Cereal variety mixtures reduce inputs and improve yield and quality - why isn't everybody growing them?

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Cereal variety mixtures, i.e. several varieties of the same species such as barley, sown mixed together, offer many potential benefits to the grower, namely:

- yield increases of between 5 and 15%
- reduced pesticide inputs
- improved grain quality
- stability of yield and quality[1]

Unfortunately, agricultural and industrial end users have concentrated on certain perceived disadvantages, so mixtures have been deployed infrequently in UK agriculture. Recent research, however, offers further evidence of the advantages of mixtures, and new methods to overcome the perceived problems, so it may shortly become opportune for the grain trade to re-consider its position.

How do variety mixtures work?

Disease control One of the main ways cultivar mixtures reduce inputs is by limiting spread of disease, thereby reducing or eliminating the need for pesticide applications. In the case of a pathogen such as mildew, only specific races with matching virulence (lack of an avirulence gene product which can be recognised by the host) can infect a variety with a specific resistance gene. If that race does not have matching virulence to another variety in the mixture with a different resistance gene, then it will induce resistance in that variety. It cannot therefore grow and produce spores for further infection on the resistant variety, and the resistance induced will even reduce the amount of infection by other normally virulent races on that variety. The epidemic is therefore slowed by two spatial effects in addition to the induced resistance; there are fewer susceptible hosts and the resistant host variety provides a barrier to reduce successful transmission of spores to the next susceptible host variety.

Variety mixtures are also effective against pathogens such as *Septoria tritici* and *Stagonospora nodorum*, which do not have the highly specific gene-for-gene interactions common in biotrophs such as mildew. Mechanisms to explain their effects still depend on the disadvantageous effect on a pathogen of changing from one host variety to another resulting in reduced infection overall.

Yield Reduced disease would be expected to result in a corresponding yield increase, this being the primary objective of the use of mixtures, but, in practice, the use of variety mixtures often gives more yield increase that would be expected from the level of disease control. Clearly, the yield response is not simply due to a reduction in the loss of grain filling through assimilate diversion to a pathogen or damage to photosynthetic capacity. It is also due to competition and yield compensation effects within and between the components of the mixture. Whilst a mixture may be sown as, for example, three varieties in equal proportions, the harvested grain proportions may be considerably distorted if a particular component is highly competitive. If that component is intrinsically low yielding, this could even result in reduced yield compared with the mean of the equally weighted components as sown. More often, however, the yield is greater, not only through the same effect of a competitive variety, but also through better resource exploitation overall. A single cultivar may not exploit all the available root or aerial environment for nutrient and light capture at any one time. Within the heterogeneous components of a mixture there is likely to be a component ready to exploit the available resources much more of the time, to the overall yield benefit of the mixture.

The effect of increased yield and disease reduction might be expected to show some relationship to the degree of heterogeneity in the mixture (Fig. 1). Heterogeneity is most easily manipulated by changing the number of component varieties and indeed there are

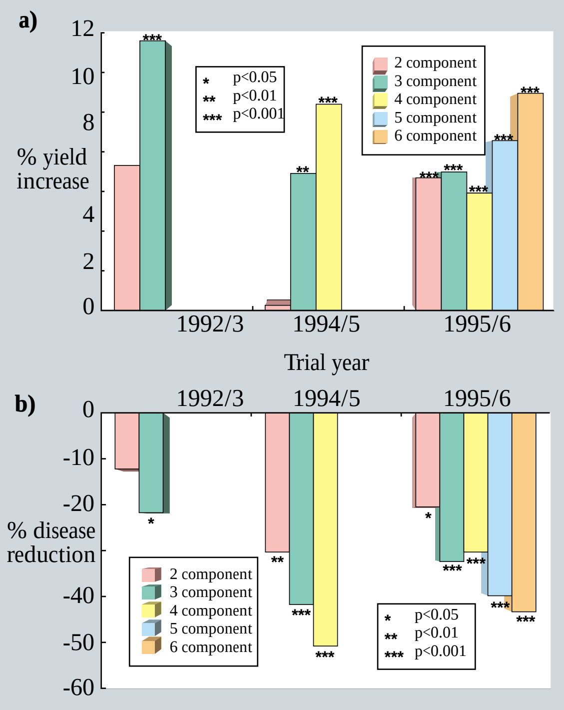

Figure 1 Reduced *Rhynchosporium* infection and increased yield corresponding with number of component varieties in winter barley mixtures. a) Increase in yield in relation to mixture component number. b) Disease reduction in relation to mixture component number.

significant correlations between increased component number and both disease reduction and increased yield, indicating that the higher the number of components the better[2]. However, a large number of varieties is impractical on farm and the number of agronomically compatible varieties available is limited. Whilst the performance of two component mixtures is somewhat variable, three component mixtures are both more reliable, practical, and achieve a high proportion of the disease control seen in much more complex mixtures.

Quality Barley cultivar mixtures are currently grown in Scotland but only as a 6-row high yielding feed quality cultivar mixed with a 2-row to increase the specific weight, i.e. to increase quality. They also tend to reduce lodging. In general, it has been assumed that quality factors such as those required for malting, will be reduced in a mixture compared with the mean of the component varieties. Nevertheless, until our recent studies, there has been very little published evidence to verify or contradict this assumption and several countries, such as Denmark and Poland, have found mixtures can be quite satisfactory for brewing. Our research also indicates that they can be very advantageous for use in malting.

Objections to mixtures

Increased heterogeneity in the malt Maltsters have been reluctant to use barley mixtures for three reasons: increased heterogeneity, verification problems, and customer preference. The first arises as maltsters 'fine tune' their systems to gain optimum performance from a specific cultivar. Therefore, the imposed regime will not be best suited to genotypes which differ in their rate of malting. Too rapid modification, or breakdown, of the endosperm structure will lead to the loss of fermentable material to the growing embryo (malting loss). Slow modification will not degrade sufficient cell wall material and protein to permit ready access of enzymes to all the starch. In addition, cell wall residues can cause viscosity problems and poor filtration. Mixtures are perceived as giving an uneven or heterogeneous malt due to differences between components in malting behaviour.

Verification and customer preference A system which permitted mixtures would require to be effectively controlled. The components and their proportions should be quantifiable and the mixture should be demonstrably what it is claimed to be. Distinguishing between grain supplies of different cultivars may be very difficult using morphological characters. Electrophoresis of storage proteins (hordeins) enables grouping of cultivars rather than individual cultivar identification and is useless for malt samples as storage proteins are substantially degraded. Both domestic and export markets favour monocultures and sales maltsters are frequently required to give assurances to customers. Mixtures are, therefore, not easy to sell at present, but there is considerable evidence that questions the validity of these objections.

Validity of objections For most malting parameters, mixtures give equivalent results to the mean of their components, so any components of inferior quality significantly increase heterogeneity and adversely affect other aspects of malting performance. However, experience, especially in Eastern European countries such as Poland and East Germany, has suggested that mixtures of malting quality cultivars can give acceptable performance in both maltings and brewhouse.

Varietal purity does not guarantee homogeneous grain samples. Even within a field, there may be differences in drainage and soil type, while grains may differ depending on position on the ear or location on main or side tillers. Maltsters do not work with homogeneity, but within an acceptable range of heterogeneity. The range of heterogeneity in well designed mixtures

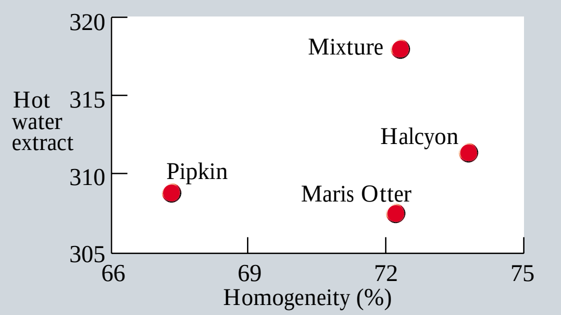

Figure 2 Improved hot water extract and homogeneity of cell wall modification in a mixture of winter barley varieties Pipkin, Maris Otter, and Halcyon.

is likely to be less than the heterogeneity amongst grain from the same variety grown in different places.

Work at SCRI, with a mixture of three cultivars derived from similar pedigrees, showed no significant increase in heterogeneity compared to any of the monocultures (Fig. 2). This was determined by a laboratory test for the evenness of modification of cell walls (Fig. 3). In addition a slight, but significant, increase in extract was observed in the mixture[3].

Exploitation of mixtures

Track record Variety mixtures have been extensively used in low input agriculture, and to a lesser extent in high input situations. Land races are locally grown selections of crops, which are heterogeneous and normally contain a mixture of resistance genes that have proved both durable, sufficiently limiting, or tolerant of the prevalent diseases. Land races rarely achieve anywhere near the yield possible with modern varieties, but the latter are dependent upon high fertiliser and, when resistance breaks down, pesticide inputs.

Figure 3 Assessing eveness of barley cell wall modification following malting using a fluorescent dye.

By contrast, the resistance of land races remains stable and they may be better at exploiting lower soil fertility. The essential feature that leads to these desirable features is not the low harvest index, but diversity, and this can be introduced and manipulated to optimise its effectiveness using mixtures of agronomically more useful varieties with high harvest index.

Cereal variety mixtures are grown extensively in several countries where they have been supported by research, notably the USA, Poland, Denmark and Switzerland. Barley mixtures have achieved high malting quality and are used for beer production in some European countries. The biggest problem is not achieving good quality, yield or disease resistance, but convincing end-users that there are not only few, if any, disadvantages to using mixtures, but also many advantages. Unfortunately one of the disadvantages is self-imposed by the end-users. Legislation restricts the sale of mixed seed and maltsters are prevented from trading in mixed varieties by their own rules.

In Switzerland, the ‘Extenso’ scheme was introduced to reduce pesticide inputs. Farmers are paid a subsidy to grow crops under a low fertiliser, no pesticide regime. Given this restriction farmers rapidly adopted variety mixtures, primarily for disease control but they quickly found the other benefits. Not least of these benefits was a price premium they received as consumers demanded products produced under such conditions. The beneficial implications of growing mixtures may extend beyond the rationale for their use once the less direct economic implications are considered.

Development

Modelling Many mathematical models have been developed to explain the host-pathogen interactions which take place within mixtures resulting in reduced disease levels. These have led to new strategies for optimising mixture composition. There is still much that is not understood about these pathological interactions, and the nature and contribution of the induced resistance component is the subject of current research by SCRI and BioSS. Yield and yield-loss modelling is yet more complex and the integration of yield, yield-loss and disease progress modelling in mixtures is necessary to disect out the effects and optimise these complex interactions. The stochastic modelling techniques we are using to understand induced resistance are likely to be the best route into this intractable jungle.

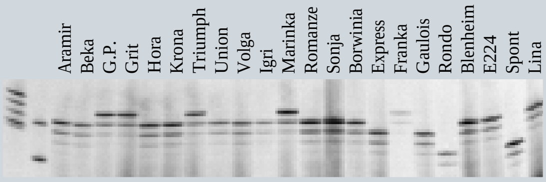

Figure 4 A simple sequence repeat DNA marker showing differences between barley varieties.

Molecular markers Protein characterisation through electrophoresis has been used to distinguish cereal varieties for several years. However, it is generally non-quantitative and not all varieties can be separated unambiguously, so contamination may not be detected. Modern DNA molecular markers, particularly 'microsatellites' or Simple Sequence Repeats (SSRs), are able both to distinguish all varieties[4], and to quantify any contamination. Using only four SSRs, it has been possible to discriminate between all varieties so far tested (Fig. 4). SSRs can therefore be used to identify or verify the composition of a mixture, overcoming a major objection of maltsters that they need to be able to be sure that a grain sample is of the specified composition. The tests are not yet rapid enough for use on grain samples as they arrive in lorries at the maltings, but such technological improvements are likely to be forthcoming. In addition, as DNA is unaffected by the malting process, molecular markers are equally applicable to verifying malt composition.

Perhaps the most important observation from our work on malting quality mixtures is that they were obtained from mixtures of available malting quality cultivars. No attempt was made to optimise the mixtures. Amongst the mixtures were some where the malting quality exceeded the mean of the components. In particular, these were related varieties deriving their quality component *via* a similar pedigree. It may therefore be possible to select components for mixtures that will have consistently better malting quality than any of the component varieties. If these criteria are brought together with optimum combinations for disease reduction and yield enhancement, substantial gains could be achieved very quickly. If combining ability for these attributes were included as criteria in breeding programmes, the gains could be still greater. All these characteristics are potential targets for manipulation using molecular markers closely linked to the desired traits.

Molecular markers are increasingly being used in breeding programmes. The marker is frequently not in the gene responsible for the character being selected, but close by. Many of the genes which contribute to quantitative characters, such as malting quality parameters, can be mapped as quantitative trait loci (QTL) and nearby markers can be used to select for these in breeding[5]. For example, there are markers for six loci controlling fermentability, the factor that determines how much of the malt extract can be fermented into alcohol[6]. Markers could similarly be used for marker-assisted component selection for designing mixtures, as favourable components may combine in a similar manner to favourable alleles in a single cultivar and could explain the high extract levels in certain mixtures.

Potential exploitation in Scotland and the UK In addition to research data, there is need to provide evidence of the commercial value of mixtures, to overcome customer resistance. Molecular markers are likely to be adopted as determinants of varietal purity and extension to mixtures will be a logical extension in a research context. Demonstration of commercial potential, however, requires a crop that is grown over a fairly extensive area, but in which varietal purity is not essential. The world record wheat yield of 13.99 tonnes per hectare was achieved with an equal proportion mixture of the varieties Virtue, Mardler and Husler grown on Mr Gordon Rennie's farm at Clifton Mains in Midlothian in 1981, demonstrating their practical agronomic advantage. Winter wheat is grown in Scotland largely as a source of starch for conversion to alcohol in grain distilling. Unlike bread wheat, high protein levels are undesirable, so distilling wheat is suited to low input systems. The choice of wheat in preference to maize is made by distillers on purely economic grounds, as there is no advantage in alcohol yield, so the optimum raw material may be defined as that which gives the highest spirit yield per unit cost. Although some varieties have been preferred over others, the reasons for this are not clear, so this would seem to be an area where mixtures could be exploited successfully.

In the longer term, benefits of malting barley mixtures of closely related cultivars could offer a means of obtaining malts with particular specifications, not met by most commercially available genotypes. At present, for example, only a few cultivars have low levels of glycosidic nitriles (GN), which undergo a series of chemical reactions during fermentation and distillation, leading to traces of an undesirable component of Scotch whiskies. Tighter regulation or the absence of suitable new cultivars could make distillers seek to extend the commercial lifespan of some existing cultivars. Varietal mixtures could offer a means of achieving this whilst maintaining profitability, especially as all low GN genotypes can be traced back to a common ancestor, so may demonstrate some similarity in malting behaviour. High diastase varieties are also required for grain whisky distilling. It may be possible to obtain sufficient expression of such characteristics from certain components of a mixture, whilst still maintaining high expression of other desirable characteristics from other components.

Mixtures - the answer to arable farming needs?

Mixtures may not be the full answer for farming needs, but could make a very significant contribution, which is being neglected for the wrong reasons. Mixtures have tended to get consigned to the 'alternative technology' box along with 'organic' agriculture and other 'environmentally friendly' or 'politically green' approaches. Mixtures should be regarded as an approach based on sound scientific principles applicable to many agricultural situations. There are many benefits in their use in low input and 'organic' situations where there are a lack of alternative approaches for controlling disease. Their potential and economic impact is likely to be far greater, however, in mainstream agriculture, where benefits from using the best products of modern breeding programmes and crop production techniques can be further enhanced both in their direct yield response and reliability. Biodiversity provides insurance against unforseen environmental effects. Variety mixtures is an approach that builds this protection into agricultural practice rather than keeping it in store for use in the event of disaster. Mixtures do not remove the requirement for pesticides but may enhance their effectiveness and reduce the level of active ingredient required for reliable effect.

These benefits are achievable with existing varieties and agrochemicals. Available molecular biological tools will enable verification of mixture composition to overcome end-users objections, which can then lead to removal of the legislative hurdles to exploitation. Once routine use in agriculture is established, the incentive will be present to use molecular biological tools in selecting components for mixtures, and for breeding varieties suitable for exploitation in mixtures. Whilst genetic manipulation offers the potential for major advances in disease resistance and yield in many crops, mixtures offer the opportunity to achieve further major improvements in exploiting both new and existing crop varieties. Not least of these is stability of yield and quality, an increasingly important criterion where margins are being squeezed.

In summary

Variety mixtures offer a fast method to exploit all the benefits of modern research, breeding, and agronomic advances whilst providing increased insurance and stability. They offer increased yield, reduced inputs, particularly of pesticides, and improved quality. With relatively low levels of development funding, considerable economic advantage could be achieved. Investment in exploitation of molecular methods and modelling studies would enable much more of the potential of biodiversity to be unlocked and optimised for use in mainstream agriculture. Compared with many other 'environmentally friendly' approaches, use of variety mixtures is likely to have a far greater beneficial effect on the environment as it could be readily adopted for use over a large proportion of cereal growing areas of the world. Perhaps most importantly for its prospects for adoption, it is a method of production that benefits the farmer who will receive direct economic benefit from increased yields and reduced pesticide inputs.

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