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### INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS GENEVA

## WORKING GROUP ON BIOCHEMICAL AND MOLECULAR TECHNIQUES AND DNA-PROFILING IN PARTICULAR

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ESSENTIAL DERIVATION IN LETTUCE

Document prepared by experts from the International Seed Federation (ISF), Nyon, Switzerland

# Essential Derivation in Lettuce The work of the International Seed Federation B. Le Buanec et al<sup>1</sup> ISF

In 1994, ISF (ASSINSEL at that time) made a study on essential derivation in Tomato. However, as today most of the tomato varieties are hybrids, the results were not very relevant.

In 2000, it was decided to have a new study on a self-pollinating species, Lettuce, and a working group was established.

Three cultigroups of lettuces were chosen: Greenhouse, Field Summer and Salinas Iceberg. The choice of molecular markers was AFLP. Morphological markers were also analyzed for the Greenhouse and Field Summer varieties, based on the DUS characteristics given by the Plant Variety offices in France and the Netherlands.

#### **RESULTS**

#### Molecular markers

The AFLP markers allow a very clear clustering of the 3 cultigroups

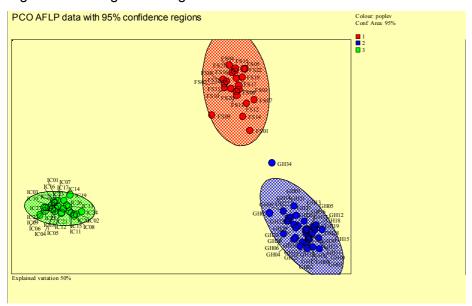
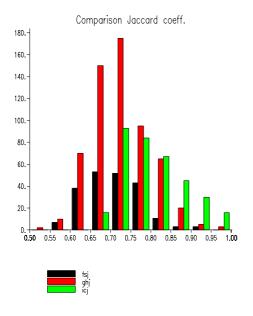


Fig. 1: Clustering according to AFLP data

and show a variability among the varieties presently on the market (Fig. 2) with a range of the Jaccard coefficient of similarity from 0.52 to 0.97 for the Greenhouse type, from 0.56 to 0.92 for the Field Summer type and from 0.69 to 1.00 for the Iceberg type. As expected, the variability within the Iceberg type is lower.

Composition of the Working Group: M. Bruins, O. de Ponti, T. Kramer, , B. Le Buanec, D. Lor, F. Moquet, Kees Reinink, F. van Eeuwijk, M. van Grinsven.

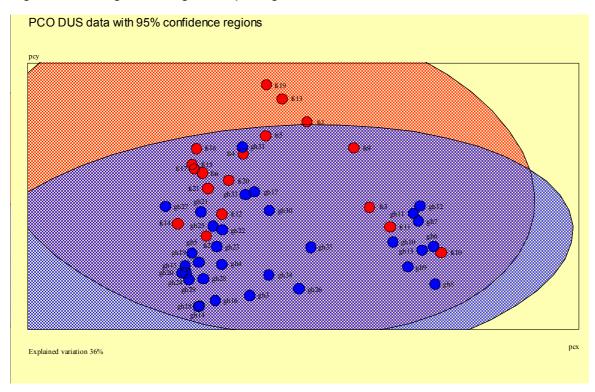
Fig. 2: Variability within commercialized varieties



#### Morphological data

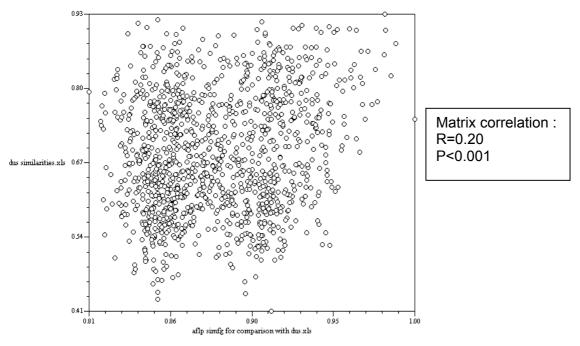
There are some differences between the two cultigroups Field Summer and Greenhouse, but a significant overlapping

Fig. 3: Clustering according to morphological data



#### **Correlation between DNA and Morphological markers**

Fig. 4: Correlation between the DNA markers and the morphological markers



That correlation is very weak.

#### **Essential derivation**

The working group considered three possibilities to define a threshold that could be a trigger point for starting a dispute on essential derivation:

- the Upper tail principle
- the Outlier principle
- the Pedigree principle

and opted for the upper tail principle.

The results are presented in Fig. 5, 6 and 7.

Fig. 5: Field Summer

zone	q	genox	genoy	compx	compy	all	errormargin
2	0.9982	8	2	4	2	1.0000	0.0000
2	0.9954	18	17	6	7	1.0000	0.0000
2	0.9925	20	2	8	2	1.0000	0.0000
2	0.9897	20	8	8	4	1.0000	0.0000
2	0.9868	10	9	4	4	0.9967	0.0043
2	0.9840	25	9	9	4	0.9935	0.0055
2	0.9811	25	6	9	4	0.9930	0.0064
2	0.9783	25	22	9	9	0.9920	0.0075
2	0.9754	22	9	9	4	0.9919	0.0055
2	0.9726	23	9	9	4	0.9919	0.0054
2	0.9698	22	6	9	4	0.9913	0.0065
2	0.9669	9	6	4	4	0.9912	0.0066
2	0.9641	23	10	9	4	0.9903	0.0051
2	0.9612	25	10	9	4	0.9887	0.0095
2	0.9584	10	6	4	4	0.9877	0.0075
2	0.9555	22	10	9	4	0.9872	0.0079
2	0.9527	15	2	6	2	0.9858	0.0126
1	0.9498	15	8	6	4	0.9858	0.0126
1	0.9470	20	15	8	6	0.9857	0.0126
1	0.9441	16	10	6	4	0.9856	0.0182
1	0.9413	25	23	9	9	0.9855	0.0083
1	0.9384	23	6	9	4	0.9843	0.0095
1	0.9356	23	22	9	9	0.9841	0.0089
1	0.9327	6	5	4	3	0.9837	0.0103
1	0.9299	21	5	8	3	0.9835	0.0092
1	0.9270	16	9	6	4	0.9824	0.0182
1	0.9242	5	4	3	3	0.9816	0.0111
1	0.9214	21	4	8	3	0.9810	0.0111
1	0.9185	9	5	4	3	0.9796	0.0070
1	0.9157	23	18	9	6	0.9792	0.0100
1	0.9128	22	5	9	3	0.9779	0.0108
1	0.9100	22	3	9	3	0.9779	0.0112
1	0.9071	25	5	9	3	0.9778	0.0107
1	0.9043	26	20	9	8	0.9778	0.0100
1	0.9014	23	17	9	7	0.9770	0.0096
0	0.8986	26	8	9	4	0.9763	0.0101
0	0.8957	26	2	9	2	0.9763	0.0101
0	0.8929	22	16	9	6	0.9763	0.0179
0	0.8900	5	3	3	3	0.9762	0.0112

FS15, 16 and 17 were sister lines in the F3

Fig. 6: Greenhouse

zone	q		genoy	compx		all	errormargin		
2	0.9990	22	21	5	5	0.9985	0.0028		
2	0.9973	19	15	4	4	0.9878	0.0079		
2	0.9956	11	8	3	3	0.9850	0.0073		
2	0.9939	13	11	3	3	0.9816	0.0093		
2	0.9922	10	8	3	3	0.9759	0.0095	range	0.0449
2	0.9906	35	30	6	6	0.9740	0.0159	median se	0.0220
2	0.9889	13	8	3	3	0.9717	0.0135		
2	0.9872	27	25	5	5	0.9702	0.0132		
2	0.9855	12	8	3	3	0.9686	0.0154		
2	0.9838	11	10	3	3	0.9684	0.0120		
2	0.9822	18	16	4	4	0.9671	0.0128		
2	0.9805	35	31	6	6	0.9669	0.0109		
2	0.9788	12	2	3	1	0.9667	0.0141		
2	0.9771	31	30	6	6	0.9651	0.0107		
2	0.9754	30	18	6	4	0.9651	0.0138		
2	0.9738	13	12	3	3	0.9650	0.0158		
2	0.9721	12	10	3	3	0.9639	0.0142		
2	0.9704	20	16	4	4	0.9619	0.0108		
2	0.9687	16	14	4	4	0.9618	0.0124		
2	0.9670	12	11	3	3	0.9612	0.0129		
2	0.9654	35	18	6	4	0.9593	0.0169		
2	0.9637	8	2	3	1	0.9587	0.0160		
2	0.9620	13	10	3	3	0.9582	0.0160		
2	0.9603	33	18	6	4	0.9580	0.0196		
2	0.9586	35	33	6	6	0.9579	0.0154		
2	0.9570	30	14	6	4	0.9570	0.0159		
2	0.9553	31	16	6	4	0.9565	0.0099		
2	0.9536	27	24	5	5	0.9553	0.0154		
2	0.9519	20	14	4	4	0.9538	0.0165		
2	0.9502	17	15	4	4	0.9536	0.0185		
1	0.9486	10	2	3	1	0.9534	0.0208		
1	0.9469	33	31	6	6	0.9524	0.0201		
1	0.9452	33	30	6	6	0.9519	0.0152		
1	0.9435	31	29	6	6	0.9511	0.0208	range	0.0087
1	0.9418	12	3	3	1	0.9507	0.0181	median se	0.0282
1	0.9402	35	16	6	4	0.9503	0.0146	1	
1	0.9385	30	16	6	4	0.9501	0.0161		

GH21 and GH22 were selected in the same F4

GH8 and GH11 come from same F3

GH30 and GH35 come from same F3

GH27 is from a cross involving GH25

Fig. 7: Iceberg

zone	q	genox	genoy	compx	compy	all	errormargin
2	0.9982	8	2	4	2	1.0000	0.0000
2	0.9954	18	17	6	7	1.0000	0.0000
2	0.9925	20	2	8	2	1.0000	0.0000
2	0.9897	20	8	8	4	1.0000	0.0000
2	0.9868	10	9	4	4	0.9967	0.0043
2	0.9840	25	9	9	4	0.9935	0.0055
2	0.9811	25	6	9	4	0.9930	0.0064
2	0.9783	25	22	9	9	0.9920	0.0075
2	0.9754	22	9	9	4	0.9919	0.0055
2	0.9726	23	9	9	4	0.9919	0.0054
2	0.9698	22	6	9	4	0.9913	0.0065
2	0.9669	9	6	4	4	0.9912	0.0066
2	0.9641	23	10	9	4	0.9903	0.0051
2	0.9612	25	10	9	4	0.9887	0.0095
2	0.9584	10	6	4	4	0.9877	0.0075
2	0.9555	22	10	9	4	0.9872	0.0079
2	0.9527	15	2	6	2	0.9858	0.0126
1	0.9498	15	8	6	4	0.9858	0.0126
1	0.9470	20	15	8	6	0.9857	0.0126
1	0.9441	16	10	6	4	0.9856	0.0182
1	0.9413	25	23	9	9	0.9855	0.0083
1	0.9384	23	6	9	4	0.9843	0.0095
1	0.9356	23	22	9	9	0.9841	0.0089
1	0.9327	6	5	4	3	0.9837	0.0103
1	0.9299	21	5	8	3	0.9835	0.0092
1	0.9270	16	9	6	4	0.9824	0.0182
1	0.9242	5	4	3	3	0.9816	0.0111
1	0.9214	21	4	8	3	0.9810	0.0111
1	0.9185	9	5	4	3	0.9796	0.0070
1	0.9157	23	18	9	6	0.9792	0.0100
1	0.9128	22	5	9	3	0.9779	0.0108
1	0.9100	22	3	9	3	0.9779	0.0112
1	0.9071	25	5	9	3	0.9778	0.0107
1	0.9043	26	20	9	8	0.9778	0.0100
1	0.9014	23	17	9	7	0.9770	0.0096
0	0.8986	26	8	9	4	0.9763	0.0101
0	0.8957	26	2	9	2	0.9763	0.0101
0	0.8929	22	16	9	6	0.9763	0.0179
0	0.8900	5	3	3	3	0.9762	0.0112

IC17 and IC18 possible copies.

IC9 and IC10 are BC5 and BC6 from same cross.

It has been agreed to propose to the ISF Vegetable and Ornamental Section a threshold of 0.96 Jaccard similarity for the Butterhead group. No proposal was made for the Salinas Iceberg, as the inter-varietal variability is too small at the moment, the upper tail principle giving a possible threshold of 0.986.

It is interesting to note that the pedigree of some of the closest pairs was voluntarily given by the members of the working group. In none of the cases the varieties were essentially derived, but sister lines coming from a same starting cross and then selected separately at F3 or F4 level.