

Delivering drought tolerance to those who need it: from genetic resource to cultivar

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Abstract. Drought limits crop yields, and with climate change, the severity of water stress is projected to increase in many production environments. Therefore, it has never been more important to deliver the findings of drought research to farmers. The maintenance *in situ*, collection and characterisation of key genetic variability for stress tolerance, its introgression into agronomically adapted materials, and the subsequent deployment of improved cultivars is a continuum. This paper focuses on one segment of the pathway—the process from genetic characterisation to cultivar delivery—and possible efficiencies are discussed with emphasis on wheat, one of the world's most important food crops.

The first efficiency is to limit the initial exploitation of genetic resources to close relatives, as much of this variation remains uncharacterised, rather than attempting gene transfers from unrelated species, which is time-consuming and has a low probability of success. Synthetic wheat, developed by crossing modern tetraploid wheat to *Aegilops tauschii*, the donor of the D-genome, has provided new genetic diversity for stress tolerance and yield advantages under drought in excess of 30% have been reported. Synthetic wheat can also be made using *Triticum dicoccum*, often referred to as emmer wheat, thus introducing new variation for all three wheat genomes.

The second efficiency is better coordinated, field-based phenotyping. The Australian Managed Environment Facility and similar national facilities established in India and China provide a basis for accurate field-based phenotyping and the weighting of physiological traits related to water-use efficiency on a national scale. The calculation of a water balance, careful management of site heterogeneity and judicious use of rain shelters maximise trait expression and improve the relevance of results.

The third efficiency is to maximise locally the benefits of global public-good research. The global wheat improvement programs of the Consultative Group on International Agricultural Research (CGIAR) have a mandate to 'tame' genetic diversity and distribute these improved materials globally in nurseries targeted to specific environmental conditions such as drought. Nevertheless, the exploitation of these materials is rarely managed well at the national level. The CIMMYT Australia ICARDA Germplasm Evaluation (CAIGE) program is a nationally coordinated germplasm introduction and evaluation program. Key CGIAR nurseries are received and grown by Australian quarantine, increased at one or two locations, tested for disease resistance and subsequently grown nationally in yield trials. The data are stored on the CAIGE website and, along with all supporting data generated by the international centres, are publicly available.

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Introduction

Drought affects most crop-production environments and its severity varies depending on location and year. Global productivity increases of major crops such as wheat and rice have averaged close to 1% per annum over the past 50 years (Waddington *et al.* 1986; Byerlee and Moya 1993; Manès *et al.* 2012); however, the rates of progress tend to be lower in the less productive environments (Perry and D'Antuono 1989; Jain and Byerlee 1999). In addition, it is difficult to separate improvements in the agronomic system from genetic advances, as the two are frequently interdependent. For example, one of the greatest genetic advances in the 20th Century was the development of semi-dwarf wheat and rice cultivars, which gave rise to the Green Revolution. However, the impact of the shorter cultivars with their inherent higher harvest index was only realised through

improved crop nutrition, particularly nitrogen (Trethowan *et al.* 2007). The impact of the Green Revolution has slowly declined, and at the turn of the 21st Century, rates of genetic advance in crop productivity have slowed and even stagnated in many environments, particularly the drier areas (Graybosch and Peterson 2010).

Nevertheless, rates of genetic advance can be increased if significant unexploited genetic variation present in most of the key crop gene pools can be accessed and exploited. Past attempts to introduce new genetic variation into crops such as wheat have focused on wild ancestors of the A and D genomes and alien introgression, largely focussed on disease resistance, and using cytological techniques (Trethowan and Mujeeb-Kazi 2008). However, significant genetic variation for response to limited moisture exists within the adapted gene pool, and although

breeders have steadily exploited this over the years to attain the reported 1% annual rates of genetic advance, the full impact of this variation is yet to be realised. This lack of progress reflects the inefficiency of ‘crossing the best with the best’—the mantra of the plant breeder—when the allelic diversity of the materials is unknown. Recent advances in genotyping and the cost-effectiveness of SNP (single nucleotide polymorphism) and GBS (genotyping by sequencing) technologies have drastically altered the plant-breeding landscape. Breeding methods that better utilise genetic variation for water-use efficiency in adapted germplasm can now be introduced.

The power of new-generation genotyping technologies can only be realised if the old problem of relevant and high-throughput phenotyping can be overcome. When addressing water-use efficiency or drought tolerance, it is desirable that phenotyping be conducted in the field. Controlled environment testing can augment field-based phenotyping, particularly if optimisation of trait expression is required, but it should not be the primary basis for breeder selection. If heterogeneity in the field can be controlled, managed or measured, then rates of genetic advance will improve. Field-based phenotyping can be conducted in the target environment and on sufficient scale to allow efficient management of gene frequency. Nevertheless, the stress treatments imposed must reflect the most probable environment type facing farmers.

Whereas efficient exploitation of genetic diversity and accurate field-based phenotyping are essential for most regional breeding programs targeting moisture-limited environments, additional advantage can be levered from global public-good research. The international centres of the Consultative Group on International Agricultural Research (CGIAR) have a mandate to use genetic diversity to improve the productivity of cropping environments in the developing world. Centres such as the International Maize and Wheat Improvement Centre (CIMMYT), the International Centre for Agricultural Research in the Dry Areas (ICARDA), the International Crops Research Institute for the Semi-arid Tropics (ICRISAT) and the International Rice Research Institute (IRRI) host extensive gene banks of the world’s most important crops and develop adapted germplasm, which is distributed globally each year. However, the true value of these materials is often never realised because of *ad hoc*, piecemeal evaluation at the national level. Better coordinated exploitation of these materials would produce significant dividends in the short to medium term, thus allowing national programs to lever the significant international investment in agricultural research.

The three efficiencies of better exploitation of genetic resources, improved and relevant phenotyping, and enhanced exploitation of global public good research are examined in the context of one of the world’s most important crops: wheat.

Stepwise exploitation of genetic resources

Plant breeders have traditionally crossed ‘the best with the best’ when designing their crossing strategies. However, new genotyping technologies have made it possible to better target such approaches. The breeder will now cross the individuals with the highest trait values that represent the greatest allelic diversity. New breeding schemes and techniques such as

association analysis of whole breeding programs offer improved efficiency. One such example is the association analysis of the CIMMYT wheat-breeding program (Crossa *et al.* 2007). In this case, 170 wheat genotypes, evaluated in multi-environment trials over a 25-year period, were genotyped using DArT (diversity arrays technology) markers, and an association analysis was subsequently used to study four traits: grain yield, and resistance to stem, leaf and stripe rusts. Once the known genes that influence yield were accounted for, such as vernalisation, photoperiod and plant height, a genetic blueprint of grain yield on a global scale was identified. A summary of the findings is presented in Table 1. There are significant DArT markers on several chromosomes including 1AS, 1BL, 1DS, 2AL, 3AL, 3BS, 3DS, 5AS, 5BS, 5DS, 6AS, 6AL, 6BS and 6BL that do not carry known genes or previously reported quantitative trait loci (QTLs) that influence yield.

CIMMYT breeders are now able to combine significant markers in targeted crosses to improve the global adaptation of wheat. This principle can also be applied to regional breeding programs. Atta (2013) conducted an association analysis of a regional Australian wheat-breeding program, including parental lines and derived progeny tested in multi-environment, multi-year trials, and identified several significant markers for grain yield located on different chromosomes, once the marker effects linked to known rust-resistance genes were removed (Table 2). A crossing program was subsequently initiated to combine as many of the significant markers as possible linked to grain yield.

A key trait linked to improved water-use efficiency is root disease resistance. Clearly, a healthier root is better able to exploit available soil moisture, and root diseases tend to be most severe under drought as the plant is unable to out-grow the disease. The inheritance of resistance to root diseases is generally less complicated than water-use efficiency *per se*. If the problem is identified in the target region and genetic variation is available, then these traits should be among the first breeding targets. An example is the improvement of crown rot (caused by *Fusarium pseudograminearum*) resistance in wheat. The inheritance of this disease is complex and the symptoms are exacerbated under dry conditions; economic losses of up to AU\$79 million are recorded annually in northern New South Wales and Queensland (Murray and Brennan 2009) and progress through traditional breeding and selection has been slow. A marker assisted recurrent selection (MARS) scheme as outlined by Charmet *et al.* (1999) was implemented to exploit genetic variation in the adapted wheat gene pool. Prior to the implementation of the scheme, significant resources had been committed to identifying QTLs in bi-parental crosses (Collard *et al.* 2006; Li *et al.* 2012) and introducing these into adapted backgrounds, with marginal success. In the MARS scheme, three-way crosses were made among polymorphic parents that represented some degree of resistance, and up to 300 F₃ progeny were derived from each cross and genotyped. Following a seed increase, the progeny were then tested for resistance under both controlled and field conditions. The controlled environment evaluation used a pot test in which infected wheat bran was spread on the pot surface after planting, as described by Raju *et al.* (2013). The field-based screening used a combination of yield loss, based on

Table 1. Significant diversity arrays technology (DArT) markers associated with grain yield from association analysis of 25 years of CIMMYT multi-environment yield trials
Shaded areas indicate significant DArT markers not associated with known or previously published genes that influence yield. Adapted from Crossa *et al.* (2007)

Chr.	Short arm (S)		Long arm (L)		Unknown arm	
	DArT (wPt)	Gene and/or QTL	DArT (wPt)	Gene and/or QTL	DArT (wPt)	Gene and/or QTL
1A	5374, 4765, 2872, 9266, 2311, 7030, 4029	—	3870, 3904, 5274, 4709, 6754, 8016, 0128	QTL	9592, 1011, 6853	QTL
1B	8949, 1328, 7094, 3465, 4434, 0974, 6427, 8986, 1781, 5065, 5678, 5363, 6777, 5801, 6117, 6833, 8616	1B/1R transl.	2315, 0944, 2526, 4129	—	5316, 1139, 2019	QTL
1D	3738, 9380	—	7711, 9857, 3743, 4427, 2897, 1263, 7092	QTL	—	QTL
2A	7901, 6687, 9793, 3114	<i>Ppd-A1</i>	5647, 2087	—	6207	QTL
2B	0100, 4916, 6706, 8326, 3983, 5672, 4125, 7757	<i>Ppd-B1</i>	3132, 9350, 7350, 9336, 2135, 3378, 7360, 0049	<i>Eps</i> QTL	0094, 0950, 4210	QTL
2D	4144	<i>Ppd-D1Rht8</i>	4413	<i>Eps</i>	—	QTL
3A	9369, 1681	QTL	7756, 2698, 7992, 2938, 4725, 9268, 4407, 1688	—	—	<i>Eps</i>
3B	0365, 0995, 5716, 7015, 8352, 8983, 7142, 9310, 4209, 9510, 5390, 9170, 6047, 5105, 6802, 8886, 0280, 0384, 8238, 8096, 4412, 8845	—	—	QTL	9368, 2757	<i>Vrn</i> QTL
3D	1365, 9401	—	—	QTL	—	<i>Ppd</i> QTL
4A	2788	QTL	7280, 2084, 3795, 7807, 8271, 4620, 2533	QTL	4660, 7924	<i>Eps</i>
4B	3908, 6149	<i>Rht-B1</i>	1505, 8292	QTL	3608, 5265	<i>Eps</i> QTL
4D	—	<i>Rht-D1</i>	—	QTL	—	<i>Eps</i>
5A	1165, 0605	—	5231	<i>Vrn-A1</i>	4249	QTL
5B	6136, 1420, 9666, 5175	—	9467, 0103, 4996, 7101, 5896, 3030, 9598	<i>Vrn-A2</i>	4418, 4703, 4936	QTL
5D	1400	—	—	<i>Vrn-A3 Vrn-4</i>	—	QTL
6A	7475, 0864, 8006, 7938, 9075	—	9690, 2573, 0959, 7063, 1642, 7204	—	—	QTL
6B	9532, 1922, 3130, 9990, 6994, 8239, 4720, 1547, 6282, 3116, 3304, 1852, 7777, 4706, 3733	—	6116, 0171, 1264, 3045, 9256, 1541	—	—	<i>Eps</i> QTL1
7A	6034, 8789, 4835	QTL	3393, 6495	QTL	2266, 4553, 4748	QTL
7B	—	—	6273, 9925, 7887, 5343, 8921, 5547, 5280, 0194, 6156, 4300, 7108, 6320, 0504	QTL	1533, 9746	<i>VrnB4, Vrn5Eps</i> , QTL

uninoculated and inoculated paired plots, and symptom scoring under crown rot pressure. An association analysis was then used to identify significant marker effects in both controlled and field screening. Those progeny carrying significant markers that expressed in both tests were then combined in crosses and progeny carrying pyramided markers identified (Table 3). The identification and pyramiding of cross-specific marker effects overcomes the problem of poor QTL expression in backcross-derived lines based on QTLs identified in biparental mapping populations. The next step in breeding efficiency would be the identification of suitable training populations for genomic selection, as the cross-based phenotyping required for MARS eventually becomes unmanageable in most breeding programs.

Once the genetic variation in the existing adapted gene pool has been exhausted, it makes sense to exploit variation in the ancestral A, AB and D genomes. The donor of the B genome in wheat is unknown, although some believe that *Aegilops speltoides* is the most likely contender (Salse *et al.* 2008). Nevertheless, significant genetic variation exists in both *Triticum dicoccoides* (wild emmer) and *T. dicoccum* (cultivated emmer) to provide sufficient B genome variation. Much has been published on the development and exploitation of synthetic wheat, and the breadth of work is reviewed in Trethowan and Mujeeb-Kazi (2008) and Trethowan and van Ginkel (2009). Synthetic hexaploid wheat is derived by crossing both adapted and unadapted tetraploid wheat with *Aegilops tauschii*, the donor of the D-genome. These materials offer significant advantages in water-limited environments, and cultivars have been released in several countries: Lalma in Pakistan (CIMMYT Wheat Atlas), Maravilla in Mexico

Table 2. Number of significant DArT markers for grain yield and their chromosome location based on association analysis of 300 parental lines and derived progeny evaluated in multi-environment trials across 3 years

Major effects linked to known rust resistance genes have been removed.
Modified from Atta (2013)

Chromosome	No. of significant markers linked to yield
1A	3
1B	1
1D	4
2A	6
5A	2
5B	4
6A	12
6D	3
7A	20
7D	6

(CIMMYT Wheat Atlas), Carmona in Spain (van Ginkel and Ogonnaya 2007), and Chuanmai 28, 42, 43 and 47 in China (Yang *et al.* 2009).

One example of the improved water-use efficiency of synthetic wheat is the derived line D67.2/P66.270//AE. SQUARROSA (320)/3/Cunningham. Reynolds *et al.* (2006) showed that the improved water-use efficiency of this line in Mexico could be attributed to greater investment in root biomass at depth. When the same line was evaluated in north-western New South Wales at Narrabri, it showed greatly improved water-use efficiency compared with Cunningham, the adapted Australian cultivar used to develop this genotype (Atta 2013) (Fig. 1). Nevertheless, the development and exploitation of these materials is time-consuming, because much of the new diversity carries agronomically unfavourable traits such as poor threshability and poor grain-processing quality (Trethowan and van Ginkel 2009). The CIMMYT wheat-breeding program began making primary synthetics >25 years ago and derived lines have only recently been released in several countries as discussed earlier. While the vanguard synthetic materials were derived from crosses of *Aegilops tauschii* with modern durum wheat (*Triticum turgidum* var. *durum*), some recent primaries are based on crosses with cultivated and wild emmer wheat. The emmer wheats represent a considerable pool of underexploited genetic variation for a range of agronomically important traits, including heat and drought tolerance (Trethowan and Mujeeb-Kazi 2008). These emmer-based synthetics offer extended genetic diversity on all three genomes for many traits. However, emmer wheat can also be directly crossed to hexaploid wheat and AB genome recombinants identified.

The author believes that it is currently unnecessary to utilise wild wheat relatives such as the *Agropyron* and *Aegilops* species (*A. tauschii* being the exception) for water-use efficiency in wheat breeding, as little progress can be realised in the short to medium term and the associated costs are high. This will remain the situation until existing and easily exploitable genetic variation in the adapted and ancestral gene pools has been exhausted.

Coordinated and relevant phenotyping for plant response to limited moisture

The reliability and value of marker–trait associations are completely dependent upon the phenotype used. The importance of an accurate and relevant phenotype has been recognised for many years, as has the need for field-based screening. If plant response to limited moisture can be attributed to physiological traits, each with a higher heritability than water-use efficiency or drought tolerance *per se*, then

Table 3. Significant SNP markers linked to crown rot resistance in the three-way cross CSCR16/2-2-49/CUNNINGHAM//KENEDY/3/SUNCO/2*PASTOR based on association analysis of controlled environment and field symptom assessment
Chromosomes underlined carry the same significant SNP markers in both tests (GRDC annual progress report US000054, 2012)

Test conditions	Chromosomes carrying significant SNP markers linked to crown rot resistance
Controlled environment screening (adult plant pot tests)	<u>1AL</u> , 1BL, 1DL, 2AL, <u>2BL</u> , 2BS, <u>3AL</u> , 3DL, 4AL, <u>4BL</u> , <u>4DL</u> , 4DS, 5AL, 5AS, 5BS, 5DL, 6AL, <u>6BL</u> , 6DL, 7AS, 7BS
Field symptom expression	<u>1AL</u> , <u>2BL</u> , <u>3AL</u> , 3B(?), <u>4BL</u> , <u>4DL</u> , 5BL, <u>6BL</u>

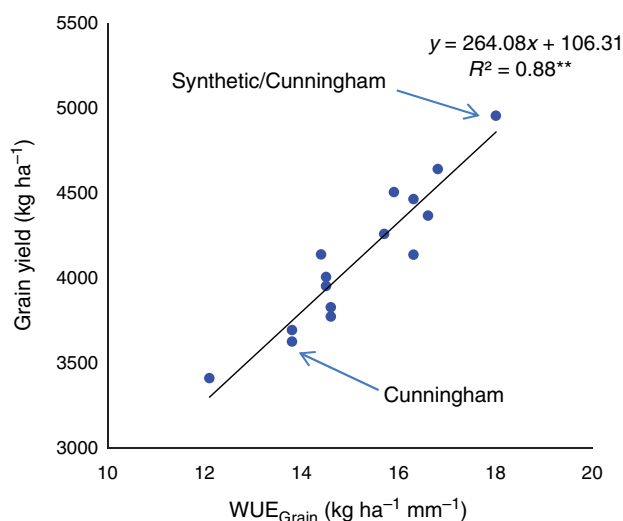


Fig. 1. Water-use efficiency (WUE) of 15 diverse genotypes including the synthetic derived line D67.2/P66.270//AE.SQUARROSA (320)/3/Cunningham (abbreviated Synthetic/Cunningham) based on three years of testing at Narrabri in NSW (adapted from Atta 2013).

response to selection will improve as will total productivity. The use of controlled-environment screening is not discounted; however, it must be used judiciously and only to maximise trait expression once the relevance of traits and trait combinations has been confirmed in the target environment through field-based screening.

Field-based screening is fraught with difficulty and greatly compromised by heterogeneity inherent in the screening location or in seasonal fluctuations. This can be managed through careful site selection, minimisation of heterogeneity through rotation, assessment of in-season weather, calculation of an accurate water balance, strategic deployment of rain shelters, and targeted stress treatments. The CIMMYT Wheat Program based in Mexico has deployed field-based screening for drought tolerance in the Sonoran Desert for many years, by using carefully managed sites and limited irrigation to generate stresses representative of target stress environments globally. The techniques employed are described in Trethowan *et al.* (2001). The advantage of desert-based screening (with ample available irrigation water) is that the same stress pattern can be generated each year, thereby improving response to selection over time. The only caveat is that the stress pattern generated is representative of the target environments; in the case of CIMMYT, these are the dry spring-wheat areas of the developing world. The strategy has been successfully used to develop materials globally adapted to water-limited environments in Mexico (Trethowan *et al.* 2002), and rates of genetic advance have been significant since 2001 (Manès *et al.* 2012) (Fig. 2).

However, a slightly different strategy to that employed by CIMMYT is required in regions that experience in-season rainfall. One such strategy is the Australian national Managed Environment Facility (MEF). The MEF comprises three locations that are representative of the three primary grain-growing regions of Australia. The premise for the MEF and a description of the strategy can be found in Rebetzke *et al.*

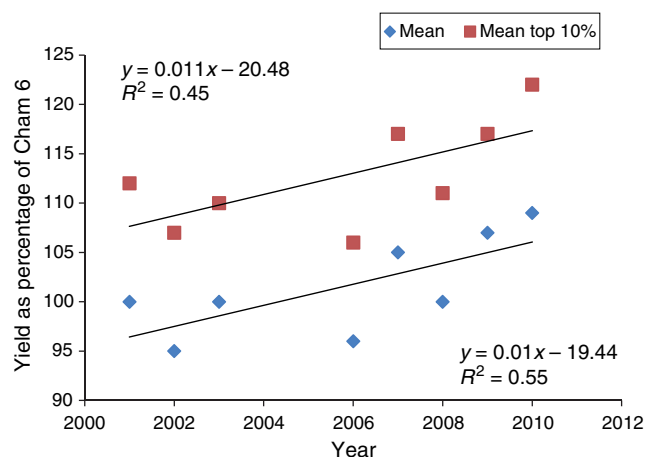


Fig. 2. Trial means and means of the top 10% of entries expressed as a percentage of Cham 6 in 216 low-yielding environments, for SAWYT 8 to 17 (2001–10). Adapted from Manès *et al.* (2012).

(2013). The MEF provides a network of representative sites across which physiological trait values are assessed and a physiological ideotype developed as a breeding target for each of the three grain-growing regions. The stress patterns generated at each regional MEF are based on the calculation of most probable stress patterns using 123 years of weather records and knowledge of the planting and harvesting dates for wheat (Chenu *et al.* 2013). Once trait weighting and optimised trait combinations are determined, it will be essential to develop linked markers to facilitate trait pyramiding in commercial wheat-breeding programs.

Programs similar to the Australian MEF example have also been established in India at four sites (Ludhiana, New Delhi, Powarkheda and Pune) and in China at four locations (Beijing, Hengshui, Yuncheng and Urumchi) (www.generationcp.org/wheat-dt-wheat-china; www.generationcp.org/wheat-dt-wheat-india). In addition to estimating trait values, plant breeders at these locations use the national MEF facilities to develop more water-use-efficient wheat cultivars using MARS schemes. The critical phenotype is determined at each carefully managed field site, and molecular marker pyramiding schemes are used to combine significant QTL effects at each location and across locations to develop both regionally and broadly adapted materials. It is expected that improved drought-tolerant cultivars developed using the MEFs will be released to farmers within the next 5 years in Asia. The availability of MEF sites in different countries and continents also offers an opportunity to develop truly broadly adapted materials based on trait combinations that are effective globally under dry conditions.

While MEF networks are powerful tools for assigning physiological trait values and for establishing phenotypes for molecular breeding strategies, it is also important to keep screening relevant to local farming practices at each location. For example, in northern New South Wales, >80% of grain growers have adopted conservation agriculture, in which tillage is minimised and crop residues are retained on the soil surface (Haskins 2012). This is a more water-use-efficient farming practice, and the total productivity of such

environments can be enhanced if water-use-efficient cultivars, better adapted to conservation agriculture, are deployed. To examine the adaptation of wheat to zero-tillage, a study was conducted to exploit genotype $\times$ tillage practice interactions and to identify QTLs linked to improved yield under zero-tillage (Trethowan *et al.* 2012). A mapping population was evaluated with and without tillage over 2 years and QTLs linked to superior response to zero-tillage were identified (Table 4). Although the inherent yield QTLs found on chromosome 5A were more significant, there were at least three significant QTLs, located on 2D and 5B, found only in progeny that performed well under zero-tillage. The 5B QTL is located in the same region as the *Tsn1* gene (Faris *et al.* 2010), which affects resistance to necrotrophic pathogens such as tan spot; these diseases tend to increase when crop residues are retained. Therefore, it should, be possible to pyramid these QTLs to enhance crop adaptation to conservation agriculture, thus enhancing overall system productivity.

Maximising the benefits of public good research

The CGIAR spends US\$900 million each year across 16 global research centres to reduce poverty and enhance the productivity

and sustainability of food production. Two primary centres are responsible for wheat, CIMMYT and ICARDA, and both now operate under a Cooperative Research Program (CRP) for wheat. Both centres house >200 000 unique germplasm accessions, including many ancestral forms that represent much of the available global genetic diversity for wheat. Given the recent enactment of biodiversity laws in most countries and the increasingly restricted movement of genetic resources across borders, these banks hold an increasingly important position in global public good research, as much of the available genetic diversity was collected before the imposition of these restrictions. Wheat breeders in the CRP have access to this diversity and the mandate and resources to introduce diversity for key traits, such as water-use efficiency, into adapted germplasm. This is a vital service for all global wheat improvement programs, because the costs of ‘taming’ such diversity are prohibitive, particularly for small regional programs in developing countries. It is much easier to transfer this diversity from agronomically superior materials to local germplasm than resorting to the original, unadapted source. Nevertheless, the importation and exploitation of these materials by most countries has been *ad hoc* at best, and considerable efficiencies and impact can be derived in the

Table 4. Significant QTL effects for yield under contrasting tillage regimes on two soil types in 2 years at Narrabri, New South Wales (adapted from Trethowan *et al.* 2012)

Markers in *italics* represent significant QTL under conventional tillage (CT) only; markers in bold type represent QTL significant in zero-tillage (ZT) only. Soil type either Grey Vertosol or Red Kandosol (Isbell 2002). Reproduced from Trethowan *et al.* (2012)

Chromosome	Marker interval	Treatment	Soil type	Additive effect %	Allele
1B	<i>gwm268/wPt-3475</i>	CT	Grey Vertosol	8	K
1B	<i>wPt-1313/gmw140</i>	CT	Grey Vertosol	10	K
1D	<i>cdf19/wmc216</i>	CT	Red Kandosol	10	K
2D	wPt-3728/cfd44	ZT	Grey Vertosol	9	K
2D	gmw484/wmc27	ZT	Red Kandosol	9	B
5A	<i>cfa2155/wPt1370</i>	ZT	Grey Vertosol	25	B
5A	<i>cfa2115/wPt1370</i>	CT	Grey Vertosol	14	B
5A	<i>cfa2115/wPt1370</i>	CT	Red Kandosol	9	B
5B	wmc99/wPt2373	ZT	Grey Vertosol	12	B

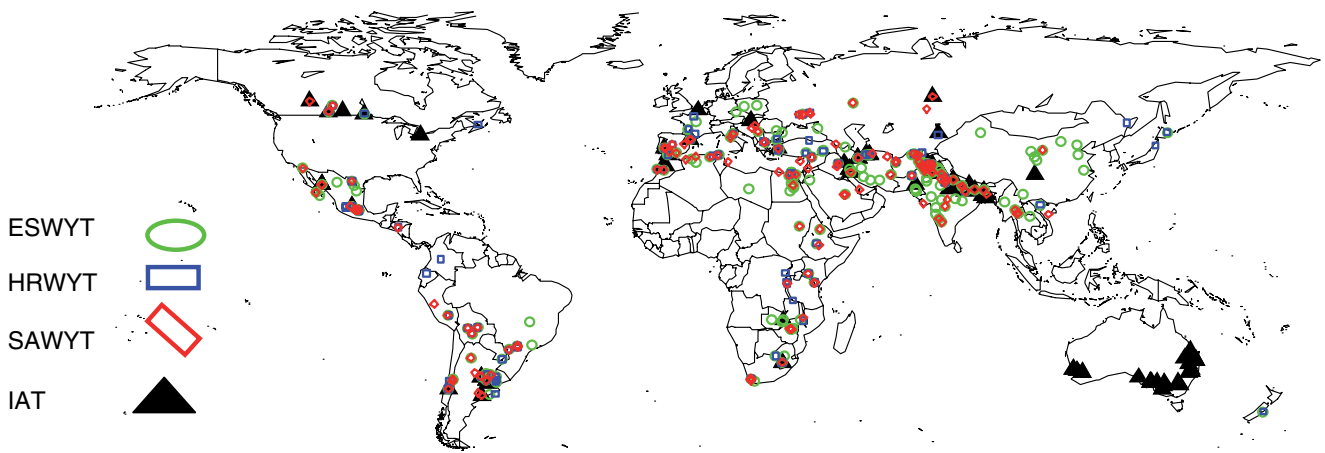


Fig. 3. Global distribution of the CIMMYT international yield trials between 1994 and 2004 and the distribution of the International Adaptation Trial (IAT) from 2001 to 2004. ESWYT, Elite Spring Wheat Yield Trial; HRWYT, High Rainfall Wheat Yield Trial; SAWYT, Semi-Arid Wheat Yield.

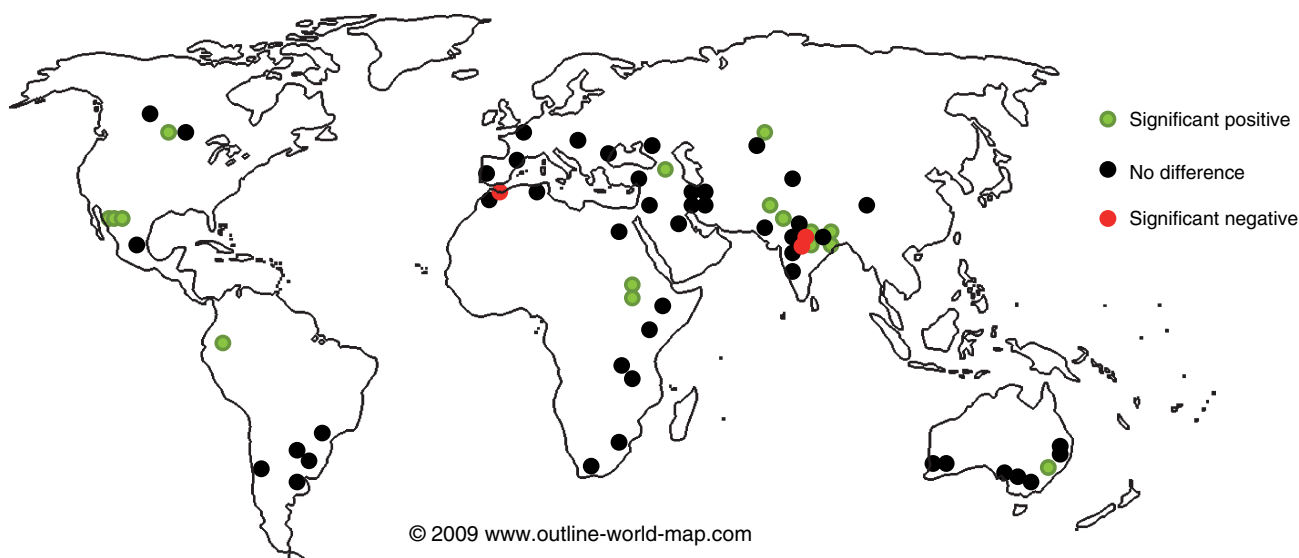


Fig. 4. Yield difference of the isogenic pair Gatcher GS50A and Gatcher at international trial locations in the International Adaptation Trial 2001–07. Simplified from Trethowan (2014).

short to medium term by introducing nationally coordinated strategies for exploitation of these resources.

One such example is the CIMMYT Australia ICARDA Germplasm Evaluation (CAIGE) program. The impact of wheat germplasm from CIMMYT in Australia has been significant and it can be traced to the introduction of the first semi-dwarf wheats in the 1970s (Syme 1970). Some estimates suggest that benefit of CIMMYT-derived wheat cultivars to the Australian wheat industry to be \$30 million per annum (Brennan 2007). However, this impact was realised despite the poor up-take of CIMMYT wheat germplasm by Australian breeders. Quarantine restrictions, limited post-quarantine availability and up-take, and piecemeal evaluation of materials significantly restricted impact.

The marginal exploitation of CIMMYT wheat germplasm in Australia was highlighted by the lack of trial information under Australian conditions. No Australian yield-trial data for lines imported from Mexico were sent to CIMMYT before 2001; it was therefore not possible to calibrate Australian locations with global wheat-growing environments. This shortcoming led to the establishment of an International Adaptation Trial (IAT) with the support of the Grains Research and Development Corporation (GRDC). This trial included 37 contrasting pairs of materials, many near-isogenic, for a range of traits including soil-borne pathogens, and is described in Mathews *et al.* (2007). The distribution of key CIMMYT nurseries, including the Elite Spring Wheat Yield Trial (ESWYT), Semi-Arid Wheat Yield Trial (SAWYT) and High Rainfall Wheat Yield Trial (HRWYT), and the IAT between 1994 and 2004 is described in Fig. 3. The IAT was eventually sown at 165 sites in 32 countries before being discontinued on 2007. The data were subsequently used to (i) estimate key determinants of genotype $\times$ environment interaction based on contrasting germplasm pairs, and (ii) calculate genetic correlations between key Australian breeding locations and global wheat environments.

Table 5. Summary of genetic correlations between Roseworthy in southern Australia and International Adaptation Trial locations based on 183 trials grown in 40 countries, 2001–04
Adapted from Mathews *et al.* (2011)

Region	Percentage of locations	
	Positively correlated with Roseworthy	Negatively correlated with Roseworthy
Asia	12	88
Europe	75	25
North America	39	61
Middle East	79	21
South America	100	0
Oceania	81	19
Other	40	60

An example of one pair of contrasting IAT lines, the near-isogenic pair for root-lesion nematode (*Pratylenchus thornei*) resistance, is described in Trethowan *et al.* (2010) and summarised in Fig. 4. Black dots represent trial sites where the isogenic pair did not differentiate for yield. Green dots indicate sites where the resistant line significantly outyielded the susceptible genotype. Clearly, a differential yield is only indicative of nematodes, and soil and root sampling is required to confirm the presence of the disease. Once these contrasts are overlain, a picture of the primary drivers of genotype $\times$ environment interaction becomes possible. Table 5 summarises the genetic correlations between Roseworthy, a historically important breeding location in southern Australia, and global IAT evaluation sites (Mathews *et al.* 2011). Locations in South America and the Middle East tend to be positively correlated with Roseworthy (as are many Australian sites, thus underpinning its importance to Australian wheat breeding), whereas sites in South Asia are negatively correlated. A breeder working at Roseworthy would therefore emphasise the acquisition of CIMMYT materials that perform well in South America and

Table 6. Site groupings based on principal component analysis of grain yield from CIMMYT Australia ICARDA Germplasm Evaluation (CAIGE) trials sown at nine locations in 2011

Group	Site	State	Mean rainfall (mm)	Latitude (south)	Longitude (east)
1	Narrabri	New South Wales	662 ^A	30.31	149.76
1	North Star	New South Wales	625 ^A	28.93	150.39
1	Wongan Hills	Western Australia	362	30.90	116.71
2	Toodyay	Western Australia	523	31.55	116.45
2	June	New South Wales	522	34.86	147.56
3	Roseworthy	South Australia	400	34.53	138.74
3	Horsham	Victoria	446	36.71	142.20
4	Esperance	Western Australia	620	33.86	121.89
4	Merredin	Western Australia	314	31.10	118.41

^ASummer-dominant rainfall pattern.

the Middle East, based on CIMMYT trial data, when accessing CIMMYT germplasm. The IAT highlighted the need to better coordinate the exploitation of this international germplasm, and the CAIGE program was subsequently established in 2007.

The CAIGE program was initially focused on CIMMYT materials and later extended to include elite lines and genetic resources from ICARDA. The two key CIMMYT wheat nurseries of greatest relevance to Australia, the ESWYT and SAWYT, and 100 elite ICARDA materials with semi-dwarf stature, appropriate phenology and white seed are imported each year. These materials are quarantined between November and May each year and at least 30 g of seed produced. A small amount of post-quarantine seed is used for adult-plant rust assessment (stem, leaf and stripe) and the remainder split between two sites, Narrabri in the east and a location in Western Australia, for subsequent multiplication in the year following introduction. Following multiplication, the materials are tested in yield trials at up to nine locations around Australia. At the same time, selected materials (based on parentage and data provided by the Wheat CRP) are tested locally for resistance to crown rot (*Fusarium pseudograminearum*), tan spot (*Pyrenophora tritici-repentis*) and *Septoria tritici*. Where possible, molecular markers linked to known resistance genes can be used to help reduce materials before disease screening. These data and the data generated by CIMMYT and ICARDA on the same materials are publicly accessible through the CAIGE website. Under the terms of a negotiated material-transfer agreement, anyone gaining access to the material within Australia is bound by contract to supply the information they generate to the CAIGE project. This process has significantly improved breeder up-take of materials, although quantification of this impact (apart from pedigree declaration in varietal releases) within the structure of commercial breeding programs is difficult to assess because the information is considered commercially sensitive.

Preliminary evaluation of CAIGE materials, based on 2 years of yield testing Australia-wide, indicates that high-yielding and adapted CIMMYT and ICARDA lines have been identified at all sites, although the frequency of adapted lines from both centres varies with site and year. Significant numbers of lines resistant to rust, crown rot, *Septoria tritici* and yellow leaf spot have been identified each year. Results from 2011, based on yield testing at nine locations nationally, grouped locations into four basic zones of adaptation (Table 6). Group 1 sites are lower

latitude and the SAWYT nursery tends to perform better whereas the ESWYT adapts best at wetter, higher latitude, group 2 sites. The ICARDA materials performed best at group 3 and 4 sites, which are primarily in the Mediterranean environments of southern Australia. A clear demarcation between northern and southern Australian environments appears to occur and confirms earlier observations of adaptation based on the IAT (Mathews *et al.* 2007).

Conclusion

The development and delivery of drought-tolerant (or water-use-efficient) wheat cultivars to farmers is an extensive subject and this paper attempts to identify three basic efficiencies that can enhance the process in the short to medium term. Clearly, it is easier to exploit genetic variation for plant response to limited moisture in the adapted gene pool because progress will be faster and the science of genomics and access to cost-effective genotyping services improves the accuracy and efficiency. However, this does not preclude the search for genetic diversity in wild wheat relatives or the wild relatives of all crop species. This work will have significant payoff in the longer term and such research or pre-breeding should be conducted in parallel with commercial breeding.

The establishment of MEF sites is critical if rates of progress in water-use efficiency or drought tolerance are to be realised. However, as discussed earlier, controlled environment testing can add value to field-based phenotyping, particularly if greater trait resolution is required for fine mapping. Nevertheless, trait value should first be determined in the field to avoid expensive determinations of irrelevant traits. The ultimate solution to increasing productivity is a combination of water-use-efficient agronomic practices and water-use-efficient cultivars better adapted to these practices.

The optimised exploitation of crop germplasm from the CGIAR will potentially give local breeding programs the greatest improvement in genetic gain in the short to medium term. This paper focused on the Wheat CRP materials simply because they are publicly available and accessible to all. Similar strategies can be used for a wide range of international germplasm (if accessible locally), and coordinated quarantine, evaluation and information exchange can be very powerful.

The three strategies described above will improve the identification of drought-resistant or water-use-efficient

cultivars for those who need it most: farmers. However, another vital link in the delivery chain is seed production and dissemination. This process is greatly influenced by country, crop and culture, and local strategies and policies are required to optimise the up-take of superior cultivars by farmers. Some have advocated farmer participatory plant breeding (Sthapit *et al.* 1996), although the accuracy of phenotypes for drought-stress response in such circumstances is marginal and the efficiency of molecular breeding will be limited. Private industry is generally very successful at delivering seed to farmers, although these tend to be more viable with hybrid crops such as maize. Nevertheless, it is vital that local strategies are established to ensure the delivery of superior cultivars to farmers.

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