

**Varietal improvement of kailaan and
production of a kailaan-broccoli hybrid -
Stage 2**

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Henderson Seed Group Pty Ltd

Project Number: VG01092

VG01092

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This final report describes the breeding and selection of a kailaan - broccoli (Chinese kale – broccoli) hybrid

This project is funded by HAL and Henderson Seed Group Pty Ltd.

Date: 30th September 2006



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Media Summary

This Kailaan * Broccoli (Kailaan – Broccoli Hybrid) breeding project was funded by HAL and Henderson Seed Group Pty Ltd. The aim of this project was to incorporate the superior high yielding quality of broccoli into the leafy low yielding Chinese kale or kailaan. Kailaan is a leafy vegetable with a small frame popular in tropical and sub-tropical Asia. Kailaan also has a nutty flavor well liked by the Asian consumers. The leaves as well as the stems are consumed usually stir fried.

The vegetable growers will benefit directly from this Kailaan * Broccoli breeding program with the introduction of an alternative variety to Broccolini. Henderson Seed Pty Ltd and Australia will benefit from export of this hybrid.

The key outcomes from this project will be a hybrid Kailaan * Broccoli variety with superior yield and the desirable nutty flavor.

The project has concluded with the advance stage in the development of broccoli and kailaan parent lines that will be used to produce the Kailaan * Broccoli hybrid variety.

Future R&D will focus on completing the breeding of the parent lines of the Kailaan * Broccoli hybrid and packaging production agronomy for on farm Kailaan * Broccoli production.

Superior commercial hybrids will be extended to the farming community by Henderson Seed Group Pty Ltd and its agents throughout the vegetable production areas of both Australia and New Zealand. Hybrid seed of the Kailaan * Broccoli variety will also be exported by Henderson Seed Pty Ltd.

Technical Summary

Australian leafy vegetable producers have recently been introduced to a new vegetable called Broccolini. Broccolini is marketed as a hybrid between Chinese kale or kailaan, *Brassica oleracea albogabra* and broccoli, *Brassica oleracea italica*. The main economic product of Broccolini is the axillary floral shoot. Broccolini is the only hybrid between Chinese kale and broccoli that is being marketed world wide. Henderson Seed Pty Ltd with funding from HAL initiated a breeding program to produce an alternative variety of kailaan – broccoli hybrid. The hybrid is called Kailaan * Broccoli and will be trade marked as such by Henderson Seed Pty. Ltd.

The objective of this breeding program is to introgress the superior yield of broccoli into the low yielding leafy kailaan. The resultant Kailaan * Broccoli hybrid possesses much hybrid vigor. The F1 hybrid approximates the size of the broccoli plant making the plant size of the hybrid three to four folds that of the leafy kailaan variety. The economic yield of resultant intervarietal hybrid, Kailaan * Broccoli, are the tender axillary floral shoots suitable for both Eastern stir fry or served in Western meals not unlike the Asparagus sprouts.

Inbred lines of broccoli and kailaan developed from this program had enabled the production of some experimental F1 hybrids. On Station trial results of these new experimental hybrid varieties were very encouraging. The parents of the short listed hybrids are under going conversion into the cytoplasmic male sterile form. The Kailaan * Broccoli hybrid to be released in 2010 will be male sterile.

Introduction:

Kailaan or Chinese kale, *Brassica oleracea albogabra*, is a leafy green vegetable well liked by Asian consumers for its nutty flavour. Kailaan is deficient in yield and has soft leaves and small stem diameter with poor storage qualities. Chinese kale as described in Plants for A Future is as follows:

Family: Cruciferae

Genus: *Brassica*

Synonyms: *Brassica oleracea albogabra* – L.H. Bailey Musil

Source: (www.pfaf.org/database/plants.php?Brassica+oleracea+albogabra)

Broccolini is a new vegetable released in the late 1990's by Sakata Seed Company of Japan. Sakata Seed Company described it as a hybrid between broccoli and Chinese kale. The scientific classification of Broccolini which is a patented name by Sakata as defined by Wikipedia (2006) is as follows:

Kingdom: Plantae

Division: Magnoliophyta

Class: Magnoliopsida

Order: Brassicales

Family: Brassicaceae

Genus: *Brassica*

Species: *B. oleracea*

Cultivar Group: *Brassica oleracea italica* * *albogabra*

Perfection Fresh the Broccolini producer in Australia described it as: Broccolini™ baby broccoli was introduced to Australia by Perfection Fresh in 1999 – a cross between broccoli and Chinese kale.

Source: www.perfection.com.au/01_cms/details.asp?ID=145

Broccoli, cauliflower, cabbage and Chinese kale all belongs to the same species, *Brassica oleracea*. All four groups of vegetables are cross compatible. However, these vegetables are all different enough to be classified under different sub species. Broccoli is *Brassica oleracea italica* while Chinese kale is *Brassica oleracea albogabra*.

The botanical classification of broccoli on the other hand as reviewed by McMurray (1999) in his Ethno Botanical Leaflets in the web link:

Source: www.siu.edu/~ebl/leaflets/broccoli.htm is

Family: Brassicaceae

Genus: *Brassica*

Species: *B. oleracea* var, *italica* or sprouting broccoli

In its web link www.oregonstate.edu/Dept/hort/233/brassicaceae.htm, the Oregon State University website puts the two vegetables under the following classification:

Species	Botanical Variety	Chromosome Number (N)	Genome (haploid)	Common Name
<i>B.oleracea</i>	<i>italica</i>	9	C	broccoli
<i>B.oleracea</i>	<i>albogabra</i>	9	C	Chinese kale, kai laan

Source: www.oregonstate.edu/Dept/hort/233/brassicaceae.htm

It can be seen that broccoli and kailaan share a common C genome and has diploid chromosome number of 18. These similarities allow these two vegetables to be classed in the same species i.e. *Brassica oleracea* but different subspecies. These two subspecies are easily cross compatible forming fertile F1 plants. The cross compatibility of these two species are utilized in the breeding of kailaan * broccoli hybrid in this HAL and Henderson Seed Pty Ltd funded project. Henderson Seed Pty Ltd has named this F1 hybrid - Kailaan * Broccoli.

The Kailaan * Broccoli:

Kailaan * Broccoli is a hybrid derived from crossing broccoli with kailaan. It has the desired traits of both kailaan and broccoli. The yield potential of Kailaan * Broccoli is higher than kailaan because of hybrid vigour and the marketable flower shoot of Kailaan * Broccoli has the nutty flavour of kailaan.

The Kailaan * Broccoli plant exhibits many features of the sprouting broccoli or calabrese plants. Kailaan * Broccoli has white flowers and large floral buds or beads, features shared by some calabrese varieties. Sprouting broccoli varieties or calabrese varieties are land races of broccoli planted by Italian farmers before the introduction of modern single stem large headed broccoli varieties. Sprouting broccoli belongs to the same species as modern broccoli. Figure 1 and 2 show the kailaan and Kailaan * Broccoli plants respectively. The Kailaan * Broccoli plant is as tall as or even taller in plant frame than the broccoli plants. The Kailaan * Broccoli plant exceeds the kailaan plant frame and stem thickness by two or three folds. The Kailaan * Broccoli plant produces a loose small flat head very similar to that produced by the sprouting broccoli or calabrese plant. The Kailaan * Broccoli plant has a strong tendency to produce side shoots which develop into branching inflorescence (Figure 3). These young side shoots bearing young inflorescence are the economic product (Figure 4). Side shoot production is induced by topping the young terminal inflorescence of the Kailaan * Broccoli. The economic product as shown in Figure 4 has a slender, fleshy stem, small leaves and a small head with flower (buds) beads larger than the broccoli parent lines. The flower buds (beads) of the Kailaan * Broccoli are larger than those of the broccoli parent line because the kailaan parent lines have large flower beads. The flower of the Kailaan * Broccoli is white in color similar to that of the kailaan parent line in contrast to the yellow color of the broccoli parent line. This is because white petal color is dominant over yellow petal color in *Brassica oleracea*.

Comparing the phenotype of Broccolini and Kailaan * Broccoli brings into question if Broccolini is truly a Chinese kale (kailaan) by broccoli hybrid. The phenotype of Broccolini from the leaf shape to the very fine bead size suggests more similarity to the calabrese or sprouting broccoli than Chinese kale by broccoli hybrid. Moreover the flower of Broccolini is yellow. As almost all kailaan varieties have white flower, Broccolini if it is a Chinese kale by broccoli hybrid, must have been made with a yellow flower Chinese kale parent line.

The botanical classification of Kailaan * Broccoli is as follows:

Kingdom: Plantae
Division: Magnoliophyta
Class: Magnoliopsida
Order: Brassicales
Family: Brassicaceae
Genus: *Brassica*
Species: *B. oleracea*
Cultivar Group: *Brassica oleracea italica* * *albogabra*

Materials and Methods:

Breeding F1 Kailaan *Broccoli:

The breeding of Kailaan * Broccoli F1 hybrid involves first the development of inbred parent lines of both kailaan and broccoli. Henderson Seed Pty Ltd has an active broccoli breeding program and has developed numerous broccoli parent lines which can be used for making Kailaan * Broccoli hybrid. On the other hand no kailaan inbred parent is available and kailaan inbred parents have to be developed. The HAL funding has enabled Henderson Seed Pty Ltd to initiate the kailaan inbred parent line development.

Development of Inbred Broccoli Lines

Selection Criteria:

The following selection criteria are commonly applied by broccoli breeders when they breed their inbred lines. The broccoli head must have fine flower buds commonly referred to as beads. The beads must be uniform. Variable bead sizes are not desirable as the larger beads tend to open up. Beads that are open make the broccoli head unattractive on the supermarket shelf. Another important trait not desired is brown beads. Brown beads are caused by loss of chlorophyll due to death of the flower bud. There are some inbred lines that consistently breed brown beaded F1 hybrids suggesting genetic control for this trait.

The broccoli head must have a dark green color. The color of the broccoli head is enhanced by cold weather and some varieties of broccoli can assume a purplish green color during the cold winter months. These varieties have high anthocyanin in their floral heads. Some producers prefer a normal green instead of purple green head. However, there are some varieties that are light green in color and during warmer season the broccoli head can become very pale green. Varieties that are too pale green are not preferred.

Another feature of the broccoli head that is not desired is the presence of ‘cats eyes’ or ‘starring’ in the beads. As the term suggests ‘starring’ is star formation in the broccoli head caused by fine floret buds surrounded by bigger floret buds. Starring is usually caused by unseasonal fluctuating temperature leading to differential growth in the buds.

An important trait in the breeding of Kailaan * Broccoli is the selection of broccoli parent lines that are able to produce side shoots. Prolific side shooting will enable the Kailaan * Broccoli hybrid to produce more harvestable shoots.

Other than the traits describe above for the broccoli head, the broccoli plant must be vigorous and have a large frame and be tolerant to the major broccoli diseases.

Sources of Genetic Variations for a Broccoli Breeding Program

Land races, open pollinated varieties and F1 hybrids are the main sources of genetic variation for breeding improvement of broccoli varieties. Land races and open pollinated varieties do not as a rule have all the traits for a well domed, heavy and fine beaded broccoli head. Most of the yield traits of a well domed and heavy dense broccoli are derived from F1 commercial hybrids. The commercial hybrids that are available form the bulk of the genetic materials for initiating a broccoli breeding program. On the other hand land races are ideal because of its natural sprouting ability for use as the broccoli parent line in this Kailaan * Broccoli breeding project.

Plant Selection.

A sizable number of broccoli varieties must be planted out to initiate a plant selection program. If open pollinated varieties are used 100 to 200 plants per land race or open pollinated variety need to be planted. At heading time, plants producing superior quality heads are selected from the land races or the open pollinated varieties. Depending on the variation within each of the land races and open pollinated varieties, one to a few plants is selected from each variety. For F1 hybrids, fewer plants need to be planted per plot. Usually 25 plants are planted for the F1 hybrid to be characterized but only one plant needs to be taken at random because all F1 plants are genotypically very similar. The plants selected are tagged individually with a paper tag. Each plant is identified with the year and the plot number of the variety. For example the tag with 04-105-1 is for the first plant selected from plot 105 planted in 2004. The entry for plot 105 is recorded in the pedigree book. Thus plot 105 may be a broccoli variety called Marathon. Thus 04-105-1 is the first plant selected from the F1 variety Marathon. The second plant selected from

this plot will be labeled as 04-105-2. A date attached to the tag will indicate the relative maturity of each plant selected. The selected plants are potted and the plants transferred to a plant house. Plants are healthier if maintained in a sheltered plant house as rain can cause the head to break down leading to more diseases.

Pedigree Record

The pedigree of each selected plant must be recorded carefully. As explained above the selected plant is identified thus: the first 2 numerical digits represents the year the selection is carried out. This is followed by the name of the variety. The last entry in the pedigree identity code is the plant number. The first plant is allocated 1 with the next plant being allocated 2 and so on and so forth.

The selected plants are then selfed pollinated and the seed collected planted out as a family to a row. Thus the seed collected from the plant 04-105-1 are planted to a family row the following year. Assume the plot number for this family row planted in 2005 is 05-208. This is the F2 family of the variety Marathon. Plants are selected within this F2 family. Each plant selected is again allocated a number. The first plant selected is allocated the family number which is 05-208-1. The second plant selected has an extension of -2. Thus the third plant selected from this F2 family will be 05-208-3. Here the code 05 represents the year and 208 is the plot this F2 Marathon family was planted on.

This pedigree number is extended by one digit for each generation advanced. The first plant selected from the F3 family derived from selfing plant 05-208-3 will be Marathon - 1-3-1. The selection continues until the line has reached F7 or F8 generation. When such advance inbreeding has been reached, the inbred line is homozygous enough for the line to be named. The line will be referred to by the name independent of the generation of inbreeding from now on. We may for example call line Marathon-1-3-1-2-1-3-1 by the line name ED-P1.

Preparing the Plant for Pollination

Pollination can be carried out on the flowers from the main stem or from the side shoots. Some plants do not have any side shoots and therefore the main broccoli head has to be used for pollination. The main central head can be removed if the broccoli plant has side shoots. The flower heads from the side shoots are smaller and are less susceptible to head rot. It is preferable to use the side shoots for pollination. The broccoli head has many florets. To prevent disease especially head rot, most of the florets are removed by cutting with a pair of scissors. The top one third of the broccoli head can be removed initially. This is followed by pruning off the side florets. Usually only three branches of florets are left. The three branches rapidly develop and more training is required to remove excessive florets. The florets are trained by removing side florets. Regular removal of side floret results in a three branch inflorescence. The inflorescence is ready for pollination when a dozen or so flowers have appeared in each branch of the

inflorescence. The mature flowers are sources of pollen for the pollination. The older shriveled flowers are removed leaving behind the fresh flowers.

The younger smaller buds in the inflorescence are removed by cutting the tip of the inflorescence off. The dozen to 20 prime larger buds and the mature flowers are left in each branch of the inflorescence. All three branches in the inflorescence are similarly prepared ready for bud pollination.

The tip of each flower bud, usually the top one fifth of the bud, is removed using a pair of forceps. This is done by cutting the tip of the bud just beneath the sepal and petal layers of the bud. The stamens and anthers can be removed together with the petal and sepal but care must be taken not to injure the style or stigma. Any abrasion can cause the style to break or the injury can result in fungal or bacteria infection on the wound. The infection can cause the ovary to rot off. A well prepared bud exposes the stigma surface. A neat pollinator usually trims off only sufficient petal and sepal tissue to barely expose the stigma surface. The exposed stigma surface allows the pollinator to pollinate the bud. The bulk of the sepal and petal tissues forming the jacket around the ovary are still intact.

Pollination

Self bud pollination is carried out by gently rubbing a ripe anther on the stigma surface. Care must be taken to use the surface of the anther bearing the pollen in the pollination. This is the surface facing the stigma. A fine layer of yellow pollen can be seen on the surface of the stigma after a couple of light touches with the mature anther. If no yellow layer of pollen is visible, the pollen load is inadequate for good seed set.

Self Incompatibility Test

During the bud pollination process, a self incompatibility (SI) test is carried out. This test allows the plant breeder to determine the SI status of the plant. The plant can have a very strong SI or it may be completely self compatible. The SI test is carried out by self pollinating the buds and the open flowers in the same inflorescence branch. A wire ring is used to demarcate the open flowers from the buds. The buds above the ring and the flowers below the ring are pollinated using pollen from the same plant. After all the buds and flowers in the plants are pollinated, the inflorescence is bagged usually with a transparent porous bag to prevent any undesirable cross pollination by stray insects.

Depending on the growing temperature, the bags are removed 7 to 20 days after pollination. At seed harvest which is usually 90 days after pollination, a SI reading is made. The breeder reads the seed formed in each pod starting from the bottom of the inflorescence. Each pod in the inflorescence is examined. A full pod is assigned a +, a pod with a few seed assigned \pm and an empty pod assigned a -. A typical SI reading for a

very strong SI which is assigned an SI rating of 5 (the SI rating scale of 1 to 5; 1 for very weak or self compatible to 5 for very strong SI or highly self incompatible) is
- - - - - / - - ± + + + + + . Reading is from the bottom of the inflorescence to the top. In this case the nine – represents nine pods without seed. The / is the demarcation ring between the flowers and buds. The three – after the ring represents the three mature buds that did not set any seed despite being bud pollinated. The ± after the – represents a few seed set in this pod. This suggests a very strong SI as even the more mature buds are unable to set or set very few seed. The next nine buds set good seed.

Broccoli Seed

Broccoli seed are ready for harvest when the pods turn yellow to brown, although some pods could be picked even when green. Green pods with yellow to brown seed have reached maturity. Harvested pods are placed together with the plant tag identifying its pedigree into a brown paper bag. Seed pods from each plant are placed in a bag. The seed pods are either sun dried or forced dried with a warm air of around 40 ° C. Seed are considered sufficiently dried at a moisture content of around 7%. When sufficiently dried the seed pods or siliqua split readily to release the seed. The seed are threshed and removed from its pods. Seed from each plant are placed in a little seed packet. Again the tag with the pedigree goes with the clean seed into the seed pocket. The pedigree can be written on the seed packet and seed are filed according to their ascending pedigree number. This system of filing allows easy tracking of any lines in time of need.

The seed are used for the next cycle or generation of plant selection.

Development of Inbred Lines.

The seed collected are sown for another cycle of selection and self pollination to continue the inbreeding process and at each generation, further selection of desired plants is carried out. After six generations of inbreeding the lines are sufficiently homozygous and therefore phenotypically uniform for the lines to be evaluated for their potential as parent lines. Parent lines are classified as potential male and female lines based on their SI status. Plants with very strong SI of 5 are classed as female lines as these lines do not set their own seed readily. The other lines with weaker SI of 4 or lower can be used as the male lines in the different hybrid combinations.

Specific Combining Ability

The female inbred lines with strong SI can now be tested crossed with all the male lines. Top crosses are made using all the available male lines on each of the female lines. Top crossing is achieved by crossing 20 mature flowers of the female lines with the pollen of each of the donor male line. The female flowers used are the fresh prime flowers in each of the inflorescence. Older more shriveled flowers are first removed. The buds and smaller immature flowers are also removed to leave only the prime flowers. The use of

the prime flowers and the removal of the smaller younger flowers allow a better seed set as all the energy can be channeled to the pollinated flowers. Cross pollination is achieved by gently rubbing the anther of the male line against the stigma surfaces of the female flowers. The donor anther used for pollination is held with a pair of fine forceps. The anthers in the flowers of female parents need not be removed as the strong SI female should not be receptive to its own pollen. After each of the flower has been pollinated, a tag with the cross combination is written and tied around the pollinated flowers. An example of a label for a new cross is with female line A and male line B is A*B followed by the initial of the pollinator and a date when the pollination was carried out. Care must be taken to clearly tag each cross so that at harvest each cross can be harvested separately.

The seed from each cross combinations is grown in a competitive trial using the best commercial varieties as check varieties. Cross combinations that perform as well or better than the check varieties are noted. More hand crossed seed of these combinations that perform well are made in the following season. A few hundred seed are made for replicated trials across different locations to verify the superiority of these new hybrid combinations. A further short list is made from the results of these trials. Sufficient seed for multi-location trials can be made using cage or tunnel isolations. This method of F1 seed production is described below in the section under cauliflower breeding.

Male Sterility and Their Use in Hybrid Seed Production

The other system used in hybrid seed production makes use of a male sterile line as the obligate female line. There are two systems of achieving male sterility in plants. One is cytoplasmic male sterility and the other is genetic male sterility. In the cytoplasmic male sterility system, sterility factors that are carried in the mitochondria in the cytoplasm prevent anther formation. In some species such as onion, *Allium cepa*, the cytoplasmic male sterile factor acts in tandem with a single male sterile gene. The recessive alleles will bestow male sterility in a plant with a male sterile cytoplasm. However, if the dominant allele is present the dominant allele over rides the male sterility factor in the cytoplasm and the plant is fertile. A fertile cytoplasm will result in a fertile plant no matter what the genetic make up is. A plant with fertile cytoplasm and double recessive sterile alleles is used as a maintainer for the production of the male sterile line. This is achieved by crossing a male sterile line with male sterile cytoplasm and double recessive sterile gene to a plant with male fertile cytoplasm and double recessive sterile gene. However, in some plant genus such as the *Brassica* there is no gene interacting with the sterile cytoplasm. The sterile cytoplasm which had been transferred to this genus from *Raphanus* acts on its own to produce the male sterility. Thus the breeding of cytoplasmic male sterile lines in the *Brassica* genus is less complicated compared to onion.

The male sterile plant produces only atrophied anthers. The atrophied anthers do not have any pollen, thus the female plant can accept pollen freely from the male line to

make the F1 hybrid seed. Here again nature is not perfect. There are a small percentage of F1 hybrid plants produced by the cytoplasmic sterile female plant that is defective. These defective F1 plants are often referred to as aberrant plants and are widely reported in cauliflower hybrids made from cytoplasmic sterile parent. These aberrant plants are the results of abnormal gamete formation due to disturbance in chromosome movement during meiosis.

Producing CMS *Brassica* Hybrid Seed

Brassica species do not have a natural source of cytoplasmic male sterility in its populations. The cytoplasmic male sterility found in modern *Brassica* hybrids originated from the genus *Raphanus*, specifically Radish. Ogura transferred the sterile cytoplasm from radish to the *Brassica* species. Initially there was problem using the Ogura male sterile cytoplasm in temperate *Brassica*. The chloroplast found in the Ogura cytoplasm was not suited for temperate low temperature production. The leaves of the *Brassica* crops with the Ogura cytoplasm tend to be yellow under low temperature growth. Protoplast fusion has resulted in the replacement of the chloroplast with more adapted chloroplast and the improved Ogura cytoplasm is now widely used in temperate *Brassica* hybrid seed production. Another source of *Brassica* male sterile source is the Anand cytoplasm.

CMS female parent lines must be developed first before CMS *Brassica* hybrids can be produced. To initiate the CMS breeding, a source of CMS must be secured. Improved Ogura cytoplasmic sterile sources are controlled by private patents. A license must be granted before the cytoplasm can be used for commercial seed production. To initiate CMS breeding, any sterile plant carrying the cytoplasm of interest can be used. To commence conversion, the line to be converted usually a stable inbred line is crossed to the source of male sterile cytoplasm. The conversion of the inbred line to a CMS line can be achieved by backcrossing the line with the recurrent fertile inbred line. The converted line is ready for use at B/C 6 when almost the whole of the genome of the recurrent fertile line has been transferred to the CMS line. The percentage of transfer of the genome of the fertile line to the CMS line in the back cross program is as follow:

CMS Plant * Fertile Line =F1 (50%)

Backcross 1 = 75%; Backcross 2 = 87.5%, Backcross 3 =93.75; Backcross 4 = 96.875

Backcross 5 = 98.4375%; Backcross 6 = 99.12875%

At B/C 6 the CMS LINE has almost the same genotype as the recurrent parent line and the line has successfully been converted and is ready to be used as a CMS female line.

Developing Kailaan Lines:

As discussed above the other parent line for producing a Kailaan * Broccoli F1 hybrid variety is a Kailaan inbred line. Kailaan inbred lines are bred using the same method as that for developing broccoli inbred lines. Kailaan lines are developed mainly from open pollinated Kailaan varieties. Kailaan open pollinated varieties are planted out in the desired production environment. Single superior plants are selected from adapted varieties and self pollinated as in the broccoli inbreeding process. Six cycles of inbreeding of selected plants are carried out. Fertile stable inbred Kailaan lines so developed are used for test crosses with male sterile broccoli lines.

Figure 1: Inbred Kailaan Line



Test Cross Kailaan * broccoli Hybrids:

Hybrids can be made from self incompatible (SI) or cytoplasmic male sterile (CMS) inbred lines acting as parent line. The Kailaan * Broccoli hybrids were made by using Henderson Seed Pty Ltd's self incompatible (SI) broccoli inbred lines as the female parent.

The Kailaan lines generated from above are used as pollen or male lines to make experimental hybrids with the broccoli female lines. The hybrids so generated are tested for their productivity and agronomic traits. Desired traits in Kailaan * Broccoli are:

Plant Size

Maturity

Shoot Length

Shoot Productivity

Bead Size

Shape of the Flower Head

Stem tenderness i.e. low fibre in the stem

Tolerance to disease especially Downy Mildew

Superior yielding Kailaan * Broccoli hybrids with the desired traits listed above are short listed for further multi-location yield and adaptation trials.

Results:

A yield trial of an experimental Kailaan * Broccoli hybrid KBL 88 was carried at the Henderson Seed Pty Ltd Research Farm at Templestowe in Victoria in the Autumn/Spring season. The plants were transplanted in the paddock on 20th April 2006. The harvest was from early July to Mid September. A total of 11 harvests were made. Harvest was made on demand and varies from bi-weekly to weekly pick. A pick was made when there were sufficient shoots that need to be picked.

Figure 2: Kailaan * Broccoli Plant with Primary Head



Harvest Agronomy:

The Kailaan * Broccoli plant was planted to a spacing of 50 cm between plants and 75 cm between rows. Twenty plants planted to a single plot were used in this study. The plants were given the same cultural practice as in the production of commercial broccoli. Kailaan * Broccoli KBL 88 headed 12.5 weeks after transplant. The terminal head was removed soon after heading by cutting it off 3cm below the head. This allows the plant to start producing side shoots. The side floral shoots are the commercial Kailaan * Broccoli product. Kailaan * Broccoli shoot harvest commenced one week after the removal of the primary head. Floral shoots are picked with a view to induce more shoot proliferation. This is done by picking the shoot just above the axil of the leaves, allowing usually two axillary buds to develop into 2 floral shoots for future harvests. By carefully picking the shoots this way the Kailaan * Broccoli plants are groomed to produce a large number of floral shoots.

In this study each plant produced an average of 36.5 shoots over 11 harvests giving a total yield of 519 grams. The shoot length average 20.7 cm.

Figure 3: Kailaan * Broccoli Plant-Primary Head Removed



The shoot number at harvest showed a decline mid season. This is mainly due to the lower than usual rainfall received during July to early August. However, the shoot yield increased significantly with higher rainfall and irrigation and fertilizer applied mid august to September. The increased in shoot number is however, compensated by lower shoot weight. The shoot size has declined to a low of 10 g per shoot. The economics of harvesting shoots of less than 10 g is questionable. The last harvest with shoot weight of less than 7.5g per shoot was not included in this yield record. The tables below summarize the data collected in this trial for experimental, Kailaan * Broccoli variety KBL 88.

Figure 4: Kailaan * Broccoli Shoot – Harvested From Experimental Hybrid KBL 88



Yield of Kailaan * Broccoli KBL 88 at Templestowe Autumn/Spring 2006

Variety	KBL 88
Harvests	11
No of Plant	19
Average Shoot Weight (g)	16
Average Shoot Length (cm)	20.7
Yield Shoot Weight/Plant (g)	519
Yield Number of Shoots/Plant	36.5

Harvest Date	Shoot Nos	Weight (g)	Weight/Shoot (g)
11-Jul 2006	27	896	33.2
14-Jul 2006	80	2164	27.1
18-Jul 2006	39	1337	34.8
23-Jul 2006	18	295	16.4
28-Jul 2006	11	180	16.4
1-Aug 2006	7	149	21.3
8-Aug 2006	10	181	18.1
15-Aug 2006	45	698	15.5
21-Aug 2006	137	1657	12.1
28-Aug 2006	163	1733	10.6
5-Sep 2006	198	2360	11.9

Discussion:

An experimental self incompatible form of F1 hybrid of Kailaan * Broccoli called Kailaan * Broccoli - KBL 88 had been bred and was identified for further breeding to developed a male sterile form of KBL 88. This Kailaan * Broccoli hybrid produced floral shoot with an average length of 20.7 cm and weight ranging from 30 g to 10 g per shoot depending on the age of the plant at harvest. The first harvest on the younger plant produced a heavier shoot and the shoot weight declined as the picking season progressed. Average shoot weight across season was 16g per shoot and total shoot yield per plant was 519 g from a total of 36.5 shoots per plant. Shoot could be harvested 14 weeks after transplant and up to 10 harvests could be made over the 2 months harvest period. Weekly to twice weekly harvests can be made. Yield of shoot is dependent on availability of water and fertilizer input. Shoot production is very dependent on rainfall and or irrigation and fertilizer top dressing.

More work is required to complete the development of male sterile broccoli parent lines. The male sterile broccoli parent line will then be used to produce seed of the commercial F1 Kailaan * Broccoli hybrid –KBL 88.

Technology Transfer

Little technology transfer has taken place in this breeding project. Variety release is still two to three years away, it is therefore not desirable to publicize the new hybrid as the request for trial seed cannot be made. However, extension and research officers from Department of Primary Industries and Henderson Seeds' sales personnel are constantly being exposed to the research progress in this project. Extension and development work are planned when the first pre-commercial sample of male sterile hybrid seed are made in two years time.

Recommendations:

The experimental Kailaan * Broccoli hybrid made during the course of the HAL-Henderson Seed Pty Ltd funding period was based on the self incompatible (SI) system. The weakness of the SI female line is the frequent occurrence of the female line setting its own seed. The result is the failure to achieve 100 % hybridity in the seed crop. The presence of even a small percentage of the inbred female in the hybrid seed lot is not acceptable. The hybrid seed quality specification often does not allow more than 5% inbred seed level in a hybrid seed lot.

Another problem of the presence of inbred female parent seed in the hybrid seed lot is the danger of the loss of the inbred parent lines to competitor seed producers.

The cytoplasmic male sterility (CMS) system can be used to overcome the deficiencies above. With the CMS system, no self pollination can take place and therefore the hybrid seed lot produced can be free from presence of inbred parent seed. The use of CMS hybrid in Brassica seed is increasing at a rapid pace and Henderson Seed Pty Ltd is in an advance stage of developing CMS broccoli parents. It is recommended that the Kailaan * Broccoli F1 hybrid production program be based entirely on the CMS system. The change over from SI hybrid to CMS hybrid will delay the release of a Kailaan * Broccoli hybrid by three years as the development of CMS broccoli lines at Henderson Seed Pty Ltd is three years behind the SI lines.

References:

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