A COMPARATIVE STUDY OF MORPHOLOGICAL AND BIOCHEMICAL TRAITS IN THE GENUS BRASSICA

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Introduction

The adaptation of the numerous Brassica species to wide ecogeographical regions of the world together with human selection for desirable traits has led to their differentiation into large number of polymorphic forms. The present study reports findings from a comparative evaluation of numerous morphological traits, seed soluble proteins and certain seed isoenzymes in number of different taxa of Brassica including natural occurring and synthesized amphidiploids and CMS forms.

Materials and methods

The material comprised of a set of 11 taxa of Brassica representing 30 lines from diverse eco-geographical regions of the world, including ten artificially synthesized B.juncea involving various A-genomic parents and two cytoplasmic male sterile lines of B.juncea in different nucleus backgrounds.

For carrying out the morphological investigations, the material was sown in a complete randomised block design with three replications in the winter of 1985. Data on twenty significantly varying characters was subjected to the multivariate statistical analysis to obtain Mahalanobis Generalised Distance /D^2/ and Canonical variates /Z-values/.

The investigations on biochemical traits included quantitative estimations of seed oil, sugar contents and seed

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proteins and enzymes. The seed soluble proteins were extracted by the modified method of Yadava et al. /1979/ and were subjected to vertical PAGE according to the modified method of Davis /1964/. Isoenzymatic analysis of esterase-B /E.C. 3.1.1-/ was subsequently carried out in accordance with the method of Shaw and Prasad /1970/. Esterase-B enzyme was chosen, since it is known to display extremely low organ specificity and would ease the assay procedure. However, due to a practical limitation of sample size for such studies, only one or few representatives from each taxa were selected and studied. Band relative fronts /Rf's/ were calculated based on an average of 5-7 runs.

Results and discussion

Based on D^2 values, 30 lines were grouped in 15 distinct clusters /Table 1/. The D^2-values between lines ranged from as low as 38.71 /D^2_{3-4}/ to as high as 7793.99 /D^2_{2-28}/. Genetic distance was found to be maximum /83.32/ between clusters I and XIII and minimum /20.61/ between clusters VI and X, thus suggesting wide diversity among the chosen lines included in the study.

The other classificatory approach based on morphological characters was that of Canonical roots in which vectors or Canonical roots were calculated to represent the chosen lines in the graphical form /Rao, 1952/. The amount of variation absorbed by the first two roots was 81.93%, therefore, a two-dimensional representation of the relative positions was attempted /fig.1/. A tentative grouping of the various lines based on the data from Tocher's classification was also carried out on the Z_1-Z_2 graph /fig.1/.

The morphological study based on both D^2 and Canonical analysis revealed wide diversity in the material. The clusters were, in general, in accordance with the accepted pattern of grouping. All Canadian cultivars /Candle, Ante and Torkel/ clustered into one group. The same was observed for the leafy forms of B.campestris, the two local B.nigra

accessions and most of the artificially synthesized B.juncea. Amphidiploids with both leafy as well as oleiferous A-genomic parents grouped into one cluster possibly due to the monomorphic B-genomic parent. However, the amphidiploid YN-2 fell into the same group as its B.campestris parent. In addition, distinction among the three B.campestris ssp. oleifera ecotypes viz. yellow sarson, brown sarson and toria could also be made. The wide divergence of the two male steriles and B.tournefortii from the rest confirmed the role of cytoplasm and wild ancestors respectively in origin of genetic diversity.

At the biochemical level, seed soluble protein electrophoresis was used to detect differences at the primary gene product level. Some bands were found to be present in almost all the Brassica species while others occured rarely. B.carinata showed as low as only 6 bands, while B.juncea displayed 17. These n=18 lines were remarkably homogeneous /as also observed in morphological study/ and had high "similarity index" of 86.69% with the artificially synthesized lines. Similar trend was also indicated while comparing the natural elemental and digenomic species. A quantitative difference of 7 bands was recorded for B.campestris, which was highly divergent from the n=10 wild species B.tournefortii /s.i.=65%/. Similar was the case with the other wild species B.fruticulosa /n=8/ which showed a difference of 7 bands with B.nigra /n=8/. Unique bands were also found in B.oleracea var. alboglabra /n=9/.

The different species of Brassica were also analysed for isoenzymes of Esterase-B also called Carboxylesterase /fig.2/. Although 9 isoenzymatic forms could be recorded, much variation was observed between and within the species at both quantitative as well as qualitative levels. Bands 2 and 4 were most prevalent while 1 and 7 were present only in a few cases. Interestingly, the exotic B.juncea lines exhibited many isoenzymatic forms. Two amphidiploids YN-2 and JN had the same banding pattern. The monogenic species had predominance of bands 2 and 4. The polymorphic

n=10 species in general displayed a large variation. In its comparison, the wild n=10 species had no band in common.

On carrying out overall comparisons of the four methods viz., D^2, Canonical, Seed protein PAGE and Esterase-B isoenzymatic analysis a few commonalities could be observed. For example, lines even from diverse geographic backgrounds showed remarkable homogeneity. Polymorphic n=10 species also revealed wide diversity in protein and isoenzymatic profiles. The wild species showed appreciable divergence from the rest of the taxa studied. Finally, different statistical classificatory procedures resulted in similar grouping of the various taxa of Brassica as was reported previously by Denford and Vaughan /1977/ based solely on isoenzymatic studies. Thus one statistical approach using the morphological characters in this particular investigation revealed a close relationship with that obtained from biochemical traits and, therefore, highlights the importance and use of this classificatory procedures.

References

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Table 1 Clustering pattern of 30 representative lines of *Brassica*

Cluster number	Number of treatments	Treatments
I	3	(1)*Candle, (3)Ante, (4)Torkel (*B. juncea*)
II	2	(2)*parachinensis*, (8)*toria* D-3 (*B. campestris*)
III	1	(5) Yellow *sarson* cv. DYS-1
IV	1	(6)brown *sarson* cv. Pusa kalyani
V	1	(7) brown *sarson* cv. DBS-1
VI	2	(9)DYS-3 (yellow sarson), (17)YN-2 (yellow *sarson* x *nigra*)
VII	4	(10)*narinosa*, (11)*chinensis*-1, (12)*japonica*, (13)*chinensis*-2 (*B. campestris*)
VIII	1	(14) *B. tournefortii*
IX	2	(15)*nigra*-1, (16) *nigra*-2
X	8	(18)BN-1 (brown *sarson* x *nigra*), (19)TN-1 (*toria* x *nigra*), (21)NN-2 (*narinosa* x *nigra*), (22)PN-BPL (*pekinensis* x *nigra*) broad petiolated leaf), (23) JN(*japonica* x *nigra*), (24)CN (*chinensis* x *nigra*), (25)RN-BL68(*rapifera* x *nigra*; broad leaf), (26)RN-2.
XI	1	(20) RN-1
XII	1	(27)MS(PB) (CMS *B. juncea* cv. Pusa bold)
XIII	1	(28)MS (Var) (CMS *B. juncea* cv. varuna)
XIV	1	(29) EBJ 408 (*B. juncea* from Tibet)
XV	1	(30) EBJ 2611 (*B. juncea* from Nepal)

* Bracketed numbers as shown in fig.1.

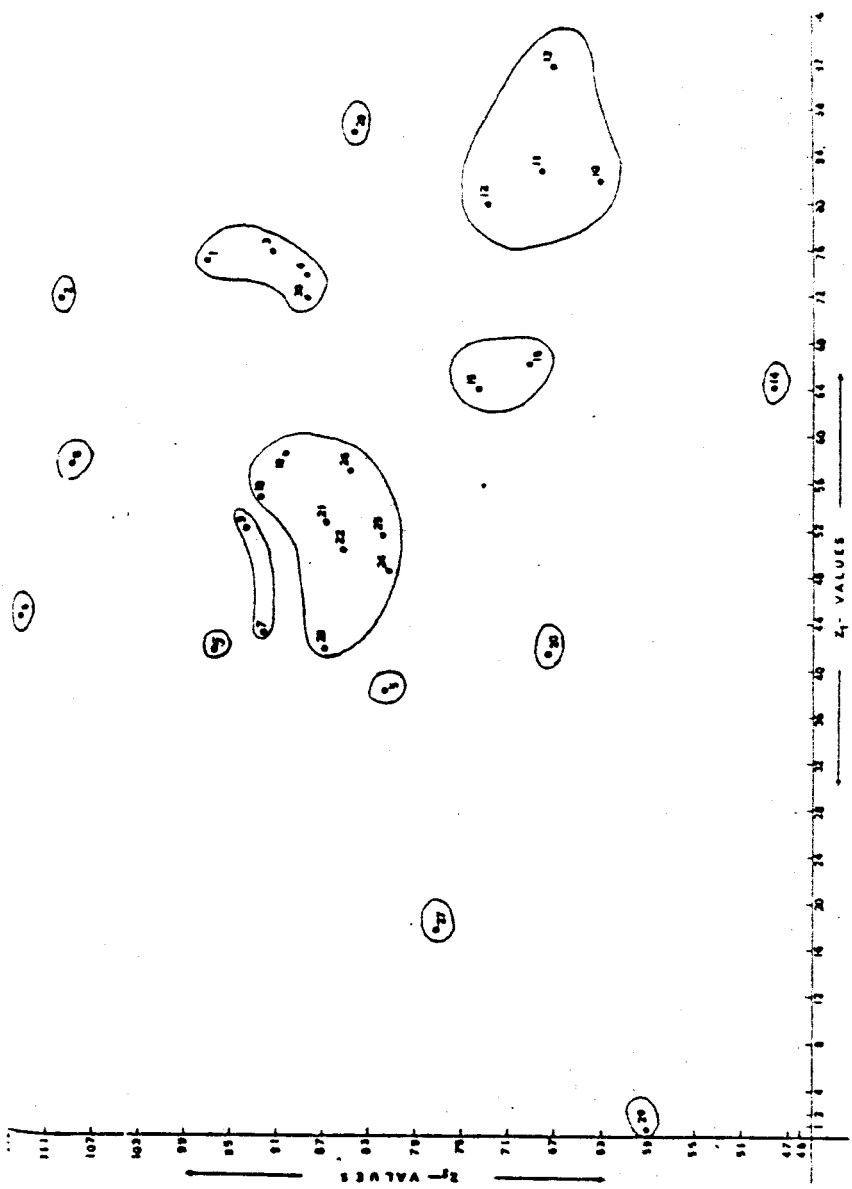

Fig. 1 GRAPH OF FIRST TWO CANONICAL ROOTS (Z_1 & Z_2) OF Biggsia SPECIES.

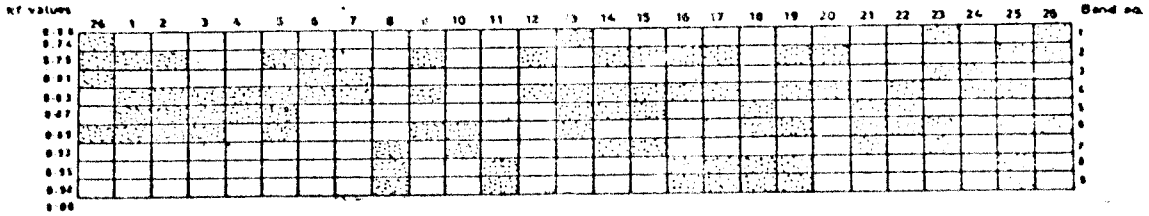

FIG.2 ZYMOGRAM FOR THE COMPARISON OF SEED ESTERASE-B ISOENZYMES FOR VARIOUS BRASSICA LINES

(1) Candle, (2) Span, (3) chinensis, (4) parachinensis, (5) pekinensis, (6) Dichotoma(DBS-1), (7) Trilocularis(DYS-1), (8) Torkel, (9) nipposinica-1, (10) Dichotoma(toria D-3), (11) narinosa, (12) japonica, (13) tournefortii, (14) nigra-1, (15) nigra-2, (16) YN-2, (17) JN, (18) EBJ 408, (19) EBJ 2503, (20) EBJ 2611, (21) Pol-1, (22) JR-7-2691-7, (23) nipposnica-2, (24) Albo-441, (25) EBJ 2469, (26) MS (Bj).