

Analysis of plant tissue test samples stored for up to 11 years

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Introduction

The operation of a tissue test service presents the opportunity to collect samples from a wide genetic and geographic range and store them for use in future studies. Such samples can be used to establish calibrations for previously unanalysed constituents and for testing new NIR equipment. We took this opportunity fifteen years ago for Australian rice samples and now have a large collection stored dry, in the dark and at ambient temperature. A few samples are also stored dry, in the dark at -20°C.

With time, samples fade from greenish to browns but show no other apparent changes. We have lost some samples to book lice and now take precautions against these and other insects. Samples from the past 11 years are stored together under identical conditions so that any trends due to storage time would be apparent.

Materials and method

When the rice tissue test was set up in 1988 a Bran and Lubbe 400 was used. This was upgraded to a 450, 19 filter instrument. The initial calibrations were made using MLR and the "UK" software in CPM. We soon moved to an IBM 286, again with "UK" software. In 1992 a NIRsystems 6500 was tested, using NSAS calibration software through several versions. In the first year MLR was used but was quickly replaced by PLS. In 1996 we changed to ISI software and have continued to use this through its updates to the present. All calibrations have only used the NIR wavelengths.

The rice tissue test includes calibrations for nitrogen, starch, phosphorus, potassium, sulphur, zinc and growth stage. Nitrogen and starch re-calibrations incorporate new samples from the past season, new varieties as they were released and samples that extended the range of constituent values.

Samples from most years since 1992 were recently reanalysed using the 2003 calibration in order to assess the progression of the test over the past 11 years and to evaluate the samples themselves for changes due to age and storage. The set analysed has a full range of nitrogen content (from 0.8% to 3.2%) based on the original Kjeldahl or Leco analysis (Figure 1).

Results and discussion

Nitrogen

Nitrogen is the major nutrient that influences rice yields and the major reason for tissue testing. In Figure 1 a set of samples from most years since 1992 was re-analysed using the 2003 calibration. Samples are well grouped about the trend line with the two samples furthest from the line being

both 1992 samples. When individual years are plotted, as in Figure 2, it can be seen that most years have trend lines very close to the “all years” average line and the 45° line that would represent perfect agreement between the current and past calibrations. Two years, 1995 and 2000, have considerable bias differences—possibly result from the inclusion of a number of new variety samples in those years. The calibration for 1992 which has both a slope and bias difference was developed on the old filter equipment.

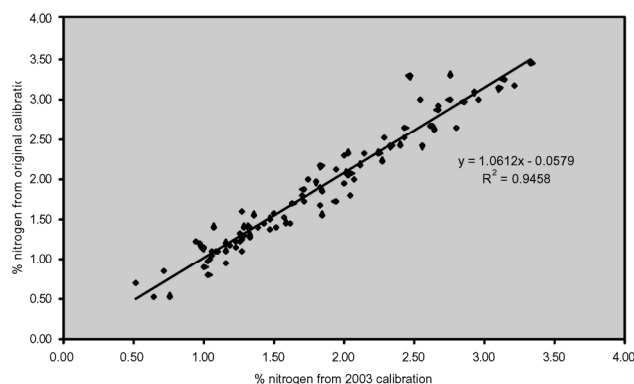


Figure 1. Nitrogen content of samples collected from farmer tissue tests between 1992 and 2002 plotted against their 2003 re-analysis

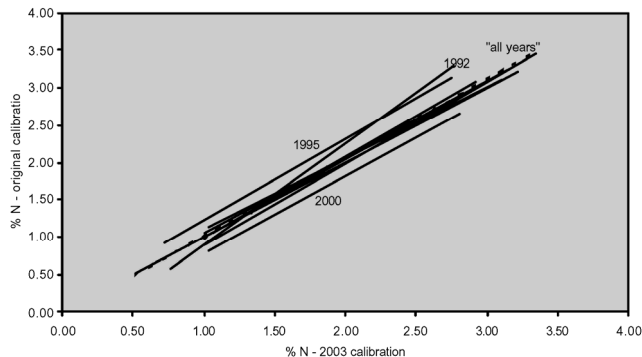


Figure 2. Nitrogen content of samples from each year from 1992 to 2002 plotted against the "all years" regression line

Starch

The rice tissue test uses starch as an indicator of the energy status of the crop. The analysis of starch in plant material is a much more difficult analysis than nitrogen and the starch content of rice tissue at panicle initiation time can vary from near zero to more than 35%, dry basis. Figure 3 shows the individual trend lines, for most years since 1992, for a set of samples from each year re-analysed using the 2003 starch calibration. Much greater variations in both slope and bias can be seen than in the nitrogen graphs in Figure 2. These variations may reflect the greater difficulty of starch analysis and be due to analytical technique in the reference method. Variation in the samples is unlikely, as no trend due to sample age is apparent.

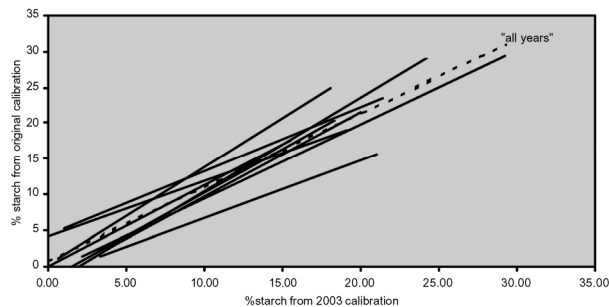


Figure 3. Starch content of samples from each year from 1992 to 2002 plotted against the "all years" regression line.

Conclusions

Ground rice shoot samples which have been stored for 11 years have maintained their integrity. The re-analysis has demonstrated that the stored sample bank is suitable for use in testing new equipment for the tissue test and can be used in developing new calibrations.

Acknowledgements

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