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RESEARCH PROJECT CO-FINANCED BY THE COMMUNITY PLANT VARIETY OFFICE (CPVO): "MANAGEMENT OF WINTER OILSEED RAPE REFERENCE COLLECTIONS"

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BMT/9/8 page 2

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Background:

1. NIAB has been carrying out a research project investigating the use of DNA microsatellites (simple sequence repeats, SSRs) for DUS testing of oilseed rape (OSR). The initial phase of the project utilised SSR markers made available by Long Ashton Research Station (LARS, Bristol) under a BBSRC programme. A number of these (49) were screened, to produce an initial set of 15, which were polymorphic within a set of 10 oilseed rape varieties. The analysis of these 15 was then optimised for use on two different detection platforms, and their utility compared by analysing 48 individuals of the 10 varieties, at both LARS and NIAB. Factors investigated included the ability to analyse the distinctness and uniformity of varieties, but also the ease of analysis and scoring of products, the accurate detection of products, simultaneous analysis (multiplexing), and map position. Comparability between the different automated systems in use at NIAB and LARS was an important element of the programme. The results showed that D and U could be assayed with these SSRs, but also that there were concerns about 5 of them, in terms of the reproducibility of the data using different detection systems.

2. In Phase 2 of the project, an additional 5 SSR markers were optimised and tested to replace the unreliable markers. The 5 new markers were used to analyse D and U of the same 10 varieties as previously. The objective was to produce a set of 15 well characterised SSRs, which can be reliably scored independent of the detection platform, and which could be used for further study.

3. In parallel with this work, colleagues at GEVES (France) were also evaluating some of these markers, and others from the BBSRC set, in a separate series of experiments. The preliminary results from both of these projects indicated that it would be useful to consider a combined set of markers, applied to the analysis of a larger variety collection.

Project Proposal:

4. Although varying approaches to DUS testing of OSR are taken within Europe, there are a number of problems common to all of the countries where OSR is a major crop. The number of candidate varieties entered for DUS testing is increasing annually, and the existence of different types of varieties (lines, synthetics, hybrids of various kinds, GMOs) complicates the DUS trial design and increases its complexity. For instance, the trials have to contain the parent lines of hybrids, whether or not they are themselves entered for PBR/NL purposes, since a description of them is needed for certification purposes and in some countries, distinctness of the hybrid can be based on the distinctness of the parents or on the parental formula. A major problem arises from the increasing size of variety reference collections and the requirement to compare new varieties with those whose existence is a matter of "common knowledge" at the time of application. Within the EU context, this includes all varieties with European rights (c. 145 currently) and/or listed on the Common Catalogue (c. 560). To include this many varieties in a replicated field trial is logistically and financially onerous.

BMT/9/8 page 3

However, it is desirable that in order to maintain the strength of protection offered by PBR, the principle of comparing new varieties with those of common knowledge should be upheld, and that variety reference collections should be as comprehensive as possible. Clearly, some means of "managing" reference collections is highly desirable, i.e. comparing candidate varieties with the reference collection prior to sowing the field trial, in order to on the one hand reduce the number of varieties that need to be grown whilst on the other, not erode the quality of PBR.

5. One attractive means of such management would be to use molecular markers (DNA profiling) to compare new varieties with the profiles of those in a database, eliminate those which do not need to be compared in a field trial (according to pre-defined criteria) and then only grow the most similar varieties for detailed DUS testing. At the moment, it has been agreed within UPOV that such a scheme would be acceptable if it could be demonstrated that there is a relationship or association between the variety "distances" as calculated using molecular markers ("genetic distance"), and those calculated based on the phenotypic markers currently used in DUS testing, the "Option 2" approach. One difficulty with such an approach is the lack of suitable data sets to test the models. In OSR, SSRs have not been applied on the scale necessary. Thus the objective of this project is to generate a large (>400 varieties) data set of DNA profiles of OSR varieties, using a common set of c. 25 - 30 SSR markers, and then utilise these data in a range of ways to explore the possibilities for managing OSR reference collections.

Current Situation:

6. In 2003/04, NIAB and GEVES, in collaboration with DIAS (Denmark) and BSA (Germany) proposed a project to the Community Plant Variety Office (CPVO), who agreed to co-finance (in conjunction with the relevant national authorities) a 3-year programme based on the use of an agreed set of SSRs to analyse a large variety collection from the four countries, to measure the genetic distances between these varieties in a number of ways, and to compare these with phenotypic distances. This would thus be a UPOV Option 2-type approach.

7. The project started in January 2005, and NIAB, GEVES and DIAS have been undertaking a series of preliminary experiments, evaluating the available markers. The results were compared at the first meeting held recently in Cambridge, and agreement reached as to which markers to accept, which to abandon and which required further study. An agreed set will be finalised by August 2005, along with an agreed variety collection.

8. The presentation will consider some of the more recent NIAB data, and outline the project and anticipated timetable.

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