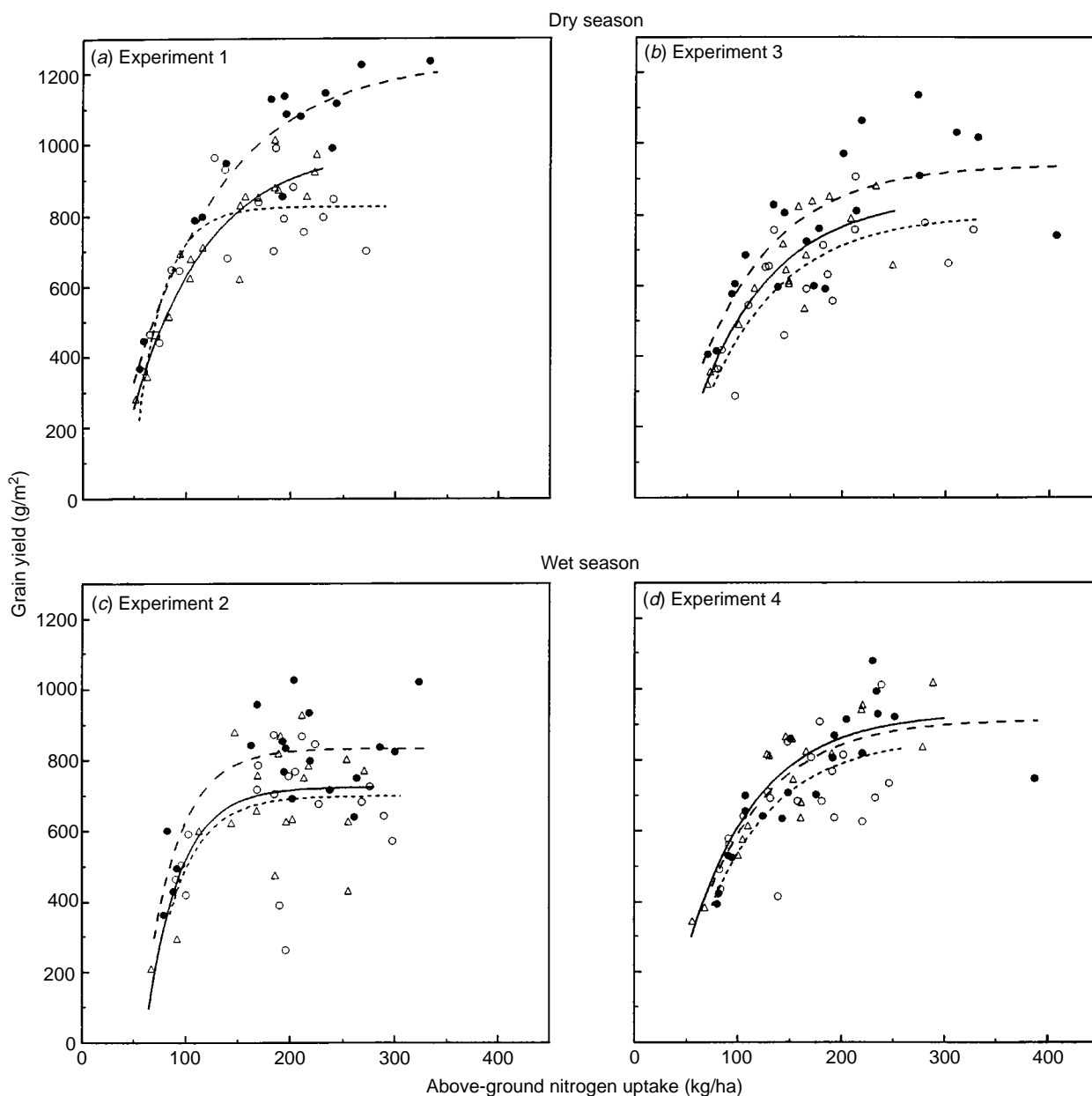
Fig. 5. Relation between above-ground nitrogen uptake and grain dry mass at maturity in two dry seasons (a, $r^2 = 0 \cdot 870$; b, $r^2 = 0 \cdot 675$) and in two wet seasons (c, $r^2 = 0 \cdot 467$ and d, $r^2 = 0 \cdot 670$) for three rice genotypes: Lemont ($\triangle$—$\triangle$), Newbonnet (●–●), Starbonnet (○---○). Exponential curves were fitted to the data.

At low N availability, grain yield was proportional to N uptake in all genotypes, indicating constant N-limiting concentrations in both the grain and straw (Fig. 5). With increasing N uptake the concentration in the grain and straw at maturity increased (Table 2), resulting in a reduced slope, and finally a plateau was reached with uptakes of about 130 kg N/ha (Expt 2) and 190 kg N/ha (Expts 1, 3, and 4) where some factor other than N availability limited yield.

In Expt 1 (Fig. 5a), both the linear and the nonlinear components of curvature were significantly different ($P < 0 \cdot 05$) among genotypes, with Starbonnet reaching a plateau very rapidly (at an uptake of about 105 kg N/ha) while Lemont and Newbonnet responded more similarly attaining 90% of their estimated maximum yield at N uptakes of 189 and 225 kg N/ha, respectively. At high levels of N supply (N uptake > 200 kg/ha), yield was greatest for Newbonnet and least for Starbonnet, with Lemont intermediate.
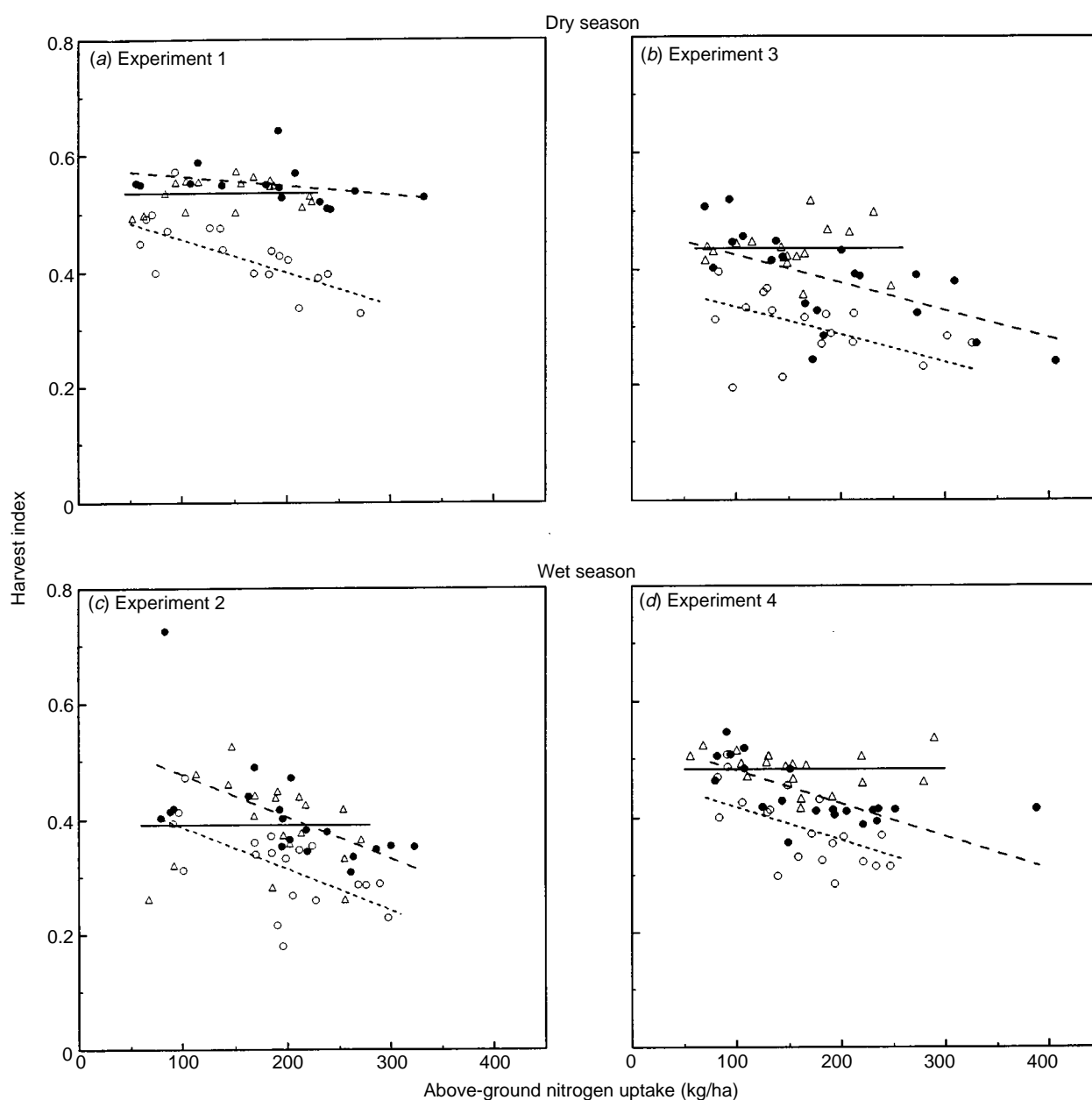
Fig. 6. Relation between above-ground nitrogen uptake and harvest index in two dry seasons (a, $r^2 = 0.813$; b, $r^2 = 0.482$) and in two wet seasons (c, $r^2 = 0.487$); d, $r^2 = 0.497$ for three rice genotypes: Lemont ($\triangle$—$\triangle$), Newbonnet ($\bullet$--$\bullet$), Starbonnet ($\circ$---$\circ$). Linear regressions were fitted to the data.

For Expts 2–4, genotypes had similar rates of increase in grain yield with total N uptake but differed in the level of yield (i.e. curves were parallel; Fig. 5b, c, and d). In Expt 2 (Fig. 5c), genotypes showed responses in grain yield up to about 130 kg N/ha (based on attaining 90% of maximum yield) while for most genotypes in the other experiments responses in grain yield to N uptake were evident up to 180–200 kg N/ha.

Yield plateaued at an N uptake of about 190 kg/ha in Expts 3 and 4, equating to N applied of about 170 kg/ha. This fertiliser application, together with the 80 kg N/ha supplied from the soil, equates to a total of 250 kg N/ha available to the crop. Of the 250 kg N/ha available, 190 kg/ha was taken up by the crop, i.e. 76% of the available N was recovered by the crop. Similar logic can be applied to Expts 1 and 2.

Nitrogen concentration in the grain was similar for N applied up to 140 kg/ha but increased thereafter without any subsequent increase in yield (Table 2). This indicates that N applied above 140 kg/ha did not increase yield but simply accumulated in the grain.

Efficiency of N use for harvest index (NUE_{hi})

The effect of N uptake on the partitioning of carbohydrate between grain and non-grain parts is important. NUE_{hi} is an index of this partitioning, expressed as HI units/kg total N uptake·ha. A pooled analysis over N treatments and genotypes found seasonal variation in NUE_{hi}: Expt 1 (0·0041 units/kg N·ha) > Expt 4 (0·0032 units/kg N·ha) > Expt 3 (0·0027 units/kg N·ha) > Expt 2 (0·0022 units/kg N·ha). NUE_{hi} (units/kg N·ha) was highly negatively correlated ($r^2 = 0.99^{**}$) with ADMT (°C) during the 30 days before anthesis ($y = 2·28 - 0·072x$). NUE_{hi} also declined ($P < 0·01$) with increasing uptake of N from 0·0057 units/kg N·ha (0 kg N/ha applied) to 0·0017 units/kg N·ha (280 kg N/ha applied).

Nitrogen uptake did not influence HI for Lemont in any of the 4 experiments and, hence, was constant over the range of N uptakes (Fig. 6). HI for Lemont was 0·54 (Expt 1, s.e. 0·01), 0·39 (Expt 2, s.e. 0·02), 0·44 (Expt 3, s.e. 0·01), and 0·48 (Expt 4, s.e. 0·01).

Newbonnet and Starbonnet had similar linear rates of decline in HI with increasing N uptake (0·003 units/kg N·ha) within all seasons except Expt 1 (dry season), suggesting a constant preferential allocation of carbohydrate to the non-grain parts with increasing N uptake. In Expt 1, the rate of decline in HI with increasing N uptake was always less for early-maturing Newbonnet than for late-maturing Starbonnet (0·00016 *v.* 0·00057 units/kg N·ha, respectively). However in all experiments, Newbonnet had a significantly higher ($P < 0·05$) intercept and hence, higher HI, than Starbonnet.

Specific leaf nitrogen

Specific leaf nitrogen at anthesis was higher ($P < 0·05$) in Newbonnet than Lemont and Starbonnet in the wet and dry seasons (Table 3).

Table 3. Specific leaf nitrogen (g N/m^2) at anthesis in three rice genotypes for two seasons

Means within a column followed by a common letter are not significantly different at $P = 0·05$

Genotype	Specific leaf nitrogen	
	Wet season	Dry season
Lemont	1·24a	1·13a
Newbonnet	1·63b	1·49b
Starbonnet	1·36a	1·24a
Mean	1·41	1·29

Discussion

Effect of season

Total N uptake at maturity was about 160 kg/ha in 3 of 4 experiments, and was higher in only Expt 2 (195 kg/ha). Similarly, the proportional relationship of total N uptake/fertiliser N applied (fertiliser nitrogen uptake efficiency) was about 1·21 in Expts 1, 3, and 4, but higher in Expt 2 (1·51). Furthermore, some variation in the response of grain yield to N uptake was observed among seasons, suggesting that these responses, although similar, were not invariable, i.e. 1 function could not be fitted for a given genotype across all seasons. Hence, the efficiency of N use for grain production (NUE_g) varied among seasons. In contrast, a study of winter wheat in the Netherlands by Groot (1987) found the relation between N uptake and yield to be invariable for 3 sites that showed large differences in the N applied/grain yield response curves.

NUE_g was highly negatively correlated ($r^2 = 0.99^{**}$) with the average daily mean temperature (°C, ADMT) during the 30 days before anthesis (reproductive period), such that ADMTs below 26·6°C were required to obtain NUE_g values >0·5 kg/kg. This suggests that nitrogen was used more effectively by the rice crop to produce grain compared with non-grain parts when ADMTs were lower during the reproductive period. This is highlighted by the negative correlation ($r^2 = 0.91^*$) between N HI (N uptake in grain/total N uptake) and ADMT during the reproductive period. For example in Starbonnet, N uptake in the straw and grain, respectively, was 62 and 95 kg/ha in Expt 1 when the ADMT during the reproductive period was 25·8°C, compared with 93 and 76 kg/ha in Expt 3 when the ADMT was 27·8°C over the same period. The highly negative correlation ($r^2 = 0.99^{**}$) between NUE_{hi} and ADMT during the reproductive period further supports this hypothesis.

Effect of nitrogen rate

Three factors varied in the relation between N applied and N uptake. Firstly, there was a trend for higher N uptake in the absence of fertiliser application (supply from soil-derived sources) in the wet than the dry season (91 *v.* 77 kg/ha). The contribution of soil N to crop growth in our experimental plots compares with 50–70 kg N/ha reported for rice in Japan (Shiga 1976), 46–65 kg N/ha in Indonesia (Ismunadji *et al.* 1973), and 92 kg N/ha for rice in southern Australia (Bacon and Heenan 1987).

Secondly, the slope of the relation between N applied and N uptake (recovery fraction) was less for N rates >140 kg/ha, particularly in Expt 2 (wet

season, Fig. 1c). In other words, the proportional relationship of total N uptake (kg/ha) to fertiliser N applied (kg/ha) declined from 1·97 to 0·85 as N rate increased from 70 to 280 kg/ha. This suggests that the plant system was approaching saturation, possibly resulting in active exclusion of nitrogen (Prins et al. 1981); alternatively, greater N losses from fertiliser application may have occurred.

Thirdly, genotypic variation in the relation between N applied and N uptake was evident only in the dry season. For a given level of N application in this season, N uptake was higher in Newbonnet than in Starbonnet than in Lemont. The parallel nature of the curves indicates that Newbonnet, compared with Starbonnet and Lemont, was able to extract more N from the soil at low and high levels of N supply.

Nitrogen HI decreased with increasing N rate, regardless of genotype and season. The ratios were 0·62, 0·65, 0·59, 0·54, and 0·52, respectively, for fertiliser N rates of 0, 70, 140, 210, and 280 kg/ha. These values are very similar to those reported by Guindo et al. (1994) for Lemont and Lebonnet which decreased from 0·65 to 0·53 as N rate increased from 88 to 220 kg/ha. Guindo et al. (1994) suggested that this variation may be due to either partial translocation or loss of N from rice foliage.

Effect of plant type

The slope of the linear phase of the regression between N uptake and LAI at anthesis was similar for all genotypes in the dry season (about 0·038 units/kg N·ha, Fig. 2d), suggesting a constant leaf area production per unit N uptake over this range. However, for uptakes >200 kg N/ha, leaf area production per unit N uptake was less in Newbonnet than Starbonnet, fitting the criterion of Schnier et al. (1990) for a plant type with modified leaf expansion patterns. In a study of N dynamics in tropical lowland rice in the Philippines, Schnier et al. (1990) concluded that a plant type with modified leaf area expansion patterns is required for optimum growth and high yields of tropical direct-seeded flooded rice. They found that excessive leaf area production resulted in foliage N dilution and, combined with increased biomass, adversely affected grain yield due to an unfavourable assimilation–respiration balance during ripening. Guindo et al. (1994) also reported that the differential N rate response between Lebonnet (tall-statured) and Lemont (short-statured) was associated with excessive plant growth of Lebonnet at high N rates rather than uptake or translocation of fertiliser N.

In a pooled analysis across all seasons and N rates, AGNU at maturity was higher in Newbonnet (184 kg/ha) than Lemont and Starbonnet (160 and

169 kg/ha). The similarity in AGNU at maturity for Lemont and Starbonnet is surprising considering the different plant types, i.e. early-maturing and short-statured v. late-maturing and tall-statured. Norman et al. (1992) also noted a lack of difference in total N uptake at maturity in 2 very different rice plant types (Lemont and Lebonnet).

Genotypic differences in the relation between N uptake and AGDM at maturity were observed in both seasons, with the medium- and tall-statured genotypes (Newbonnet and Starbonnet) producing consistently more dry matter per unit of N uptake than the short-statured genotype (Lemont), particularly in the dry season. Therefore, Newbonnet and Starbonnet exceeded Lemont in NUE_{dm} at maturity in both seasons, indicating scope for genetically improving nitrogen use efficiency.

As the N uptake required to reach a plateau in yield response was usually similar for all genotypes within a season (except for Expt 1), yield variation was dependent on variation in the efficiency of nitrogen use for grain production (kg grain/kg total above-ground N uptake, NUE_g). Hence NUE_g was greatest for Newbonnet and least for Starbonnet since Newbonnet had the highest yield and Starbonnet the least. Higher NUE_g in Newbonnet may be associated with higher ($P < 0·05$) SLN in this genotype (1·62 g N/m^2 at anthesis) compared with Lemont and Starbonnet (1·22 and 1·33 g N/m^2 at anthesis, respectively). Sinclair and Horie (1989) found that variation among maize, rice, and soybean species in the response of radiation use efficiency to specific leaf nitrogen (SLN) was compatible with the greater nitrogen use efficiency that has been observed in C_4 than in C_3 species (Brown 1978; Schmitt and Edwards 1981; Brown 1985).

Genotypic variation in fertiliser N uptake efficiency (AGNU/fertiliser N applied) was also observed, with Newbonnet exceeding Lemont (1·35 v. 1·22) and Starbonnet being intermediate (1·28). There were no genotypic differences in fertiliser N uptake efficiency for an application of 70 kg N/ha, yet Newbonnet exceeded Lemont and Starbonnet for applications of 140 and 280 kg N/ha, although the genotype×treatment interaction was not significant at $P = 0·05$. This indicates that Newbonnet, compared with Lemont and Starbonnet, was relatively more efficient at utilising fertiliser N at higher rates. Wescott et al. (1986) found Lemont to have a total N uptake of 125 kg N/ha when 100 kg N/ha was applied under dry-seeded conditions, i.e. fertiliser N uptake efficiency of 1·25. Norman et al. (1992) found Lemont had a total N uptake of 177 kg N/ha for an application of 134 kg N/ha (split applications of 67 kg N/ha at preflood and panicle differentiation), i.e. fertiliser N uptake efficiency of

$1\cdot32$. In this study, when 70, 140, 210, and 280 kg N/ha were applied to Lemont, the respective fertiliser N uptake efficiencies were $1\cdot94$, $1\cdot23$, $0\cdot93$, and $0\cdot78$.

Genotypic variation in the partitioning of N between grain and non-grain parts was observed in all experiments. Nitrogen harvest index (N uptake in grain/total N uptake) was higher in Lemont and Newbonnet ($0\cdot60$ and $0\cdot62$, respectively) compared with Starbonnet ($0\cdot53$). These values are lower than those reported by Westcott *et al.* (1986) for Lemont, which ranged from $0\cdot68$ to $0\cdot74$. Guindo *et al.* (1994) found N harvest index in short-statured Lemont ($0\cdot62$) to be higher than in tall-statured Lebonnet ($0\cdot58$). However, the current study indicates that medium-height genotypes such as Newbonnet can have N harvest indices as high as short-statured genotypes such as Lemont.

Linkages among the functional components of yield

Improving yield by increasing one or more of the functional components of yield (N uptake, NUE_{dm}, and HI) assumes the independence of components (Richards 1987). Attaining further yield increases in flooded rice depends on the extent to which genetic linkages between N uptake and both NUE_{dm} and HI can be broken. Our studies provide evidence that some of these linkages among components can be broken. For example, the fact that HI did not decline with increasing N uptake in Lemont and, to a lesser extent, in Newbonnet (see Expt 1, Fig. 6) suggests that HI does not always decline with increasing N uptake. Furthermore, despite similar negative correlations between N uptake and NUE_{dm} in all genotypes, the y-axis intercept was higher in Newbonnet and Starbonnet compared with Lemont. The example of Newbonnet shows that, to some extent, it is possible to increase yield by increasing each of the functional components independently within a specific genotype.

Conclusions

Seasonal differences in N uptake, NUE_{dm}, and HI were observed. NUE_g and NUE_{hi} were both highly correlated ($r^2 = 0\cdot99^{**}$) with the average daily mean temperature (°C) during the 30-day period before anthesis, suggesting that nitrogen was used more effectively by the rice crop to produce grain compared with non-grain parts when ADMTs were lower during the reproductive period (panicle initiation to anthesis). Seasonal differences appeared to affect the partitioning of dry matter between grain and non-grain parts more than it affected the production of dry matter *per se*. Some variation in the response of grain yield to N uptake was also observed among seasons, indicating that these responses were not invariable.

Three factors varied in the relation between N applied and N uptake. Firstly, there was a trend for higher N uptake from soil-derived sources in the wet than the dry season. Secondly, the recovery fraction was less for N rates above 140 kg/ha, i.e. the fertiliser N uptake efficiency declined from $1\cdot97$ to $0\cdot85$ as N rate increased from 70 to 280 kg/ha. Thirdly, genotypic differences were observed in this relation in the dry season such that N uptake was higher in Newbonnet than in Starbonnet than in Lemont.

Genotypic variation was found in N uptake, NUE_{dm}, and HI. The capacity to capture these components in crop improvement programs depends on the extent to which genetic linkages between N uptake and both NUE_{dm} and HI can be broken. High grain yields in Newbonnet were associated with increased nitrogen uptake and nitrogen use efficiency, particularly in the dry season. In this season, the greater response of yield to N fertilisation in Newbonnet was due to both increased N uptake when N was limiting (i.e. at zero N application) and increased uptake at high N rates due to increased growth. In the harvest index–AGNU regression, Newbonnet had a significantly higher intercept, and therefore higher HI, than Starbonnet. This suggests that, to some extent, the negative linkages between nitrogen uptake and both HI and NUE_{dm} can be broken, providing an avenue for further improving yield in nitrogen-limiting environments.

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