'HAKURAN' .. An Artificially Synthesized Heading *Brassica napus* L.

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The fact that the ***Brassica napus*** L. is an allopolyploid of two primary ***Brassica*** species, ***B. oleracea*** L. and ***B. campestris*** L. which have 9 and 10 chromosomes, respectively, was proved by late Dr. N. U., one of the founders of the breeding system F_1 hybrid of crucifers using self-incompatibility in Japan. The spontaneous ***Brassica napus*** L. has 19 haploid chromosomes and belongs to so-called secondary species of ***Brassica*** along with the other two species of ***B.carinata*** Braun and ***B.juncea*** Hemsl. which have 17 and 18 chromosomes, respectively. This species is supplying many important oil seed or fodder crops as rape and rutabaga in various places of the world. Though the ***B. oleracea*** L. and ***B. campestris*** L., the component species of ***B. napus*** L., include cabbage and Chinese cabbage, respectively, both have edible heads when matured, there are no indications that ***B. napus*** L. hold some heading crops as for as we know. So, many of research workers in the field of ***Brassica*** breeding tried to synthesize the artificial ***B. napus*** L. from said two primary species in aiming not only to get more informations about breeding and/or genetics on the ***Brassica*** species but also to synthsize artificial heading new ***B. napus*** L.

Until 1960, there reported 7 to 8 records of artificial hybrids between ***B. oleracea*** L. and ***B. campestris*** L. in the world, however, all of them were obtained from the combinations where the latter was used as female parent. Nobody succeeded in obtaining hybrids if the former was used as the female parent. These hybrids were proved to have 19 diploid chromosomes and no one could maintain their progeny except 'CO' which was produced by Dr. U. Mizushima, Prof. Emeritus of Tohoku Univ., in 1943 and brought to have 38 diploid chromosomes in latter generation. This crop was mainly used for fodder but not had any heading character.

At the Hort. Res. Sta. (Min. of Agr. and Forest.), Hiratsuka, Japan, the studies on the interspecific and intergeneric hybridization of ***Brassica*** relatives have been conducted since 1956 to obtain disease resistant Chinese cabbage varieties. Though their consumption is decreasing year by year for the past twenty years on account of the changes in people's diet, Chinese cabbage is still one of the main vegetable crops in Japan. The most troublesome problem in the Chinese cabbage production is the damages by Bacterial soft rot, a soil-born disease caused by ***Erwinia aroideae*** (Townsend) Holland. The 9 chromosomes ***Brassica***, cabbage family, is highly resistant to this disease. Then, the interspecific breeding project between cabbage and Chinese cabbage materials has been carried out aiming to give soft rot resistant character to Chinese cabbage. As mentioned above, this hybridization was extremely difficult by ordinary methods, but the application of 'embryo culture' technique gave unexpectedly good results in getting many hybrids and many of them had heading character in F_1, 19 diploid chromosomes, stage.

For example, the results in the early stages of this experiments are as follows. In 1958, in the case of crossing where Chinese cabbage were used as the female parent, only seven seeds were harvestd from 1,998 pods produced through 2,925 flower pollinations. These seven seeds were larger than

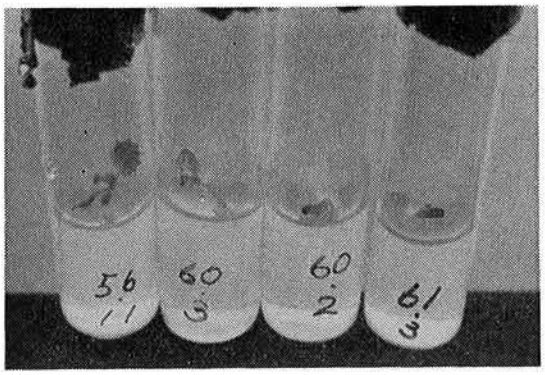

Fig. 1. Inoculated hybrid embryos
Left two : after 3-4 weeks of inoculation
Right two: after 1-2 weeks of inoculation
(just emerged and rooted)

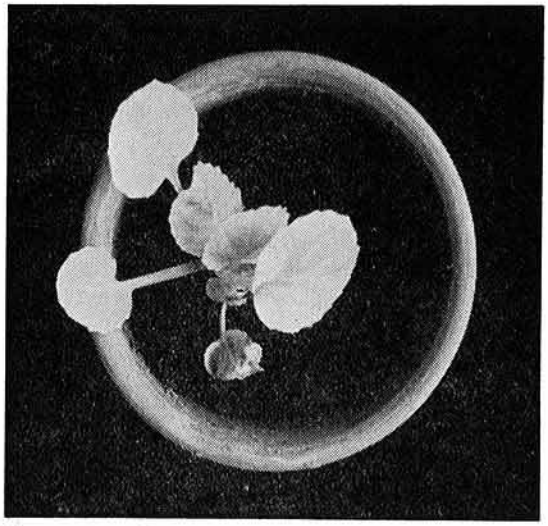

Fig. 2. Hybrid (F_1) seedling transplanted to the pot (about two months after the inoculation)

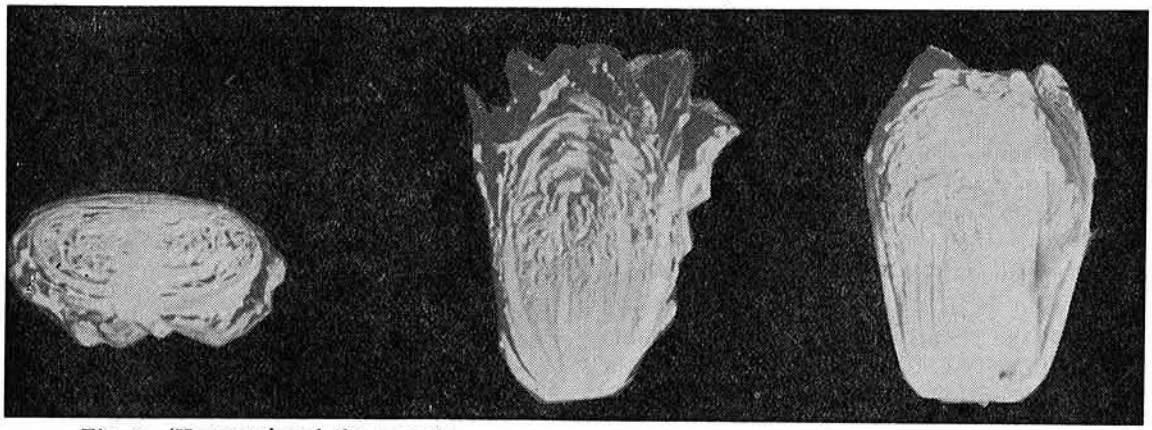

Fig. 3. 'HAKURAN' and the parents
From left to right: cabbage, HAKURAN and Chinese cabbage

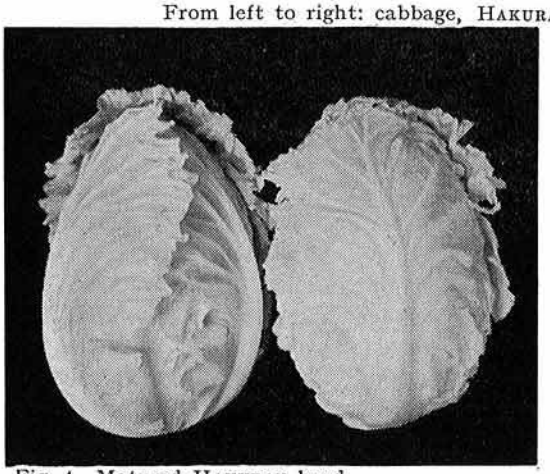

Fig. 4. Matured HAKURAN head

usual Chinese cabbage seeds and all produced matroclinous seedlings, however, all the other ovaries ceased their growth in early stages and their embryos degenerated so early that their artificial culture was regarded as impossible with our embryo culture techniques at that time. However, in the case of crossing where the cabbage was used as the female parent, 155 flowers were pollinated and eight young embryos were removed 40 days after pollination. These embryos were cultured in a culture media modified from White's basic solution and three hybrid seed-

lings were grown successfully. In the other case, where cabbage was used as the female parent, 46 hybrid embryos were removed 30 days after crossing where 250 flowers were pollinated. Twenty two hybrid seedlings were reared from these embryos. However, from the pods pollinated and left on the female cabbage plants six large seeds were harvested, all of which produced matroclinous seedlings. In the following year 1956, where the cabbage varieties were used as female parent, 395 pods were produced from 533 pollintions and 30 embryos were removed about 30 days after pollinations. These embryos were grown in the culture media and 5 hybrid seedlings were obtained. In the case where no embryo culture were carried out, 1,273 flowers were pollinated and no seeds were harvested from 637 pods.

The appropriate stage of removal and the appropriate ingredients of culture media should be determined to obtain better results, but the embryo culture technique was obviously so useful as a means of the progress of the studies on the interspecific hybridization of cabbage and Chinese cabbage. It is very interesting that in the early stage of the experiments hybrids were only obtained from the combinations where cabbage was used as female parent, never seen in the past trials where the ordinary pollination methods were used. On the contrary, no hybrids were raised when the Chinese cabbage was used as female parent, by which all of the past hybrids were obtained. After several trials, accompanied by progress in experimental techniques, both types of hybrids became available when appropriate parental varieties or strains were used for the combinations. Table 1 shows the number of hybrid seedlings raised by embryo culture during the past eleven years since 1958.

These F_1 hybrids were almost completely sterile in both of self-pollination and cross-pollination with cabbage or Chinese cabbage. So, it seemed very difficult to make back-crossing with Chinese cabbage materials, however, a certain amounts hybrid seeds were obtained through back-crossing every

Table 1. Number of hybrid seedlings raised by embryo culture

Year	Combination	
	a×c *	c×a *
1958	—	30
59	—	32
60	—	54
61	—	73
62	—	—
63	23	—
64	22	—
65	—	—
66	98	50
67	78	70
68	50	120
Total	271	429

* Genome; a ~ Chinese cabbage
c ~ cabbage

experimental year. Disease resistant breeding program is still under way.

While, these hybrid seedlings gave us tremendous amounts of breeding materials for heading new *B. napus* L.. F_1 hybrids were treated with colchicine and became artificial *B. napus* L. having 38 diploid chromosomes, and most of them showed highly heading character. These synthesized *B. napus* L. mostly showed high self-incompatibility while the spontaneous *B. napus* L. proved to be highly self-compatible when open-flower pollinated. This is an interesting phenomenon from the viewpoint of the inheritance of compatibility in cruciferous vegetable crops.

Further improvement and seed growing of synthetic *B. napus* L. obtained in early experiments has been achieved in some Prefectural Exp. Sta. and Seed Co., and heads are being shipped to the markets under the name 'HAKURAN' (Chinese cabbage and cabbage are called in Japanese as 'HAKUsai' and 'kanRAN', respetively) in the western Japan since 1966. It taste like heading lettuce with juicy, soft and less fibrous character of leaves and is good for pickles and for fresh uses. From the cultural viewpoint, HAKURAN is highly resistant to soft rot and other bacterial diseases and is very easy grow because of their highly

resistant to drought and heat. Present time, their cultivation is limited in certain area of Japan because of lack in ecotypes. However, in future, they will expand to several area of this country when more HAKURAN varieties were raised through breeding project.

In experiments for the past two-three years, it became rather difficult to obtain F_1 embryos for culture when particular strains of cabbage and/or Chinese cabbage were used for hybridization. As the results of pollination test, these parental strains mostly contributed from seed campanies or Pref. Exp. Sta. and supposed be the parents for F_1 breedings, showed a rather high rate of self-incompatibility. These facts might suggest that the self-incompatibility of parental strains may influence the production of F_1 embryos and their further growth, and also that some parts of self-incompatible genes of both species are common to each.

'HAKURAN', a new vegetable crop, will also be useful as the material for disease resistant breeding of Chinese cabbage and for the study of inheritance of several characters including heading, both projects are now under research at several places in Japan.