Breeding – what's in the pipeline?

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Summary

There are significant price and profitability challenges for Australian cotton at present and likely into the future. Changes at all levels of the production and processing steps can address the challenges and it will be best if all contribute some innovation. As plant breeders we recognise our responsibility and this paper discusses some of the technical challenges for progress and presents some likely new varieties and traits which may become available in the short to medium term.

Yield needs to increase to maintain profitability. Protection from disease will help realise yield potential, but discoveries are needed in increasing yield potential. New molecular traits will become available in the short to medium term to assist with some production challenges. Molecular tools should assist with the speed and effectiveness of breeding for many objectives.

Our fibre quality needs slight improvement to ensure our markets will source our product preferentially because of the quality and value. There may be opportunities for a small fraction of our production to address more premium markets. The biggest immediate challenge for breeders is to **combine** all the yield, disease resistance and fibre quality targets **with** the most popular transgenic traits.

Introduction

Breeders, growers, ginners and spinners have challenges to achieve industry targets and viability and all groups need to be innovative and effective for genuine progress. At this Conference in 1990, Norm Thomson raised the dilemma facing breeders about over reliance on breeding to solve problems: "challenge... to first decide on whether any particular solution to a problem is best sought by breeding or whether it is possible and preferable to go for the non-breeding solution". This comment was made at that time in a plea for what was a long list of expectations from breeders. The list of expectations and targets for breeders is now longer: more yield on top of the world's best; better quality when there is a yield penalty; more disease resistance; and combine all of these with traits such as Bollgard® II/Roundup Ready Flex®. So even if the breeding program is bigger in 2006, the challenge is substantially bigger.

Yield

Cotton lint yield in Australia continues to increase by about 22.7 kg lint (0.1 bale)/ha/year through better varieties and better management. Can that rate of progress continue? High yields are needed for growers to remain viable, so both breeding and farming systems (especially together) have a challenge. Our targets for breeding aim to at least maintain the genetic yield gain, if not double it. Combining yield, fibre quality, disease resistance and transgenic traits is a massive task and strategic planning, appropriate levels of investment and pre-breeding are important to achieve and deliver genuine improvements. That combination takes significant time.

Disease

Over the last 20 years great progress has been made in improving the disease resistance of cotton varieties in Australia. A number of diseases have caused severe damage to crops, but breeding for resistance has alleviated some of the problems. However, new disease challenges such as Fusarium wilt mean that as breeders we still have much work to do, particularly in combining disease resistance with best yield and fibre quality and transgenic traits.

Bacterial blight arose as a serious problem during the 1970's and 1980's, causing significant yield losses. Beginning with the release of Siokra 1-1 in 1985, all subsequent CSIRO varieties have possessed near immunity to blight and now the disease is very rare. It is only found in susceptible Pima varieties and this should also soon be overcome with the imminent release of a blight resistant Pima variety by CSIRO. While our current sources of resistance appear to be very stable we need to be vigilant in case new strains of blight should arise.

Verticillium wilt has always been a significant challenge to the modern irrigated cotton industry with areas such as the Namoi and Macquarie valleys and the Darling Downs suffering severe losses in cooler seasons. The development of resistant varieties such as Sicala V-2 have had a major impact on reducing the levels of Verticillium and the current good level of resistance amongst CSIRO varieties seems to be adequate to hold the disease in check. Again we need to be vigilant about more virulent races developing and we continue to search for even greater levels of resistance.

The identification of Fusarium wilt on the Darling Downs in the early 1990's and its subsequent discovery in most other growing regions represents a serious challenge to cotton production. In the most severely infected fields on the Darling Downs, cotton production is no longer economic. However, we are making significant progress in increasing resistance – for instance in the 2005/06 season the seven most popular CSIRO varieties all had Fusarium Resistant Ranks (F Ranks) greater than 100. When we commenced our major Fusarium effort in the mid 1990's only Sicot 189 (F Rank = 100) had any appreciable resistance. We have achieved an F Rank of 200 with Sicot F-1, but under severe disease pressure even this is inadequate. The suggestion by some that an F Rank of 150 may be

adequate to cope with the disease in most areas makes the dangerous assumption that the disease will not reach the severe levels that are found elsewhere such as that present on the Downs.

There is a yield penalty with the highest levels of Fusarium resistance. We have made progress in breaking this linkage, indicating that some of the yield depression may be due to the exotic germplasm sources being used to bring in resistance. For instance, we will release a variety in 2007 with the yield potential of Sicot 71 with an F Rank greater than 150. Unfortunately, the limited sources of resistance available within *Gossypium hirsutum* make immunity to Fusarium a very difficult target. Other *Gossypium* species and biotechnology may offer the best (though long term) chances to achieve the target of immunity. This is also true of another significant cotton disease, black root rot, to which *Gossypium hirsutum* does not appear to have any resistance. We hope in future the use of DNA markers in the selection process will improve the speed and efficiency of breeding for disease resistance.

Fibre quality

Fibre quality marketing and spinning requirements are not static, with considerable shifts in spinning capacity and production in different countries. Competition has become fiercer as a result and so our future fibre quality package and delivery to market may also need to change.

Unfortunately the speed of these changes is quicker than we can breed new varieties. One aspect of competition is that Australia needs to get improved fibre onto the spinning market *before* our competitors do. This establishes (maintains) our reputation and facilitates market pull.

From a breeding perspective in developing fibre breeding targets, there are two issues; first, the negative association between yield and fibre quality and second, the dilemma on exactly how much effort should be expended on research and development of a broader fibre quality portfolio, when there is uncertainty in the exact future requirements or adoption by the growing or spinning industry.

Yield and quality. Within a variety, better growing conditions and yields usually mean a better fibre quality package, except for the possibility of high micronaire. Within a breeding population however, the better yielding lines generally have reduced fibre quality, especially in fibre length, length uniformity, fibre fineness and strength. Thus heavy selection pressure for yield during breeding will reduce fibre quality on average (ie shorter fibre with higher micronaire). Some of the negative associations between yield and quality may be genetic linkages and as can be seen from the scatter of the relationship, there are outliers which give hope for identifying good combinations of yield and quality as long as the population size is large enough and the measurements are accurate. These requirements cost time and money; both of which are now scarce.

Other negative associations may be due to functional or physiological linkages such as between finefibred types having lower yield because there are fewer fibres per seed in which to stack cellulose and hence yield. To reduce the likelihood of high micronaire, it may be necessary to also breed for more fibres per seed to increase the capacity of the seed surface to draw resources away from other tissues in the plant competing for the available sugar produced by the leaves.

Broader portfolio. Globally, fibre quality is improving for a range of reasons including response to demand and more attention / success by breeders, especially with HVI instrumentation allowing more, quicker and cheaper testing. It may be that a broader portfolio of fibre quality packages is needed at the research and breeding level to be ready for possible shifts in longer term fibre quality preferences by the spinning industry. In addition to a slightly improved package than we have at present, a genuine longer and stronger premium fibre type may have a small demand in the spinning industry. Sicala 350B, a premium fibre Bollgard®II variety has been released and is attracting some interest and a price premium in the market. In addition, there is continued interest in Pima and it is worthwhile maintaining some activity in this area. The current relative prices for Pima fibre are attractive.

Biotechnology

The imminent release of CSIRO varieties with the enhanced Roundup Ready Flex® trait signals the end of the first generation of transgenic cotton crop protection traits developed by Monsanto (Ingard® and Roundup Ready®). These traits have had positive impacts on the efficiency and economics of cotton production in Australia. Release of a small variety suite with the alternative herbicide tolerance trait from 2006, Liberty Link® cotton, conferring resistance to the glufosinate ammonium herbicide, will provide growers with other options for weed control and this may relieve pressure on selection for Roundup tolerant or sensitive weeds.

All of the major biotech companies maintain active product pipelines targeted at many of the major broadacre crops, including cotton, Although Monsanto has focussed mainly on crop protection traits it has recently been moving towards traits concerned with crop performance and quality (nitrogen use efficiency, drought tolerance, modified oil and protein content) and some of these traits are now being evaluated in cotton (http://www.monsanto.com/monsanto/layout/investor/company/productpipeline.asp#). Other companies are developing similar traits (Bayer CropScience with insecticide traits and an alternative glyphosate resistance trait; Dow AgroSciences with insecticide traits (eg., WidestrikeTM); and Syngenta with insecticide traits (eg., VipcotTM)). Some of these are being evaluated in Australia. Important considerations will be how they comply with existing resistance management strategies and delivery into well adapted germplasm.

There are some new traits of interest for cotton in the Monsanto pipeline, but as they are still in the proof of concept or discovery phase they are not likely to appear in breeding programs for some years. These include possible traits for drought tolerance, experimental material of which is currently being imported by Monsanto Australia for preliminary internal evaluation. In the a world of growing climatic unpredictability, such a trait would have significant value if it can deliver some buffering of yield or quality against water stress or even if it enhances water use efficiency.

So for the immediate future, our breeding pipeline will rely on conventional enhancements to the genetic backgrounds in which we will deliver the existing traits of Bollgard®II, Roundup Ready Flex® and Liberty Link®. This will include the yield, quality and disease tolerance traits discussed earlier.

How can we be smarter in developing such enhanced germplasm? Modern biology can contribute not only transgenic traits but also better conventional breeding tools. Other crops are starting to benefit from such tools, but cotton has lagged behind. Stacking rust resistance genes into a single wheat variety to give more durable disease resistance is a difficult task (as it is hard to tell which plants have the multiple resistance genes just from their tolerance to the rust fungus). Using DNA markers that are specific for each of the rust resistance genes being combined makes it a simple task to identify plants with two or more different resistance genes just by running a DNA assay on a small piece of leaf – without even having to put any rust fungus on the plant! The assay serves as a substitute for doing a biological disease resistance test and takes away the uncertainty in identifying plants carrying resistance genes and can significantly reduce the numbers of breeding lines that need to be taken to the field for detailed evaluation. How much easier would it be for cotton breeders if they could use one or two simple tests on young seedlings in the glasshouse to track fusarium resistance through breeding populations instead of having to test multiple lines in a fusarium nursery over two or three successive years? The same would be true if molecular markers could be identified for other diseases such a Cotton Bunchy Top or for fibre yield or quality traits. The negative associations between yield and some fibre traits might be tackled with the right DNA markers as they would allow much larger numbers of plants to be screened in early generations to break the negative genetic linkages between such associated traits.

So why don't we already have useful DNA markers for important traits in cotton? The answer is complex but involves at least two main components.

- Cotton is a relatively new plant on an evolutionary scale and has gone through a number of
 genetic bottlenecks, some historically recent, that means that most modern cotton varieties are
 closely related to each other and so lack the diversity in DNA changes in their genes that are
 essential to find and implement DNA markers (this is not true for many other crops).
- 2. A lack (globally) of commitment to investment in this area in cotton probably because the first point means that it is harder and more expensive to find useful markers in cotton and hence high risk when there are limited research funds.

This slowness in discovery in cotton is now being addressed at an international level and CSIRO led the founding the International Cotton Genome Initiative (ICGI) that has brought together cotton scientists from around the world to combine their efforts in DNA marker discovery in cotton. The eventual aim is to generate high density genetic maps of different cotton species and sequence the cotton genome so that cotton will have the same type of genetic and genomic information enjoyed by scientists working in rice, maize, wheat and barley. Already in the last year a number of groups in the

US, China and France have published the use of DNA markers in experimental crosses in cotton to tag genomic locations that confer high fibre quality, so now we can see if those markers can be used to select for high performing combinations of those traits in real breeding populations. CSIRO aim to utilise this information to speed up how we select for fibre quality, or screen DNA markers being lodged in International databases to find markers linked to disease resistance. We already use such DNA markers in our transgenic breeding to follow and select traits like Bollgard®II and RRFlex®, so technically we should have no difficulty in using similar methodologies for yield, quality or disease traits once useful markers are identified.

Breeding challenges

The conventional breeding program forms the basis for new discoveries in all areas; yield, disease resistance and fibre quality. However, due to the high adoption rate of transgenic varieties, the commercial demand for conventional varieties is small. In the future we expect that most new discoveries will only be released in transgenic varieties. While this is the commercial reality, it has the effect of delaying the introduction of new discoveries by at least two years while a transgenic version is produced. While some breeding can be done between and among different transgenic varieties the available genetic pool out of which new characteristics can be selected is too small to offer any large advances for many important performance and disease traits. Of possible greater importance however, is the difficultly in finding conventionally sprayed trial sites to test new conventional lines. In some regions the uptake of transgenic varieties is such that the only conventional cotton grown is in unsprayed refuges. The impact of this restriction in testing had not yet been felt, but over the next few years we feel that delivery of new discoveries may be constrained due to the lack of data from conventional sites.

Another major challenge is the balance within the breeding program between conventional and transgenic variety development. We have been in an environment of rapidly changing transgenic traits and a market that continually demands new traits in the best varieties. Because of these market pressures, our ability to allocate resources to the area of new discoveries also becomes constrained. An example of this is the introduction of Roundup Ready Flex®. CSIRO expect to have a full suite of Bollgard®II/Roundup Ready Flex® varieties to replace the existing Roundup Ready® by 2009. However, this is just through the introduction of this new trait into existing conventional or Bollgard® II varieties, without any new discoveries. The first releases in 2006 are in the Sicot 80 and Sicot 43 backgrounds; Sicot 80 was released as a conventional variety in 2001 and has since been superseded!

These challenges are significant and the way that we address them will have a major bearing on the varieties that will be available over the next ten years.

A vision for 2012

Australian cotton varieties in 2012 will have better combinations of yield, fibre quality and disease resistance compared with Sicot 71in 2006. If the Australian cotton industry maintain the 22.7 kg lint/ha/year yield increase, then in 2012, yields should be trending to an extra 136 kg lint (0.6 bale)/ ha.

Disease resistance will be improved; a FRR of 150 will be the norm, but research will still be underway to discover immunity to Fusarium wilt. The majority of fibre will be slightly improved quality compared with 2006, with another 10% to 15% as premium types. Crop management and processing of cotton will be more attuned to optimising fibre quality.

These combinations of yield, fibre quality and disease resistance will be delivered with traits such as Bollgard®II/Roundup Ready Flex®.

There may be competition from other providers for these types of traits. The concept of genes for tolerance to heat and water stress will still be discussed, with more advanced research underway. Molecular tools will be in more common use to assist with making breeding more efficient and more successful.