CSIRO PUBLISHING

Australian Journal of Agricultural Research

Volume 50, 1999

A journal for the publication of original contributions
towards the understanding of an agricultural system

www.publish.csiro.au/journals/ajar

All enquiries and manuscripts should be directed to
Australian Journal of Agricultural Research
CSIRO PUBLISHING
PO Box 1139 (150 Oxford St)
Collingwood
Vic. 3066
Australia
Telephone: 61 3 9662 7628
Facsimile: 61 3 9662 7611
Email: jenny.fegent@publish.csiro.au

Published by **CSIRO** PUBLISHING
for CSIRO Australia and
the Australian Academy of Science

Aust. J. Agric. Res., 1999, **50**, 1399–408

Grain quality of flooded rice is affected by season, nitrogen rate, and plant type

A. K. Borrell[AD], *A. L. Garside*[AE], *S. Fukai*[B], *and D. J. Reid*[C]

[A] Department of Primary Industries, PO Box 591, Ayr, Qld 4807, Australia.
[B] The University of Queensland, School of Land and Food, St Lucia, Qld 4072, Australia.
[C] Department of Primary Industries, Townsville, Qld 4814, Australia. Present address: Department of Primary Industries, Rockhampton, Queensland 4702, Australia.
[D] Present address: Hermitage Research Station, Department of Primary Industries, Warwick, Qld 4370, Australia.
[E] Present address: Sugar Yield Decline Joint Venture, BSES, C/- CSIRO Davies Laboratory, PMB Aitkenvale, Townsville, Qld 4814, Australia.

Abstract. Quality of grain, next to yield, is the most important factor for rice (*Oryza sativa* L.) production in semi-arid tropical Australia. Studies were undertaken in the Burdekin River Irrigation Area of northern Australia to improve rice grain quality through nitrogen fertilisation. This paper reports the results of 4 experiments comparing the response of 3 rice genotypes differing in maturity and stature to 5 rates of applied nitrogen (0, 70, 140, 210, and 280 kg/ha) over 4 seasons (2 wet and 2 dry seasons). The components of grain quality studied were endosperm chalkiness, whole grain millout, grain size, alkali digestion (gelatinisation temperature), and grain protein. This paper also examines the suitability of the 3 genotypes as parental material in breeding programs aimed at selecting for specific grain quality attributes.

Starbonnet was identified as a potential parent in breeding programs which aim to specifically select for reduced chalkiness and high millout in low N environments. Selection for lower chalkiness, and higher millout and protein concentration, should occur in a wide range of target environments to account for the seasonal variation observed in these parameters. Grain size appeared to be affected more by genetic than agronomic factors, since grain length and breadth were largely unaffected by N rate, yet genotypic differences were found for both parameters in all experiments. Newbonnet grain was long and slender, suggesting this genotype would be a suitable parent in breeding programs aimed at improving grain appearance. The response of alkali digestion to N rate and genotype was small for all seasons.

The importance of developing N fertiliser strategies that optimise both grain yield and quality was highlighted by differences in the responses of grain protein and grain yield to N rate. A number of linkages were examined among various components of grain quality. However, the magnitude of these linkages was small, suggesting that selection for one quality component should not be at the expense of selection for another.

Introduction

Quality of grain, next to yield, has historically been the most important factor for rice production in northern Australia. Traditionally, high-quality long-grain rices have been produced in the Burdekin River Irrigation Area (BRIA) of northern Australia.

Although rice is the most important basic food in the world, it is relatively unimportant in world commerce. Most rice is consumed where it is grown, with >5% of total world production traded internationally (Efferson 1985). Other food grains, particularly wheat and maize, are traded in much larger volumes. This small international market has relatively few buyers and sellers, and its supply and demand is relatively unpredictable. Unlike other major cereals, the consumer views rice in the unprocessed or uncooked state, making judgements about its quality.

Some of the key components of rice quality are grain appearance, whole grain millout, grain length and breadth, alkali digestion (gelatinisation temperature), and grain protein. Grain appearance is mainly determined by endosperm opacity and the degree of chalkiness, either on the dorsal side of the grain (white belly) or in the centre (white centre) (Khush *et al.* 1979). Endosperm chalkiness disappears with cooking and does not directly affect cooking or eating quality (Ikehashi and Khush 1979). However, chalkiness does influence consumer preference and affects milling

10.1071/AR98054 0004-9409/99/081399

recovery. Chalky areas are softer than translucent areas since the starch granules in the chalky areas are less densely packed (del Rosario *et al*. 1968). Therefore, grains with chalkiness are more likely to break during milling, affecting the commercial value of milled rice (Ikehashi and Khush 1979), and lowering market acceptability (Khush *et al*. 1979).

Whole grain millout (alternatively 'head rice' or 'milling recovery') is the proportion of unbroken polished grain that remains after paddy rice has been milled. Seetanun and De Datta (1973) found that an application of N at heading increased the whole grain millout due to the higher protein concentration. In a study of *indica* cultivars, an application of up to 120 kg N/ha in the dry season increased the whole grain millout of 3 chalky cultivars, but not that of a non-chalky cultivar (Nangju and De Datta 1970). In *japonica* rice, chalkiness reduced whole grain millout (Kanda *et al*. 1969).

Grain length is another important quality character since there is strong demand for long-grain rice in the world market (Khush *et al*. 1979). In addition, the grain length : breadth ratio is an important measure of overall grain shape, indicating the slenderness of grain.

The time required to cook rice depends on the gelatinisation temperature, a physical property of starch, and is defined as the range of temperature in which the starch granules begin to swell irreversibly in hot water (Khush *et al*. 1979). There appears to be a distinct consumer preference for rices with intermediate gelatinisation temperature.

Rice is one of the most nutritious of all cereals, yet the protein concentration of milled rice is relatively low (about 7% at 14% moisture) (Nanda and Coffman 1979). Even a modest increase in rice protein levels would provide a significant nutritional boost to the hundreds of millions of people who depend upon it (Gomez 1979). Nitrogen fertilisation generally increases the protein concentration in rice grain (De Datta *et al.* 1969; Nangju and De Datta 1970; De Datta *et al.* 1972; Cheang and Mohan Rao 1972), and protein also increases with better water management and weed control, probably due to the higher efficiency of N utilisation associated with improved water and weed management (Gomez and De Datta 1975).

The various components of rice quality are affected by season, N rate, and genotype. For example, environmental factors strongly influence the protein concentration of rice grain, accounting for a major portion of total variability (Gomez 1979). Similarly, environmental factors, particularly those that interrupt normal grain filling, greatly influence the development of chalkiness (Tashiro and Ebata 1975). Nitrogen fertilisation is known to affect endosperm chalkiness (Lewin *et al.* 1994), whole grain millout (Nangju and De Datta 1970; Seetanun and De Datta 1973), and grain protein (De Datta *et al*. 1969; Cheang and Mohan Rao 1972). Genotypic variation has also been reported for chalkiness (Lewin *et al.* 1994), millout (Lewin *et al.* 1994), grain length and breadth (Webb *et al.* 1985), alkali digestion (Khush *et al.* 1979; Bhattacharya 1979), and grain protein (Gomez 1979).

This paper examines how season, N rate and plant type affect the response of grain quality in 3 rice genotypes to 5 rates of N fertilisation in a semi-arid tropical environment. The components of grain quality examined were chalkiness, whole grain millout, grain size, alkali digestion (gelatinisation temperature), and grain protein. In particular, this paper examines genotypic variation in these components, recommending which genotypes may be useful parents when selecting for specific grain quality attributes in breeding programs.

Materials and methods

Agronomy

Agronomic practices used in the experiment are described in Borrell *et al*. (1998*a*). Summarising, 4 experiments were conducted in 4 consecutive seasons at Millaroo Research Station, BRIA (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 each experiment, the design was a randomised split-plot design with 5 rates of N fertiliser (0, 70, 140, 210, and 280 kg/ha) 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.

In Expts 1, 2 and 4, quadrats were cut from all genotypes on the same day after the attainment of physiological maturity in the late-maturing genotype (Starbonnet). No yield loss was observed in the early-maturing genotypes (Lemont and Newbonnet) due to this harvesting procedure (Borrell *et al*. 1998*a*). In Expt 3, however, Lemont, Newbonnet, and Starbonnet were harvested at physiological maturity (123, 130, and 137 days after sowing, respectively). Physiological maturity was defined as the time when 90% of the florets were yellow. Therefore, Starbonnet was harvested immediately after physiological maturity in all experiments, while Lemont and Newbonnet were harvested immediately after physiological maturity in Expt 3 only, and from 7 to 14 days after physiological maturity in Expts 1, 2, and 4. According to Lewin *et al*. (1994), delaying harvest of long grain rices by a 10-day period had little effect on whole grain millout or chalkiness, although delaying harvest of medium grain rices by 40 days had a marked effect. Lemont, Newbonnet, and Starbonnet are all classified as long grain.

Chalkiness of endosperm

Chalkiness was evaluated on 100 milled grains per plot. Grains containing 50% or more of white belly, white centre, white back, or a combination of these were considered chalky.

Whole grain millout

A 150-g sample of paddy rice was milled from each plot using a McGill No. 2 mill (Adair 1952). The paddy was twice passed through a Satake dehusker, polished, then separated into broken, and unbroken grain. The husk, bran, and broken and unbroken grain were weighed. The unbroken grain was expressed as a percentage of the total sample of paddy (whole grain millout).

Grain size

The length and breadth of 5 milled grains from each plot were determined using a dissecting microscope. Grain length was classified according to the Standard Evaluation System for Rice (IRRI 1976): extra long (>7.50 mm); long (6.61–7.50 mm); medium (5.51–6.60 mm); and short (≤5.5 mm). The grain length : breadth ratio was determined from these measurements.

Alkali digestion

This test is undertaken as an indication of gelatinisation or cooking temperature (Little *et al.* 1958). Gelatinisation temperature refers to the first irreversible change in the starch when the crystalline structure is lost and the granule begins swelling. It is routinely measured as the loss of birefringence on a polarising microscope (Schoch and Mayward 1956) and called Birefringent Endpoint Temperature (BEPT). Final BEPT is the temperature at which 90% of the starch granules have gelatinised or swollen irreversibly in hot water, and is classified as low (55–69.5°C), intermediate (70–74°C), and high (74.5–79°C) (Juliano 1979). Gelatinisation temperature varies with genotype and was found to range from 55 to 79°C in a world collection (Juliano 1985).

Six milled rice kernels from each treatment were placed in a solution of 1.7% potassium hydroxide for 24 h, then scored for spreading according to the Standard Evaluation System for Rice (IRRI 1976): 1, not affected but chalky, low alkali digestion, high gelatinisation temperature; 3, swollen with collar incomplete and narrow, low/intermediate alkali digestion, high/intermediate gelatinisation temperature; 5, split or segmented with collar complete and wide, intermediate alkali digestion, intermediate gelatinisation temperature; and 7, completely dispersed and cleared, high alkali digestion, low gelatinisation temperature. Alkali digestion was not recorded in Expt 4 since there was little variation observed in Expts 2 and 3.

Grain protein

Protein in the grain was calculated by multiplying the concentration of N in the grain by 5.95 (Gomez and De Datta 1975; Webb *et al.* 1985). The procedure for determining concentration of N in the grain is described in Borrell *et al.* (1998*b*).

Statistical analysis

Data were analysed by standard analysis of variance and pairwise comparisons of means were performed using the protected l.s.d. procedure at $P = 0.05$. Relations were investigated between endosperm chalkiness and both N concentration and content in the grain in Expt 1, and between whole grain millout and N content in the grain in all experiments. Relations between endosperm chalkiness and both grain protein and grain breadth, and between whole grain millout and grain protein for each genotype were also investigated. Furthermore, relations between average daily mean temperature during the 30 day period after anthesis and endosperm chalkiness, whole grain millout, grain length, grain breadth, grain length : breadth ratio, alkali digestion and grain protein were considered.

In an attempt to explain seasonal variation in grain quality components, the average daily mean temperature (ADMT, °C) for the 30-day period after anthesis (grain filling period) was regressed against each component of grain quality. This concept was based on earlier work by Yoshida and Parao (1976), who found rice grain yield in the tropics to be highly negatively correlated with ADMT during the 25-day period before anthesis, although the current application relates to the effect of temperature during the grain filling period on grain quality, rather than the effect of temperature during the reproductive period on grain yield. That environmental factors, particularly those interrupting normal grain filling, should affect some grain quality components has already been established by Tashiro and Ebata (1975).

Results

Seasonal variation

Of the grain quality components measured in this study, grain breadth in Lemont was the only component correlated ($r = -0.96$) with ADMT (°C) during the 30-day period after anthesis.

Endosperm chalkiness

The response of chalkiness to N fertilisation appeared to differ across experiments (Table 1). A significant interaction between N rate and genotype was only observed in Expt 1. In that experiment (dry season), there was no response to N rate in Starbonnet while in Lemont chalkiness was greater ($P < 0.05$) in the nil N rate compared with the applied N treatments. In Newbonnet, chalkiness decreased ($P < 0.05$) for N rates up to 140 kg N/ha. There was no response ($P > 0.10$) in chalkiness to applied N for Expts 2, 3, and 4. In both dry season experiments (Expts 1 and 3), chalkiness was greater ($P < 0.05$) in Newbonnet than Lemont and Starbonnet (Table 1).

To interpret the interaction between N fertilisation and genotype in Expt 1, relations between chalkiness and both N concentration and content in the grain were investigated. Chalkiness in Newbonnet declined exponentially with N concentration (Fig. 1*a*; $R^2 = 0.75$) and N content (Fig. 1*b*; $R^2 = 0.59$) so that N contents of 120 kg/ha (or N concentration in the grain of about 1.2%) were required to reduce chalkiness to <5%. However, neither N concentration nor content influenced chalkiness in Starbonnet and Lemont.

Whole grain millout

Fertiliser treatment interacted ($P < 0.01$) with genotype for whole grain millout in Expts 1, 3, and 4, with millout in Starbonnet and, to a lesser extent Newbonnet, relatively unaffected by N fertilisation, while millout increased with N rate in Lemont (Fig. 2).

Whole grain millout in Lemont was related to N content in the grain during the dry and wet seasons (Fig. 3), highlighting the importance of adequate grain N content to grain quality in this genotype. The affect of environmental conditions during the grain filling period on millout percentage in Lemont is emphasised by the fact that N uptakes in the grain of about 40 and 120 kg/ha were required in Expts 3 and 1, respectively, to attain an equivalent whole grain millout of 50% (Fig. 3*b* and 3*a*). This was not evident for Lemont in the wet season with N uptakes in the grain of about 80 and 90 kg/ha for Expts 2 and 4, respectively, to attain a whole grain millout of 50%. Millout for Newbonnet in the dry season was not affected by N uptakes in the grain but increased exponentially with grain N in the wet seasons (Fig. 3), attaining maximum millouts at grain N contents of about 90 kg/ha. Regardless of season, millout in Starbonnet was independent of grain N content.

Table 1. Effect of five rates of nitrogen on rice grain quality of three genotypes across four experiments

Means within a significant treatment effect not followed by a common letter are significantly different ($P < 0.05$)

	Endosperm chalkiness (%)	Grain length (mm)	Grain breadth (mm)	Grain length : breadth ratio	Alkali digestion	Grain protein (%)
			Expt 1 (dry season)			
Rate of N (kg/ha)	***	$P = 0.055$	n.s.	*	*	***
0	7.42a	7.02	2.22	3.17bc	3.02bc	6.54c
70	3.83b	7.26	2.25	3.23ab	3.09ab	6.40c
140	2.67bc	7.18	2.19	3.28a	3.00c	7.67b
210	2.50bc	7.19	2.27	3.18bc	3.12a	8.60a
280	2.00c	7.03	2.24	3.14c	3.00c	9.18a
Genotype	***	***	***	*	***	$P = 0.072$
Lemont	3.30b	7.30a	2.34a	3.13b	3.00b	7.86
Newbonnet	6.10a	7.19a	2.23b	3.24a	3.12a	7.62
Starbonnet	1.65c	6.92b	2.14c	3.24a	3.02b	7.55
			Expt 2 (wet season)			
Rate of N (kg/ha)	n.s.	n.s.	n.s.	n.s.	n.s.	***
0	5.83	7.28	2.17	3.36	3.00	7.33d
70	5.83	7.25	2.18	3.33	3.02	7.93d
140	7.00	7.13	2.19	3.26	3.00	8.77c
210	6.08	7.13	2.20	3.26	3.03	9.60b
280	4.92	7.08	2.17	3.27	3.00	10.51a
Genotype	n.s.	**	***	***	n.s.	n.s.
Lemont	5.10	7.28a	2.29a	3.19b	3.01	8.78
Newbonnet	6.65	7.18ab	2.15b	3.34a	3.02	8.77
Starbonnet	6.05	7.07b	2.11b	3.36a	3.00	8.93
			Expt 3 (dry season)			
Rate of N (kg/ha)	n.s.	n.s.	n.s.	n.s.	N/A	***
0	7.00	7.24	2.13	3.41	3.00	6.53c
70	9.75	7.30	2.17	3.38	3.00	6.83c
140	6.92	7.26	2.15	3.38	3.00	7.61b
210	5.08	7.25	2.13	3.41	3.00	8.34a
280	7.00	7.22	2.14	3.38	3.00	8.64a
Genotype	**	***	***	***	N/A	***
Lemont	5.70b	7.48a	2.26a	3.31b	3.00	8.03a
Newbonnet	10.65a	7.21b	2.10b	3.43a	3.00	7.47b
Starbonnet	5.10b	7.07c	2.06c	3.44a	3.00	7.27b
			Expt 4 (wet season)			
Rate of N (kg/ha)	n.s.	n.s.	**	n.s.		***
0	8.08	7.33	2.24a	3.27		6.96cd
70	6.25	7.29	2.23a	3.28		6.67d
140	5.67	7.23	2.21a	3.28		7.13c
210	5.17	7.25	2.17b	3.35		7.67b
280	5.17	7.27	2.17b	3.36		8.40a
Genotype	n.s.	***	***	***		*
Lemont	6.60	7.43a	2.31a	3.22c		7.57a
Newbonnet	6.15	7.26b	2.14b	3.39a		7.66a
Starbonnet	5.45	7.14b	2.16b	3.31b		6.88b

n.s., not significant ($P > 0.10$); * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$; N/A, not analysed (all data were 3).

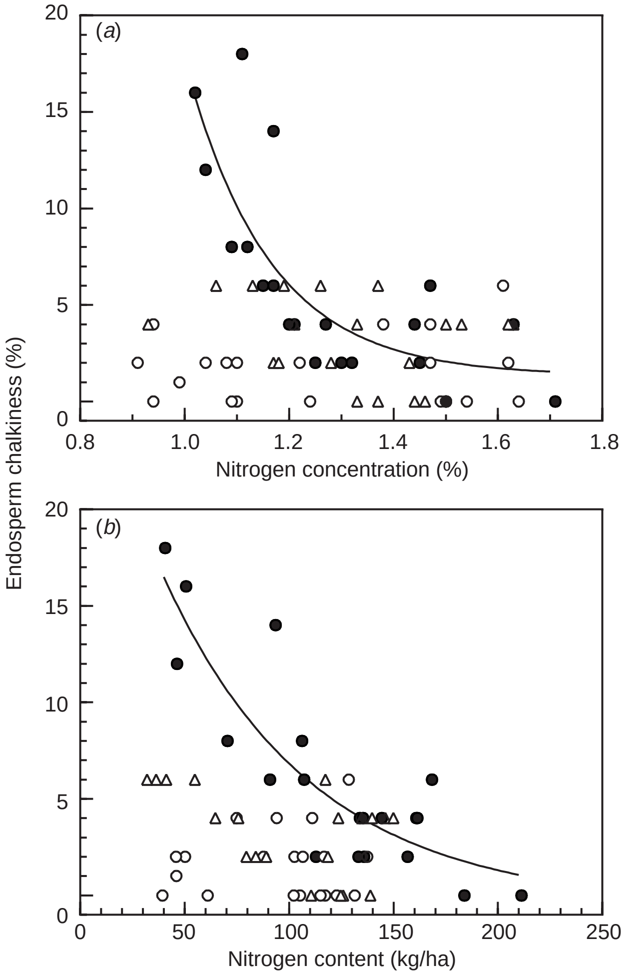

Fig. 1. Relation between endosperm chalkiness (%) in grain and (*a*) nitrogen concentration in grain, and (*b*) nitrogen content in grain for Lemont (Δ), Newbonnet (●) and Starbonnet (○) in Expt 1. Exponential curves were fitted to the data of Newbonnet.

Grain size

There was no interaction ($P > 0.05$) between N fertilisation and genotype for grain length, grain breadth and length : breadth ratio in all experiments.

Grain length (mm) in all experiments differed ($P < 0.01$) among genotypes, yet was unaffected ($P > 0.10$) by N fertilisation (Table 1). Grain was longest in Lemont, intermediate in Newbonnet, and shortest in Starbonnet. Indeed, the average length of Lemont grains even approached the extra long category (> 7.50 mm; IRRI 1976) in one dry season (Expt 3).

In 3 of 4 experiments, grain breadth did not respond ($P > 0.10$) to N fertilisation. However in Expt 4, grain breadth declined from around 2.23 mm (2.21–2.24 mm) for N rates of 0–140 kg/ha to 2.17 mm for N rates of 210 and 280 kg/ha (Table 1). Genotypic differences ($P < 0.01$) in grain breadth were observed in all experiments, with grain being broader in Lemont than in Newbonnet than in Starbonnet in the dry seasons while in both wet seasons, grain was broader in Lemont than in Newbonnet or Starbonnet.

Slenderness of grain is determined by the length : breadth ratio, i.e. grain with a high ratio is more slender than grain with a low ratio. This ratio was unaffected ($P > 0.10$) by N fertilisation in 3 of 4 experiments (Table 1). However in Expt 1, grain was more slender ($P < 0.05$) with an application of 140 kg N/ha compared with applications of 0, 210, or 280 kg N/ha. The ratio was also higher ($P < 0.05$) in Newbonnet and Starbonnet compared with Lemont in all experiments.

Alkali digestion

Variability in alkali digestion, an indicator of gelatinisation temperature, was limited. In Expt 1 values ranged from 3 (low/intermediate alkali digestion, high/intermediate gelatinisation temperature) to 5 (split or segmented with collar complete and wide, intermediate alkali digestion, intermediate gelatinisation temperature) with most samples rated as 3. An interaction ($P < 0.01$) between fertiliser and genotype was observed in this experiment with no response to N for Lemont and Starbonnet while alkali digestion differed among levels of N for Newbonnet. In Expts 2 and 3, all except 2 samples in Expt 2 were rated as 3 and hence no differences among genotypes or fertiliser treatments were observed in these experiments. Alkali digestion was not measured in Expt 4 due to the limited variability observed in the earlier experiments.

Grain protein

A treatment × genotype interaction ($P < 0.01$) was only observed in Expt 1. For N rates between 70 and 280 kg/ha, grain protein increased at a higher rate in Starbonnet compared with Lemont and Newbonnet. In all experiments, grain protein increased ($P < 0.001$) with N rate (Table 1). In the dry seasons (Expt 1 and 3) no significant increase in grain protein was observed for rates greater than 210 kg N/ha, while in the wet seasons (Expts 2 and 4) grain protein increased to 280 kg N/ha.

Differences among genotypes in grain protein were significant in Expts 3 ($P < 0.01$) and 4 ($P < 0.05$) only (Table 1). Grain protein was higher in Lemont (8.03%) compared with Newbonnet (7.47%) and Starbonnet (7.27%) in Expt 3, and higher in Lemont (7.57%) and Newbonnet (7.66%) compared with Starbonnet (6.88%) in Expt 4.

Linkages among quality components

No clear relationships between whole grain millout (%) and grain protein (%) or endosperm chalkiness were evident for any genotype although millout in Lemont tended to increase with increasing protein. Similarly, no relationships were evident between endosperm chalkiness and grain protein (%) or grain breadth although endosperm chalkiness in Newbonnet tended to decrease with both increasing protein (%) and grain breadth.

Discussion

Endosperm chalkiness

Chalkiness was higher in Newbonnet compared with Lemont and Starbonnet in the dry but not wet seasons. Of particular interest, genotypes responded differently at low rates of N in Expt 1 (dry season), indicating that endosperm chalkiness is likely to be lower in Starbonnet, and to a lesser extent in Lemont, compared with Newbonnet when N is limiting growth in the dry season. Even when grain N concentration fell below 1%, chalkiness remained low in Starbonnet (1–2%). Conversely, endosperm chalkiness greatly increased in Newbonnet from about 3.5% to >15% as grain N concentration fell from 1.3% to around 1% (Fig. 1). Ikehashi and Khush (1979) found evidence that chalkiness in rice is under genetic control, and selection for less chalky grain is worthwhile. Lewin *et al.* (1994) also reported genotypic variation in chalkiness at low N rates. In a study of 2 medium grain and 3 long grain cultivars under low (50 kg N/ha) and high (150 kg N/ha) rates of N application, they found endosperm chalkiness was less in Jarrah than Amaroo (medium grains), particularly at the lower N rate. Therefore, rice-breeding programs in low-input environments with a specific objective of reducing grain chalkiness could utilise Starbonnet as a parent. Selection for endosperm chalkiness should also occur in a wide range of seasons and target environments to account for the seasonal variability observed in the current study.

Whole grain millout

Whole grain millout varied across experiments in Lemont, and to a lesser extent in Newbonnet, while Starbonnet was largely unaffected by experiment. Large differences in the grain nitrogen content–whole grain millout

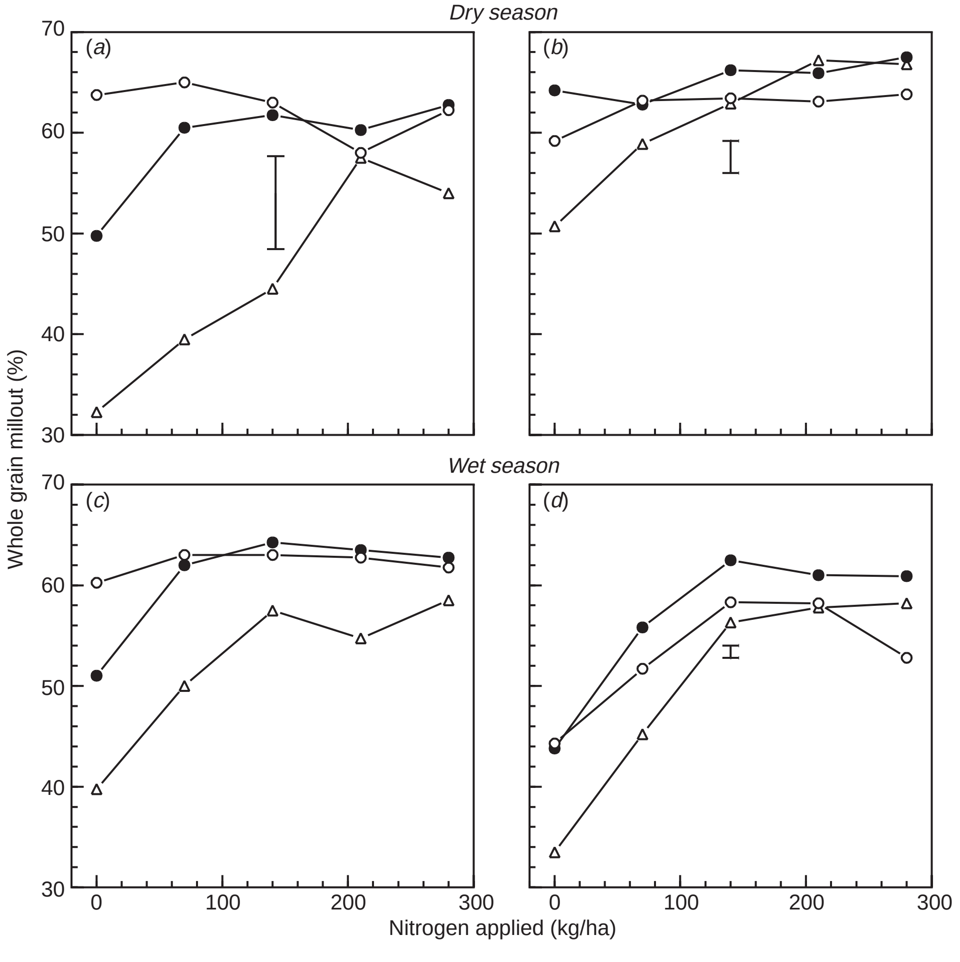

Fig. 2. Relation between nitrogen applied and whole grain millout in 2 dry seasons (*a*) Expt 1 and (*b*) Expt 3 and 2 wet seasons (*c*) Expt 2 and (*d*) Expt 4 for 3 rice genotypes: Lemont (Δ), Newbonnet (●) and Starbonnet (○). Vertical bars denote l.s.d. of interaction ($P = 0.05$).

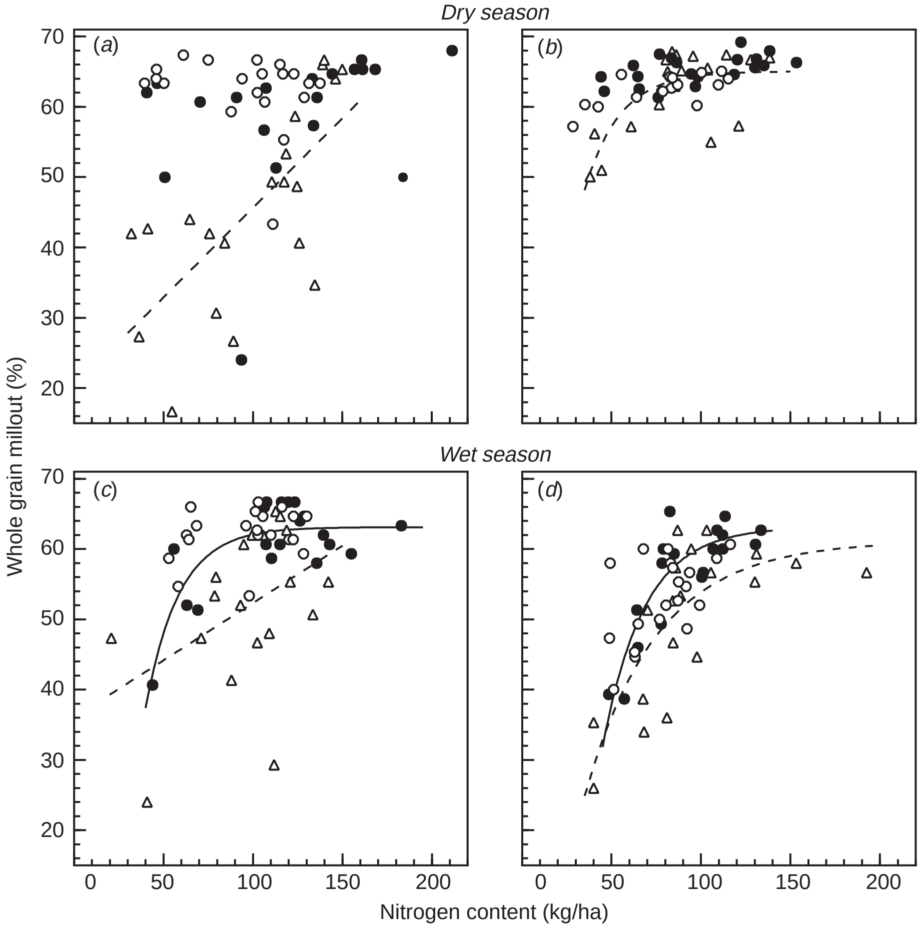

Fig. 3. Relation between whole grain millout (%) and nitrogen content in grain in 2 dry seasons (*a*) Expt 1 and (*b*) Expt 3 and 2 wet seasons (*c*) Expt 2 and (*d*) Expt 4 for 3 rice genotypes: Lemont (Δ, - - -), Newbonnet (●, —) and Starbonnet (○). Linear and exponential regressions were fitted to the data as appropriate.

regression for Lemont between Expts 1 and 3 highlight such seasonal variation. These environmental or climatic effects are emphasised by the different levels of grain N required in Expts 1 and 3 for Lemont to achieve millouts of 50% (120 and 40 kg/ha, respectively). Webb *et al.* (1985) noted that some quality characteristics vary greatly with location, and concluded that rice varieties may be area- and/or environment-specific in quality attributes. This work suggests that quality attributes of certain genotypes may also be affected by seasonal variation within the same environment.

In 3 of 4 seasons, N rate and genotype interacted such that millout in Starbonnet, and to a lesser extent Newbonnet, remained high regardless of N rate (about 60%), yet increased with N rate in Lemont from about 40% (0 kg N/ha) to 60% (210 and 280 kg N/ha). Positive relations between N rate and millout have been found by Nangju and De Datta (1970), and Seetanun and De Datta (1973), particularly in chalky genotypes, and were also related to an increase in protein concentration of grain with increasing rate of N. Further evidence that millout increases with N rate is provided by Lewin *et al.* (1994). In a study of 5 rice genotypes in southern New South Wales, Australia, they observed that millout was consistently higher with an application of 150 compared with 50 kg N/ha, regardless of harvest date. In light of these combined findings, the retention of high millouts in Starbonnet at low N rates is important, and breeding programs selecting for increased millout under low levels of N supply may benefit from using Starbonnet as a parent. Interestingly, Place *et al.* (1970) reported the whole grain millout of Bluebonnet remained high (about 68%) regardless of rate of N.

Grain size

Genotype and N treatment responses in both grain length and breadth varied across experiments. In the case of Lemont, lower temperatures during the grain filling period were associated with broader but less slender grain, and ADMTs below 27.5°C resulted in grain breadths >2.3 mm.

Resurreccion *et al.* (1977) observed that grain size (100-grain weight) was correlated with grain breadth, while Ebata and Nagato (1967) reported that high temperature during ripening resulted in a lower final grain weight for *japonica* rices.

Surprisingly, N fertilisation did not affect grain length in any experiment, or grain breadth in any experiment except Expt 4 where rates of ≥210 kg/ha reduced grain breadth. On the other hand, significant genotypic variation was observed for both grain length and breadth in all experiments, suggesting that grain size is more readily manipulated by genetic than agronomic factors. Grain of Lemont was longer than Starbonnet, but was less slender, and therefore less desirable in grain appearance. Newbonnet was intermediate in both grain length and breadth leading to the greatest length : breadth ratio. This slenderness is valuable for Newbonnet as there is a strong demand for such grain in the world market (Efferson 1985). This demand, together with the importance of genetic factors in grain size determination, suggest that Newbonnet would be a suitable parent in breeding programs aimed at developing long and slender grain for the world market. Genotypic variation for grain length, grain breadth and grain length : breadth ratio has also been found in typical commercial long-, medium- and short-grain types in the USA (Webb *et al.* 1968; Adair *et al.* 1973; Webb *et al.* 1979).

Alkali digestion

Alkali digestion was largely unaffected by N rate and genotype regardless of season. This result is in contrast to previous work showing that gelatinisation temperature varies with ambient temperature during grain development (Juliano *et al.* 1969; Resurreccion *et al.* 1977) and with genotype (Khush *et al.* 1979; Bhattacharya 1979).

Grain protein

Grain protein increased with N rate in all experiments, supporting earlier studies which have documented similar responses (De Datta *et al.* 1969; Nangju and De Datta 1970; Cheang and Mohan Rao 1972; De Datta *et al.* 1972), with the largest increases in grain protein occurring when N is applied at heading (De Datta *et al.* 1972; Seetanun and De Datta 1973). This highlights the dependence of grain protein on N supply, in contrast with other quality parameters such as grain length and breadth, which were largely unaffected by N fertilisation. Nitrogen fertiliser strategies need to be developed to optimise both grain yield and quality. For example, grain yield responded to the application of 70 kg N/ha in 3 of 4 experiments (Borrell *et al.* 1998*a*), yet grain protein increased to the highest N rate (280 kg/ha) in Expts 2 and 4 (wet seasons) and to the second-highest N rate (210 kg/ha) in Expts 1 and 3 (dry seasons).

The existence of genotypic variation in grain protein is well documented. For example, the brown rice protein concentration of 17 587 cultivars in the International Rice Research Institute world collection ranged from 4.3% to 18.2%, with a mean of 9.5% (Gomez 1979). Genotypic variation in grain protein was observed in Expts 3 and 4, and to a lesser extent in Expt 1 ($P = 0.07$), with Lemont exceeding Starbonnet in these seasons. No genotypic variation was found in Expt 2. Webb *et al.* (1985) found little difference in the grain protein levels of Lemont (7.0%) and Starbonnet (6.9%) when grown in their area of origin in the southern USA.

The environment has a strong influence on the protein concentration of rice grains, accounting for a major portion of the total variability (Gomez 1979). For temperatures >22°C, Resurreccion *et al.* (1977) found that grain protein increased with increasing temperature during grain ripening, and that temperature affected the accumulation of non-protein constituents, particularly starch, more than the accumulation of protein. These findings contrast with the current study, since no relationship was found between temperature during the grain filling period and grain protein.

Low protein is also associated with high solar radiation during grain development, and therefore protein is generally lower in the dry season than in the wet season (IRRI 1973; Gomez and De Datta 1975). In the current study, grain protein was highest in Expt 2 (wet season), intermediate in Expts 1 and 3 (dry seasons), and least in Expt 4 (wet season). As solar radiation was only measured during Expts 3 and 4, the hypothesis proposed by Gomez and De Datta (1975) that grain protein and solar radiation during grain development are negatively correlated could not be tested. However, the seasonal variation observed in the present study highlights the need to select for grain protein in a wide range of target environments.

Linkages among quality components

To what extent are the various components of grain quality linked? Since proteins occupy the space between compound starch granules, and starch granules in the chalky areas of the grain are not as firmly packed as those in the non-chalky areas (del Rosario *et al.* 1968), it is likely that protein molecules act as a binder, so that anything which increases the protein concentration of the grain (e.g. N fertilisation) increases the resistance of genotypes, particularly chalky ones, to breakage (Nangju and De Datta 1970). In this study, all plots were harvested on the same day, regardless of N rate, so low N plots may have been harvested at a lower grain moisture percentage compared with high N plots, which may have confounded moisture with protein when explaining changes in whole grain millout data. In relation to this issue, Lewin *et al.* (1994) noted that the rate of decline in whole grain millout highlights the need to sample carefully for milling quality, as results are very dependent on the stage of sampling.

No relation was observed between endosperm chalkiness and whole grain millout in any genotype. In contrast, del Rosario *et al*. (1968) observed that white belly grains tend to crack more under stress conditions, presumably due to their heterogenic texture. Kanda *et al.* (1969) also found the milling recovery of a *japonica* rice to be higher for grains without white belly compared to grains with white belly.

Neither were endosperm chalkiness and grain protein found to be correlated for any genotype, although chalkiness in Newbonnet tended to decrease with increasing protein. Srinivas and Bhashyam (1985) found that increasing the protein concentration of rice grain reduced the proportion of white belly.

Endosperm chalkiness and grain breadth were not related for any genotype in this study. Chalky grain is more often associated with broad than with slender grain of comparable length (Nakatat and Jackson 1973; Somrith 1974). In a study of 138 rice cultivars, Srinivas and Bhashyam (1985) found that chalkiness and grain breadth were positively related. For example in one cultivar, all grains were translucent when the breadth was 2 mm and almost all grains had white belly when the breadth was 2.8 mm. Cultivars of intermediate breadth displayed either translucence or white belly or a mixture. Grain length and grain length : breadth ratio were not related to the expression of white belly (Bhashyam and Srinivas 1981).

Although no clear relationships were observed between whole grain millout and grain protein for any genotype, millout in Lemont tended to increase with increasing protein. Similar positive trends between millout and grain protein have been observed by Nangju and De Datta (1970) and Seetanun and De Datta (1973).

In all the linkages examined, only a small proportion of the variation in a given component (e.g. endosperm chalkiness) could be explained by other components (e.g. grain protein concentration and grain breadth). This finding has implications for plant breeding programs utilising the genotypes considered in this study, suggesting that selection for a given grain quality component will be largely independent of other components.

Acknowledgments

This work was funded by the Rural Industries Research and Development Corporation and the North Queensland Rice Industry. The staff of Millaroo Research Station, together with Shane Wallace (Technical Assistant) and Trevor Rasmussen (Chemist) are thanked for their support.

References

Adair, C. R. (1952). The McGill miller method for determining the milling quality of small samples of rice. *Rice Journal* **55**, 21–3.

Adair, C. R., Bollich, C. N., Bowman, D. H., Jodon, N. E., Johnston, T. H., Webb, B. D., and Atkins, J. G. (1973). Rice breeding and testing methods in the United States. *In* 'Rice in the United States: Varieties and Production'. pp. 22–75. (US Department of Agriculture Handbook 289, revised edition.)

Bhashyam, M. K., and Srinivas, T. (1981). Studies on the association of white core with grain dimension in rice. *Journal of Food Science and Technology* **18**, 214–5.

Bhattacharya, K. R. (1979). Gelatinisation temperature of rice starch and its determination. *In* 'Proceedings of the Workshop on Chemical Aspects of Rice Grain Quality'. pp. 231–49. (International Rice Research Institute: Los Baños, Philippines.)

Borrell, A. K., Garside, A. L., Fukai, S., and Reid, D. J. (1998*a*). Season and plant type affect the response of rice yield to nitrogen fertilisation in a semi-arid tropical environment. *Australian Journal of Agricultural Research* **49**, 179–90.

Borrell, A. K., Garside, A. L., Fukai, S., and Reid, D. J. (1998*b*). Season, nitrogen rate and plant type affect nitrogen uptake and nitrogen use efficiency in rice. *Australian Journal of Agricultural Research* **49**, 829–43.

Cheang, Q. S., and Mohan Rao, P. K. (1972). Protein variability in Malayan varieties of rice (*Oryza sativa* L.). *Malaysian Journal of Science* **1**, 35–43.

De Datta, S. K., Magnaye, C. P., and Magbanua, J. T. (1969). Response of rice varieties to time of nitrogen application in the tropics. *In* 'Proceedings of the Symposium of Tropical Agriculture Researches'. pp. 73–87. (Agriculture, Forestry and Fisheries Research Council: Tokyo.)

De Datta, S. K., Obcemea, W. N., and Jana, R. K. (1972). Protein concentration of rice grain as affected by nitrogen fertilizer and some triazines and substituted ureas. *Agronomy Journal* **64**,785–8.

Ebata, M., and Nagato, K. (1967). Ripening conditions and grain characteristics. International Rice Committee Newsletter (special issue), 10–17.

Efferson, J. N. (1985). Rice quality in world markets. *In* 'Rice Grain Quality and Marketing'. pp. 1–14. (International Rice Research Institute: Los Baños, Philippines.)

Gomez, K. A. (1979). Effect of environment on protein and amylose content of rice. *In* 'Proceedings of the Workshop on Chemical Aspects of Rice Grain Quality'. pp. 59–68. (International Rice Research Institute: Los Baños, Philippines.)

Gomez, K. A., and De Datta, S. K. (1975). Influence of environment on protein concentration of rice. *Agronomy Journal* **67**, 565–8.

Ikehashi, H., and Khush, G. S. (1979). Methodology of assessing appearance of the rice grain, including chalkiness and whiteness. *In* 'Proceedings of the Workshop on Chemical Aspects of Rice Grain Quality'. pp. 223–9. (International Rice Research Institute: Los Baños, Philippines.)

International Rice Research Institute (1973). 'Annual Report for 1972.' (International Rice Research Institute: Los Baños, Philippines.)

International Rice Research Institute (1976). 'Standard Evaluation System for Rice. International Rice Testing Program.' Second printing, 1976. (International Rice Research Institute: Los Baños, Philippines.)

Juliano, B. O. (1979). The chemical basis of grain quality. *In* 'Proceedings of the Workshop on Chemical Aspects of Rice Grain Quality'. pp. 69–90. (International Rice Research Institute: Los Baños, Philippines.)

Juliano, B. O. (1985). Polysaccharides, proteins, and lipids of rice. *In* 'Rice: Chemistry and Technology'. (Ed. B. O. Juliano.) p. 62. (American Association of Cereal Chemists: St Paul, MN.)

Juliano, B. O., Nazareno, M. B., and Ramos, N. B. (1969). Properties of waxy and isogenic nonwaxy rices differing in starch gelatinization temperature. *Journal of Agriculture and Food Chemistry* **17**,1364–9.

Kanda, M., Ikehashi, H., and Ito, R. (1969). Evaluation of brown rice character through milling tests of samples partitioned according to grain trait. *Agricultural Technology Tokyo* **24**, 24–7.

Khush, G. S., Paule, C. M., and de la Cruz, N. M. (1979). Rice grain quality evaluation and improvement at IRRI. *In* 'Proceedings of the Workshop on Chemical Aspects of Rice Grain Quality'. pp. 21–31. (International Rice Research Institute: Los Baños, Philippines.)

Lewin, L. G., Reinke, R. F., Blakeney, A. B., and Reece, J. E. (1994). Effect of nitrogen, cultivar and time of harvest on rice physical quality attributes. *In* 'Proceedings 1st Temperate Rice Conference (Vol. 1)'. Yanco Agricultural Institute, Yanco. ISBN 0 646 20410 6.

Little, R. R., Hilder, G. B., and Dawson, E. H. (1958). Differential effect of dilute alkali on 25 varieties of milled white rice. *Cereal Chemistry* **35**, 111–26.

Nakatat, S., and Jackson, B. R. (1973). Inheritance of some physical grain quality characteristics in a cross between a Thai and Taiwanese rice. *Thailand Journal of Agricultural Science* **6**, 223–35.

Nanda, J. S., and Coffman, W. R. (1979). IRRI's efforts to improve the protein concentration of rice. *In* 'Proceedings of the Workshop on Chemical Aspects of Rice Grain Quality'. pp. 33–47. (International Rice Research Institute: Los Baños, Philippines.)

Nangju, D., and De Datta, S. K. (1970). Effects of varietal types and nitrogen level on the optimum time of harvest and the amount of breakage in transplanted rice. *Agronomy Journal* **62**, 468–74.

Place, G. A., Sims, J. L., and Hall, V. L. (1970). Effects of nitrogen and phosphorus fertilization on growth, yield and cooking characteristics of rice. *Agronomy Journal* **62**, 239–43.

Resurreccion, A. P., Hara, T., Juliano, B. O., and Yoshida, S. (1977). Effect of temperature during ripening on grain quality of rice. *Soil Science and Plant Nutrition* **23**, 109–12.

del Rosario, A. R., Briones, V. P., Vidal, A. J., and Juliano, B. O. (1968). Composition and endosperm structure of developing and mature rice kernel. *Cereal Chemistry* **45**, 225–35.

Schoch, T. J., and Maywald, E. C. (1956). Microscopic examination of modified starches. *Analytical Chemistry* **28**, 382–7.

Seetanun, W., and De Datta, S. K. (1973). Grain yield, milling quality and seed viability of rice as influenced by time of nitrogen application and time of harvest. *Agronomy Journal* **65**, 390–94.

Somrith, B. (1974). Genetic analysis of traits related to grain yield and quality in two crosses of rice (*Oryza sativa* L.). PhD thesis, Indian Agricultural Research Institute, New Delhi, India. 138 p.

Srinivas, T., and Bhashyam, M. K. (1985). Effect of variety and environment on milling quality of rice. *In* 'Rice Grain Quality and Marketing'. pp. 49–59. (International Rice Research Institute: Los Baños, Philippines.)

Tashiro, T., and Ebata, M. (1975). Studies on white belly rice kernel. III. Effect of ripening conditions on occurrence of white belly kernel. *Proceedings of the Crop Science Society of Japan* **44**, 86–92.

Webb, B. D., Bollich, C. N., Adair, C. R., and Johnston, T. H. (1968). Characteristics of rice varieties in the U.S. Department of Agriculture collection. *Crop Science* **8**, 361–5.

Webb, B. D., Bollich, C. N., Carnahan, H. L., Kuenzel, K. A., and McKenzie, K. S. (1985). Utilization characteristics and qualities of United States rice. *In* 'Rice Grain Quality and Marketing'. pp. 25–35. (International Rice Research Institute: Los Baños, Philippines.)

Webb, B. D., Bollich, C. N., Johnston, T. H., and McIlrath, W. O. (1979). Components of rice quality: their identification, methodology, and stage of application in United States breeding programs. *In* 'Proceedings of the Workshop on Chemical Aspects of Rice Grain Quality'. pp. 191–205. (International Rice Research Institute: Los Baños, Philippines.)

Yoshida, S. and Parao, F.T. (1976). Climatic influence on yield and yield components of lowland rice in the tropics. *In* 'Climate and Rice'. pp. 471–94. (International Rice Research Institute: Los Baños Philippines.)

Manuscript received 6 April 1998, accepted 7 July 1999