The biology and ecology of canola

(*Brassica napus*)

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PREAMBLE

This document addresses the biology and ecology of the species *Brassica napus* L. Included is the origin of *B. napus* as a crop plant (referred to as 'canola'), general descriptions of its growth and agronomy, its reproductive biology, toxicity and allergenicity and its general ecology. This document also addresses the potential for canola to outcross via pollen transfer and seed movement. Special emphasis has been given to the potential hybridisation between canola and its close relatives.

BIOLOGY OF CANOLA

INTRODUCTION

Origin of cultivated canola

The Brasscicaceae family (formerly Cruciferae) consists of approximately 375 genera and 3200 species of plants, of which about 52 genera and 160 species are present in Australia (Jessop & Toelken 1986). Of the 160 species of Brassicaceae present in Australia, several species are important weeds of the southern Australian cropping zone. Genera of economic importance in Australia are *Brassica* as a crop and *Raphanus*, *Sinapis*, and *Brassica* as weeds. In Australia, other important cropping weeds from the Brassicaceae family include *Hirschfeldia incana*, *Diplotaxis* spp. and *Sisymbrium* spp. (Rieger et al. 1999).

The *Brassica* genus consists of approximately 100 species, including species *Brassica napus* L., spp. *oleifera*, commonly known as oilseed rape, rapeseed or canola. *B. napus* is not native to Australia, and originated in either the Mediterranean area or Northern Europe. It is thought to have originated from a cross where the maternal donor was closely related to two diploid species, *B. oleracea* and *B. rapa*.

History of its use/domestication

Canola was cultivated by ancient civilisations in Asia and the Mediterranean. Its use has been recorded as early as 2000BC in India and has been grown in Europe since the 13th century, primarily for its use as oil for lamps (Colton & Sykes 1992). Canola was first grown commercially in Canada in 1942 as a lubricant for use in war ships. Canola was first grown commercially in Australia in 1969.

Traditionally, *B. napus* is unsuitable as a source of food for either humans or animals due to the presence of two naturally occurring toxicants, erucic acid and glucosinolates. However, in the 1970s, very intensive breeding programs in several countries including Australia produced high quality varieties that were significantly lower in these two toxicants. The term 'canola' refers to those varieties of *B. napus* that meet specific standards on the levels of erucic acid and glucosinolates. Those cultivars must yield oil low in erucic acid (below 2 %) and meal low in glucosinolates (total glucosinolates of 30 µmoles/g toasted oil free meal) (CODEX 1999), and are often referred to as "double low" varieties.

Uses of canola and by-products

Canola lines have become more important to the western world, through breeding for better oil quality and improved processing techniques (OECD Paris 1997). Edible oil was first extracted in Canada in 1956 (Colton & Potter 1999). Canola is now grown primarily for its seeds which yield between 35 % to over 45 % oil. Cooking oil is the main use but it is also commonly used in margarine. After oil is extracted from the seed, the remaining by-product, canola seed meal is used as a high protein animal feed.

Growth

General information on growth and agronomy

Canola in Australia, is mostly grown as a winter annual in winter dominant rainfall environments (between 30^{o}S and 38^{o}S). Only spring type canola varieties are grown in Australia and unlike winter varieties, do not need vernalisation (winter chilling) to flower, although vernalisation speeds up flowering. Rain-fed crops are sown with the onset of significant rain in April or May. Australian canola varieties flower for a 6-week period with crops ripening in late spring or early summer, after a 5 - 7 month growing season. This compares to a considerably longer growing season in Europe, which lasts for 12 months (due to a vernalisation requirement) and a rather short growing season (due to a long daylengths and warm temperatures) in Canada, which extends for less than 4 months.

Small areas of canola are sown in late spring - early summer in more temperate regions of Australia. These crops are located in areas that receive reliable rainfall, or have access to irrigation during summer as well as experience cool-mild temperatures at flowering. Summer grown canola crops are harvested in early autumn.

The average sowing rate of canola in Australia tends to be between 4 and 6 kg ha^{-1}, with hybrid seed sown at 3 kg ha^{-1}. These sowing rates are used to achieve a plant population of 50 - 70 plants m^{-2}. Under optimal soil moisture for germination, canola seed is sown at 2 - 4 cm depth which gives rapid emergence. When soil moisture is dry and soil temperatures high, seed can be sown into more moist areas of the soil, at depths up to 6 cm.

The growth of canola and its seed yield in Australia is almost always limited by the amount of water available to the crop, at least during seed maturation. In Australia, yields for broad acre production average 1 to 2 tonnes per hectare but range up to about 5 tonnes per hectare in situations with a long, cool growing season and adequate moisture.

Australian canola varieties are reasonably frost tolerant. Seedling losses may occur due to frosts, however unusually late frosts after flowering can result in aborted seeds and reduced yields.

Canola crops in Australia are harvested in summer. At this time, seeds have good storage characteristics due to low moisture, as well as high quality seed low in chlorophyll and free fatty acids. The majority of Australian crops are ‘swathed’ or windrowed whereby the crop is cut and placed in rows. This process hastens the

drying rate of the crop, reduces the possibility of seed losses from wind or hail and ensures even ripening. The windrow is picked up and threshed within 7 - 10 days after swathing, when the seed moisture has fallen to less than 8.5 %.

Canola is one of the most profitable crops available to grain growers in southern Australia and provides the opportunity for farmers to use more diverse cropping rotations. Like many other broadleaf crops, canola provides an important disease break during which the inoculum of cereal pathogens such as the take-all fungus (*Gaeumannomyces graminis*) decline. Canola root exudates have also been reported to have biofumigation effects on fungal inoculum (Kirkegaard et al. 1994), (Kirkegaard et al. 1998). Studies have shown that the root system of canola has beneficial effects on soil structure and soil moisture infiltration, resulting in higher yield and protein levels in the following cereal crop.

Canola is often the first crop grown following a pasture and benefits from the nitrogen fixed by legumes during the pasture phase. The subsequent crops following canola are generally wheat followed by a second wheat crop or a pulse, and then another cereal. The chosen pulse could be lupins (e.g. Western Australia) or field peas (e.g. western Victoria and South Australia). Due to the poor returns from pulses, alternating crops of canola and wheat are becoming more common, particularly in some regions of NSW (Norton et al. 1999).

Pests, diseases and weeds

INSECT PESTS

A number of insects and mites can damage canola crops. Insect pests such as the redlegged earth mite (*Halotydeus destructor*), blue oat mite (*Penthaleus major*), cutworms (*Agrotis infusa*), aphids (*Brevicorne brassicae* and *Lipaphis erysimi*), Diamond Back moths or cabbage moths (*Plutella xylostella*), heliothis caterpillars (*Helicoverpa punctigera* and *H. armigera*) and Rutherglen bug (*Nysius vinitor*) cause severe and widespread losses in some years. Western Australia recently experienced widespread problems with Diamond Back moths attacking crops prior to flowering in 2000. Significant insect damage to canola crops is most likely to occur during establishment and from flowering until maturity.

DISEASES

The most important pathogen of canola is blackleg, caused by the fungus *Leptosphaeria maculans*. Blackleg can be carried over from year to year on infected canola stubble which can kill seedlings or reduce seed yield in older plants. Scelerotinia stem rot (*Sclerotinia* spp.) is another major disease of canola. It has a wide host range of about 400 species of mostly broadleaf crops (e.g. lupins, field peas, beans) and weeds (e.g. capeweed). Growing canola after any of these crops or weeds can increase the risk of this disease. Other diseases include, phytophthora root rot caused by the fungus *Phytophtohora megasperma* var. *megasperma*, downey mildew (*Peronospora parasitica*) and alternaria leaf spot which is caused by the fungus *Alternaria brassicae* and can cause serious yield loss in wet seasons. A survey conducted in canola crops throughout Western Australia in 1998, revealed the presence of a number of viral diseases including Beet Western Yellow virus and

Cauliflower Mosaic Virus, both spread by aphids. In 1999, Beet Western Yellow virus was also detected in NSW (Howlett et al. 1999).

WEEDS

Broad leaf weeds, particularly weeds from the Brassicaceae family, are the most problematic in canola crops. There are no herbicides available to control Brassicaceous weeds in conventional canola once the seeds have germinated and seedlings have emerged (so called post-emergent herbicides). Consequently, competition from these weeds leads to significant yield losses in the crop. Furthermore, seeds of certain Brassicaceae species can contaminate canola seed, jeopardising the seed quality by increasing levels of erucic acid and glucosinolates.

Cultivation & distribution of *B. napus* in Australia

In Australia, canola is an established crop in the medium and high rainfall (400 mm and above) areas of southern Australia which represents the winter production cereal belt. However the development of early maturing varieties is expanding growing areas of canola into the low rainfall areas of the wheat belt.

Canola production has grown significantly in Australia over the last decade. Canola production has risen from approximately 100,000 ha in the early 1990s to an estimated total area of 1.4 Mha in 2000 (Colton & Potter 1999). Internationally, in 2000, China planted 6.5 Mha of canola, USA planted 5.7 Mha, and Canada planted 4.9 Mha (The Canola Association of Australia).

Canola occupies approximately 6 % of the cropped area in New South Wales, Victoria, South Australia and Western Australia (Norton et al. 1999). Canola grown in Western Australia, New South Wales and Victoria accounts for 85 % of Australia's total canola production (The Canola Association of Australia).

REPRODUCTION

Canola has entomophilous flowers capable of both self- and cross- pollination. Fertilisation of ovules usually results from self pollination since in a flowering crop, each flower produces a large amount of pollen and usually out competes with the pollen from adjacent flowers. However outcrossing can also occur at levels between 12 – 47 % (Williams et al. 1986); (Becker et al. 1992). Crosses with other plants can occur in two directions: canola can act as either a pollen donor (male) or pollen recipient (female). The level of out-crossing varies on the availability of insect pollinators, cultivar and weather. The normal means of reproduction is through seeds. There are no reports of vegetative reproduction under field conditions in Canada (Canadian Food Inspection Agency 1994).

Pollen characteristics

Most insect pollinated plants have relatively large (32-33 µm), sticky grains that do not become airborne readily. In contrast, grains from wind pollinated plants are generally light, dry and easily airborne. Canola pollen grains is an exception and intergrades in these situations. Under field conditions, canola has the ability to cross pollinate through physical contact between neighbouring plants and/or insect

pollination and whose pollen can also become airborne and potentially travel at least several kilometres downwind (Treu & Emberlin 2000).

Pollen movement

Wind

In general, wind-borne pollen plays a minor role in long-distance pollination. The vast majority of pollen travels less than 10 m and the amount of pollen decreases as the distance from the pollen source increases (Scheffler et al. 1993); (Timmons et al. 1995); (Thompson et al. 1999). The dispersal range of canola pollen is variable, from a few metres to 360 m or in extreme cases there is evidence of wind transfer up to 1.5 km (Timmons et al. 1995).

Most pollen grains released to the air flow will travel at least some way from the anthers but distances will depend on the dispersal processes operating. Air-borne pollen dispersal distances are variable in response to environmental and topographical conditions. Pollen movement will depend on wind direction, wind speed, topography (e.g. hills, slopes, valleys) and surrounding vegetation (Gliddon et al. 1999); (Thompson et al. 1999). Longer distance pollen transfer (termed 'regional pollen') occur when pollen grains are caught by upward air movements and are transported above the height of vegetation and the local air currents created by surface features. No research has been carried out on movement of canola pollen in atmospheric conditions such as convection currents, turbulent conditions and weather fronts. However research on pollen from other species has demonstrated that dispersal can occur over considerable distances (e.g. 380 km for arboreal pollen) (Tyldesley 1973).

Insect pollinators

The flowers of canola produce nectar with relatively high concentrations of sugars and have a colour and structure which makes them attractive to insects, particularly bees. In Australia, insects, particularly honeybees (*Apis mellifera*) are believed to play a major role in the transfer of pollen over long distances. However, other beneficial insects such as hoverflies (*Simosyrphus grandicornis*) which prey on aphids in canola, may inadvertently transfer pollen between plants. Bumblebees (*Bombus* spp.) play a major role in the transfer of pollen in the Northern Hemisphere (Cresswell 1999). Since bumblebees only occur in Tasmania and are geographically discrete, these insects play a minor role in the pollination of canola crops in Australia.

Pollinator behaviour

Insect foraging behaviour is complex, being dependent on a number of factors including spatial arrangement of plants, environmental conditions, plant density and availability of pollen (Rieger et al. 2002). Given abundant flowers, such as in a cultivated field, individual honey bee foragers tend to collect nectar and pollen from flowers in the same or immediately adjacent plants. Bee hives are commonly introduced into canola crops to facilitate with pollination and maximise seed set. In this situation, most foraging is carried out close to the hive and between neighbouring plants which may be a few dozen square metres in size (Nieuwhof 1963). Many studies have showed that a large proportion (up to 80 %) of bee flights are less than 1 m in distance, with the majority of pollen being transported by bees less than 5 m (e.g.

(Cresswell 1999); (Ramsay et al. 1999); (Pierre 2001). Occasionally however, bees may travel much further and studies have measured bee flight distances of 1 - 2 km (Eckert 1933), up to a maximum distance of 4 km (Ramsay et al. 1999); (Thompson et al. 1999). Loose pollen grains can be picked up from within a hive, so with the majority of honeybee colonies foraging up to 2 km in all directions from a hive, some pollen transfer and fertilisation up to 4 km may occur (Ramsay et al. 1999).

While bees will search a larger region for food during flowering, honeybees will only forage during daylight and are unlikely to carry pollen grains viable to effect fertilisation beyond 12 hours (Kraai 1962). Honeybees are very sensitive to barometric pressure, and decrease foraging distances in response to impending adverse weather (APHIS 1998). The mean distance of pollen dispersal is dependent not only on pollinator behaviour but also plant density and sparse areas of plants receive far fewer pollinator visits (e.g. (Kunin 1997))

Pollen viability

The distance and success to which pollen mediated gene flow is likely to occur is dependent not only on its dispersal in space, by either wind or insect action, but also on the length of time the pollen grain retains its potential for pollination. Pollen viability varies with environmental conditions, particularly temperature and humidity. Under controlled conditions in the laboratory, canola pollen can remain viable for between 24 hours and one week (Mesquida & Renard 1982). Under natural conditions pollen viability gradually decreases over 4 - 5 days (Ranito-Lehtimäki 1995). In Australia, canola crops flower in spring when temperature increases and humidity declines. Under these conditions, pollen viability may be reduced to 24 - 48 hours (M. Rieger, pers. comm.).

Outcrossing

The table below (Table 1) summarises a number of representative studies which have measured pollen dispersal distance and outcrossing rates between canola and related species. This summary shows a wide variation in canola pollen dispersal distance and outcrossing rates which have been influenced by factors such as local climatic conditions (e.g. wind direction, wind speed, temperature, humidity, rainfall), experimental design (e.g. size and orientation of plots), insect movements (Scheffler et al. 1993).

Canola pollen dispersal distance and outcrossing rates between commercial fields of non-GM herbicide tolerant canola and conventional canola were recently measured under Australian conditions by Rieger et al. (2002). Outcrossing rates between 0 and 2.6 km were variable. On an individual sample basis, the maximum outcrossing rate of 0.197 % was measured at 1.5 km, while between 0 and 2.6 km, outcrossing rates varied between 0 and 0.15 %. This study measured an outcrossing rate of less than 0.01 %, 2.6 km from the pollen source. More than 300,000 seeds per paddock were also tested at sites from 3 km to 6 km from the source and outcrossing was not detected. When averaged across the individual paddocks where outcrossing had occurred, at no distance was pollen flow greater than 0.07 %. Outcrossing occurred in 63 % of the fields, but only a few had outcrossing rates greater than 0.03 %. For

comparison, current EU standards allow accidental contamination of GM foodstuffs up to 1 %.

Table 1. Summary of representative studies on pollen dispersal distance and outcrossing rates between canola and related species

Reference	Country	Pollen dispersal distance (m)	% outcrossing
(Paul et al. 1995)	U.K.	0 m (mixed population)	3 - 12
(Scheffler et al. 1993)	U.K.	1 m, 3 m, 12 m, 47 m, 70 m	1.5, 0.4, 0.02, 0.00033, 0
(Staniland et al. 2000)	Canada	30 m	0.03
(Stringam & Downey 1982)	Canada	47 m, 137 m, 366 m	2.1, 1.1, 0.6
(Manasse & Kareiva 1991)	U.K.	50 m, 100 m	0.022, 0.011
(Simpson et al. 1999)	U.K.	54 m	0.05 - 0.11
Lamond, unpublished	Australia	100 m	<0.15
(Downey 1999)	Canada	100 m	0.02 – 0.28
(Scheffler et al. 1995)	U.K.	200 m, 400 m	0.0156, 0.0038
(Timmons et al. 1995)*	Scotland	1.5 km, 2.5 km	1.2, 0.8
(Rieger et al. 2002)	Australia	3 km	< 0.01
(Thompson et al. 1999)*	U.K.	4 km	5

*using emasculated 'bait' plants – petals and stamens removed.

A number of studies that have reported maximum distances of up to 1.5 km for wind-mediated pollen dispersal by measuring directly for pollen (Timmons et al. 1995) and between 400 m and 4 km for pollination by using male sterile or emasculated bait plants to detect pollination (Timmons et al. 1995; Thompson et al. 1999; Norris et al. 1999). Studies using male sterile bait plants, only represent the potential for gene flow, with pollination levels for male fertile plants likely to be much lower. Furthermore, it should be noted that while such measurements give an indication of the potential for outcrossing they will not provide information on the likelihood of outcrossing actually occurring (Salisbury 2002b).

Fruit & Seeds

Fruit & seed production

The fruiting bodies produced by the Brassicaceae family are siliques, commonly called pods (Buzza 1979). Between 15 and 25 seeds are produced per pod. Each canola plant produces hundreds of small, spherical, light brown to black seeds. Each seed is generally 1 to 2 mm in diameter (Buzza 1991). There are generally 250,000 to 300,000 seeds per kilogram of seed.

Seed ecology

Viability/germination

Optimal germination conditions for canola are 20^{o}C, high water availability (e.g.–0.2 MPa) and exposure to light (Pekrun et al. 1998). Consequently, the greatest proportion of canola plants that germinate after harvest ('volunteers') emerge in

response to tillage (Pekrun et al. 1998) since canola seed develop an ability to react to short light flashes under certain soil conditions (see below).

Dormancy

Canola seed show virtually no signs of dormancy at maturity (Lutman 1993); (Pekrun et al. 1998). However non-dormant canola seed may enter dormancy if environmental conditions are unfavourable for germination (referred to as 'secondary dormancy'). Induction of secondary dormancy in canola occurs in response to sub optimal germination conditions such as large temperature fluctuations, low soil water availability (e.g. –2.0 MPa), long exposure to darkness (Pekrun et al. 1997b), and sub-optimal oxygen supply (Pekrun et al. 1998). High temperatures greater than 20^{o}C can also induce dormancy in some genotypes (Linder 1998); (Gulden et al. 2000). Secondary dormancy can be removed at low temperatures (2 – 4^{o}C) (Gulden et al. 2000) or by alternating warm and cold temeperatures (Pekrun et al. 1998).

During a prolonged exposure to darkness and water stress, canola seeds develop light sensitivity. Light sensitivity enables seed to germinate in response to very short exposure to light, as experienced during soil cultivation. The presence of large quantities of crop residues (e.g. in zero tillage systems) can also create shaded conditions that allow canola seed to develop light sensitivity (Legere et al. 2001). The development of secondary dormancy and persistence of canola can vary between genotypes of European cultivars (Pekrun et al. 1997a) and Canadian cultivars (Gulden et al. 2000).

Persistence of canola seed is considerably longer in undisturbed soils compared to cultivated soils (Chadoeuf et al. 1998). Studies in the Northern Hemisphere have reported viable seeds of canola persisting in disturbed soils for at least 5 years and possibly up to 10 years or more in undisturbed soil (Masden 1962); (Pekrun et al. 1997a); (Vaughan et al. 1976). Light sensitivity in canola seed is the major factor contributing to a shorter persistence of seeds in cultivated soils. Persistence will also vary between soil types.

Despite the lack of Australian data, overseas studies suggest that high temperatures and low soil moisture availability experienced after harvest in Australia, may provide conditions to induce secondary dormancy which may contribute to higher persistence rates than under European or Canadian conditions. It is possible that cold and moist conditions experienced in the Northern Hemisphere after canola harvest is more conducive to germination of canola seed than warm, dry conditions experienced after harvest in Australia. The persistence of viable canola seed in the soil under Australian field conditions is poorly understood and further research is needed.

Seedbanks

Large seedbanks of canola can build up in the soil as a result of high amounts of seed losses before and during harvest. Other mechanisms responsible for adding seed to the seedbank include ungerminated seed from sowing, seed produced from volunteers and/or seed losses from mature plants due to heavy rainfall, hail, strong winds or lodging.

Loss of seed from plants, particularly in hot and windy conditions, can be up to 70 % for canola (Colton & Sykes 1992), which may lead to a substantial number of volunteers appearing in subsequent crops. Data suggest that canola seed density in the soil can range between 2000 seeds m^{-2} in Canada after harvest (Legere et al. 2001) to 10,000 seeds m^{-2} in the U.K. (Lutman 1993). Canadian studies have also shown that seedbank density declined ten-fold in the first year, but only slowly declined thereafter. Seasonal variations in seedbank density in Canada occurred as a result of seeds being produced by volunteer plants every spring thereby replenishing the seedbank (Legere et al. 2001). Recent field surveys conducted across Canada, found volunteer canola still present in fields 4 - 5 years after a canola crop had been grown, albeit at low densities (Legere et al. 2001).

While there are considerable genotypic variation in the development of secondary dormancy in canola (Pekrun et al. 1997a), generally low nutrient status and dark conditions are the major contributing factors to the persistence of the seedbank of canola. The vertical distribution of seed in the soil has a significant impact on the persistence of canola seed in the seedbank. Seeds are more likely to persist at deeper soil depths than at shallow depth in the field (Pekrun et al. 1998). A greater proportion of germination occurs at shallow depths due to dormancy breaking temperature alterations and exposure to light (Pekrun et al. 1998). Persistence will also vary with soil type. Pekrun et al. (1998) found that sandy soils, with low water holding capacity, tended to have a greater proportion of dormant canola seeds in the seedbank than clay soils which have high water holding capacity (Pekrun et al. 1998).

WEEDINESS OF CULTIVATED CANOLA

Cultivated areas

As with all crops cultivated and harvested at the field scale, some seed may escape harvest and remain in the soil until the following season when it germinates either before or following seeding of the succeeding crop. In some instances the volunteers may give considerable competition to the seeded crop and warrant chemical and/or mechanical control. Volunteers can also be expected away from the planting site (e.g. along roadsides and around storage facilities) as a result of transportation of seed out of fields (e.g. in farm equipment) and spillage during transport.

Canola occurs as a minor problem in agricultural areas within Queensland, New South Wales, Victoria, Tasmania and Western Australia (Groves et al. 2000). Surveys have shown that canola occurs as a volunteer weed in up to 10 % of cereal crops in southern Australia (Lemerle et al. 1996). Volunteer canola is among the 20 most common weeds in fields in Alberta, Canada, occurring as a residual weed in 11.8 and 10.5 % of all wheat and barley fields surveyed in 1997 (Thomas et al. 1998). Volunteer canola has been recognised as fourth ranked weed in the central cropping region of Manitoba, Canada (Kaminski 2001). Canola also occurs as a weed in cropping regions in the U.S.A (Weed Science Society of America 1992).

Non-cropped disturbed habitats

Canola is a plant of disturbed habitats (agricultural fields) and will take advantage of disturbed land (Salisbury 2002c). Canola seed can be disseminated to neighbouring non-agricultural areas by a variety of mechanisms: canola windrows can actually be

blown across or off a field in a strong wind; or during harvest, a small fraction of seed that is not collected in the grain bin but is blown out the back of the harvester with the canola straw and chaff. However, canola is a poor competitor and is not regarded as an environmentally hazardous colonising species. Unless the habitat is regularly disturbed, or seed replenished from outside, canola will be displaced by other plants (Salisbury 2002c).

Populations of canola can be found on roadside verges, in field margins and along railway lines in all countries where it is grown. An Australian survey encompassing a total of 4000 km of road and 400 observations (Agrisearch 2001) recorded incidences of canola plants growing within 5 m of the roadside in major canola growing districts in southern NSW (31 %), Victoria (13 %), SA (9 %), WA (20 %) and Tasmania (14 %). The occurrence of predominantly isolated plants suggests they had not originated from seed dropped from plants the previous season, but resulted from individual seeds being dropped during transportation. Dignam (2001) surveyed 103 local councils across Australia and canola was present in 30 %. In Australia, the density of roadside canola populations was mostly low. Only 5 % of councils and 4 % of road and rail authorities surveyed, indicated canola was present in large numbers.

Undisturbed natural habitats

Canola is not considered a significant weed, nor invasive of natural undisturbed habitats in Canada (Canadian Food Inspection Agency 1994); (Warwick et al. 1999); (Beckie et al. 2001) or Australia (Dignam 2001). Crawley et al. (2001) showed that over a 10 year period, canola was not invasive nor persistent in undisturbed non-arable habitats in the U.K. Due to selective breeding, crop plants only function optimally under managed agricultural conditions, such as high soil fertility or low plant competition. These conditions rarely occur in natural habitats, resulting in poor fitness of canola plants (i.e. reduced recruitment, low survivorship, poor competitive ability, low seed production).

TOXICITY & ALLERGENICITY

Traditional rapeseed is unsuitable as a source of food for either humans or animals due to the presence of two naturally occurring toxicants in the seed, erucic acid and glucosinolates. The presence of erucic acid in rapeseed oil has been associated with fat accumulation in the heart muscle of laboratory rats, resulting in cardopathogenic effects. Glucosinolates, located in the seed meal, were found to cause thymus enlargement in laboratory animals and therefore their presence also limited the nutritional value of the meal as feed for livestock. Canola are cultivars of rapeseed that yield oil low in erucic acid (below 2 %) and meal low in glucosinolates (less than 30 µmoles g^{-1}). Breeders have systematically replaced the seedstock with varieties that were selected with low erucic acid content (refer to '**History of its use/domestication**'). Food Standards Australia New Zealand (FSANZ), formerly called Australia New Zealand Food Authority (ANZFA), does not consider canola meal as a food fraction suitable for humans due to the presence of glucosinolates (ANZFA 2001). The quality requirements of commercial canola oil production dictate the absence of any protein in the final product.

No allergic reaction to fats (including canola oil) has been reported by humans. Allergic sensitisation to canola can occur via the lungs (through inhaling pollen) or through skin contact (e.g. during handling). High incidences of hay fever and/or

bronchial asthma have been measured in canola growing areas in Europe during flowering (Bugur & Arner 1978). Other studies conducted in Europe show a low prevalence of allergy to oilseed rape pollen (less than 0.2 %) unless the subjects were occupationally exposed. Those affected, with one exception, were already atopic and allergic to other pollens (Fell et al. 1992). Volatile organic compounds given off by growing canola plants have been shown to play a role in respiratory mucosa and conjunctiva irritation associated with airborne releases from oilseed rape (Butcher et al. 1994).

Data collected on the allergenicity of canola pollen is often confounded by the other flowering plants, particularly grasses, which flower at similar times. Soutar et al. (1995) found people who complained of symptoms in relation to the flowering of oilseed rape were rarely allergic to the plant and fewer than half were atopic. Nevertheless, they usually showed increased bronchial reactivity during the season, which may have been due in some cases to other allergens but in others to non-specific irritant effects of the air. There is no evidence of cross-reactivity between canola and grass pollen (Welch et al. 2000).

Occupational allergies to plants can take the form of either immediate hypersensitivity or delayed hypersensitivity reactions. The latter frequently occur as a consequence of handling plant material and generally manifest as contact dermititis. However studies have shown that exposure to oilseed flour (contained in animal fodder) may be a possible cause of occupational asthma in farmers (Alvarez et al. 2001). Those at risk from pollen include the rural population in general and farm workers in particular.

POTENTIAL FOR POLLEN MEDIATED GENE TRANSFER FROM CANOLA TO OTHER ORGANISMS

There are a number of factors which affect pollen mediated gene flow between plants, these include sexual compatibility of the recipient species (whether the same or different species), flowering time, size of pollen source and sink, pollination vectors, pollen viability and environmental factors.

TRANSFER BETWEEN CULTIVATED & FERAL *BRASSICA* SPECIES

Other canola crops

With a potential for outcrossing rates of 10 – 50 %, it is highly likely that gene flow will occur between populations of canola. The transfer of genes between canola populations is well documented (e.g. (Paul et al. 1995); (Scheffler et al. 1995); (Downey 1999)). Since canola pollen can travel significant distances via insects and wind, these studies (and others) have made attempts to define the probabilities of outcrossing with increasing distances between populations.

Gene flow from large commercial canola fields to small plots in Canada tended to be at low levels (46 m = 2.1 %, 137 m = 1.1 % and 366 m = 0.6 %) (Thomas 2000). In 1998, substantially lower outcrossing rates were measured between large commercial fields in Canada, ranging from 0.1 to 1.5 % at 20 metres to 0.1 to 0.4 % at 100 metres (Thomas 2000). In the U.K., outcrossing rates of 0.022 % at 50 m and 0.011 % at 100 m were measured by Manasse and Kareiva (1991). At the same site, higher outcrossing rates were measured by (Scheffler et al. 1995) 0.0038 % outcrossing at

400 m. Differences in outcrossing rates are attributed to different climatic conditions and differences in experimental design.

Although the probability of outcrossing appears to be low, the large number of canola flowers and the many small seeds produced per plant ensures a substantial quantity of outcrossed seed can still be produced. Some seed may shatter onto the ground before or at harvest and germinate the following season with the succeeding crop. Although many of these seedlings may be killed by frost, disease, insect attack, early herbicide treatments and/or tillage, a proportion of seedlings may either survive or emerge later in the season, to compete with the succeeding crop, warranting further chemical or mechanical control.

The probability of gene flow is a function of spatial scale of the introduction. Limited field experiments may not accurately predict behaviour in widespread plantings. Estimates of low levels of gene flow from moderate-sized plots to larger planted areas (e.g. (Scheffler et al. 1993)) are more representative of gene flow from a feral population to a nearby crop, than the reverse (Squire et al. 1999). At larger spatial scales there is a greater possibility for contact with sensitive species or habitats or for landscape-level changes because at larger scales, more ecosystems could be altered.

There have been a small number of studies aimed at quantifying gene flow between canola crops on a regional scale ie. sites within an agricultural system. A study conducted in the U.K. found seeds from plants which had been fertilised by pollen of a second genotype whose nearest known source was 4 km away (Thompson et al. 1999). In this case, pollination was not wholly due to wind dispersal and may have been transported by occasional visits by pollinating insects. A similar conclusion was drawn from a large scale gene flow study in Australia using imidizolinone tolerance in canola as the marker trait (Rieger et al. 2002). In this study, low levels of gene flow were detected 2.6 km from the pollen source and attributed to insect mediated pollination events.

Brassica rapa

Brassica rapa ssp. *sylvestris* (= *B. campestris* L.) (diploid) is a closely related species of canola (amphidiploid), which has a similar life history to canola, but with a shorter growing season. *B. rapa* varieties were grown on a limited scale in Australia in the 1970s and early 1980s. By mid 1980s, breeding programs on *B. rapa* ceased in Australia and *B. rapa* was superseded by the release of superior quality canola varieties (Colton & Potter 1999). *Brassica rapa* is still grown as oilseed rape crops in Europe in both spring and winter forms.

Brassica rapa ssp. *sylvestris* is distributed throughout Queensland, New South Wales, Victoria, Tasmania and South Australia and sometimes occurs as a weed of disturbed and cultivated land (Auld & Medd 1987). While not regarded as a problem in South Australia, it is a minor problem in Queensland, New South Wales and Victoria and a major problem in cereals and vegetables in Tasmania (Hyde-Wyatt & Morris 1989); (Holm et al. 1997); (Groves et al. 2000).

Brassica napus (AACC) and *B. rapa* (AA) have a common set of chromosomes, making interspecific outcrossing extremely common (e.g. (Bing et al. 1991);

Jorgensen and Andersen; (Scheffler & Dale 1994b); (Bing et al. 1996); (Brown & Brown 1996); (Mikkelsen et al. 1996a); (Mikkelsen et al. 1996b); (Salisbury & Wratten 1997); (Jorgensen 1999); (Snow & Jorgensen 1999). Viable hybrids can be produced in the field when canola is crossed with *B. rapa* (Jorgensen & Andersen 1994); (Bing et al. 1996); (Brown & Brown 1996); (Mikkelsen et al. 1996b); (Metz et al. 1997) in either direction. However, the frequency of hybrids depends on parental genotypes, experimental design, population size (Palmer 1962); (Bing et al. 1991); (Jorgensen & Andersen 1994); (Jorgensen et al. 1996), (Jorgensen et al. 1998); (Landbo et al. 1996); (Hauser et al. 1997); (Scott & Wilkinson 1998); (Jorgensen 1999).

Gene flow measurements by Scott and Wilkinson (1998) from canola to *B. rapa* populations growing outside the field boundaries, suggest that hybridisation frequencies are low and (0.4 - 1.5 %) and seedling survivorship low (less than 2 % of all hybrid seedlings survived). Hybridisation frequencies are higher when *B. rapa* occurs as a weed within canola crops, but varies significantly with experimental design. Danish studies have shown that the proportion of hybrids produced between canola and *B. rapa* can vary from 93 % to 9 %. Generally a higher frequency of hybridisation occurred between single *B. rapa* plants in canola crops (Jorgensen et al. 1996). *B. rapa* is an obligate outcrosser and consequently more hybrids are found on *B. rapa* as the female (Jorgensen & Andersen 1994); (Jorgensen et al. 1996); (Hauser et al. 1997); (Jorgensen et al. 1998). Where natural interspecific hybrids occur, hybrids have reduced fertility and low seed set (average 2-5 per pod) compared with the parents (Jorgensen & Andersen 1994). Reduced dormancy of *B. rapa* x canola hybrids relative to the persistent wild *B. rapa* (Jorgensen et al. 1999), coupled with the reduced fertility of the inter-specific hybrid (Jorgensen et al. 1999) makes it very unlikely that populations of these hybrids will persist.

Brassica juncea

Brassica juncea occurs in Queensland, New South Wales, Victoria, South Australia and Western Australia (Groves et al. 2000). This species is only regarded as a minor problem in agricultural areas in New South Wales and Victoria (Groves et al. 2000).

Canola (AACC) and *B. juncea* (AABB) have a common set of chromosomes, enhancing the likelihood of interspecific hybridisation and gene flow (Salisbury 2002a). Spontaneous occurrence of interspecific hybrids in the field has been reported in several countries (Bing et al. 1991), (Bing et al. 1996); (Frello et al. 1995); (Jorgensen et al. 1998); (Bielikova & Rakousky 2001). A hybridisation rate of 3 % was measured from *B. juncea* plants surrounded by canola as the male parent (Jorgensen et al. 1999). Similar frequencies of hybridisation (4.7 %) between canola and *B. juncea* were measured by Bing et al. (1991). In the other direction, with canola as female and pollinated by *B. juncea*, the production of hybrids, have been less successful (Jorgensen et al. 1998). Pollen fertility was low (0 – 28 %), although the canola markers were all transferred to the first backcross generation with *B. juncea* (Jorgensen 1999). Bing et al. (1991) found there was potential for hybrids between canola and *B. juncea* to produce viable seed that could survive to the next generations.

Vegetable *Brassica* crops (*Brassica napus*, *B. oleracea* and *B. rapa*)

Gene flow from canola to *B. napus* vegetables (swedes, rutabaga, Siberian kale) is possible. Gene flow to *B. rapa* vegetables (e.g. turnip, Chinese cabbage, pak choi) is also possible due to a common set of chromosomes. However, *B. napus* and *B. rapa* vegetables are generally harvested prior to flowering which minimises the chance of hybrids occurring. Furthermore, these plants are not recognised as weeds in agricultural environments in Australia.
No hybrids have been reported in the field between canola and *B. oleracea* vegetables (cauliflower, Brussel sprouts, broccoli, several kales, kohlrabi etc.). *B. oleracea* vegetable crops are generally harvested prior to flowering and seed development, unless being used as a seed production crop.
Forage rape (*B. napus*) crops rarely flower and are usually consumed by foraging animals before seed development.

TRANSFER BETWEEN CULTIVATED & WEEDY *BRASSICACEAE* RELATIVES

Many Brassicaceae species occur as weeds of disturbed habitats in Australia, particularly agricultural habitats. There are several inherent biological characteristics common to many species of Brassicaceae which contribute to its success as a weed in these habitats. In general, Brassicaceous weeds have a high level of seed dormancy and seeds can remain viable in the soil for several years. Studies have shown that *Raphanus raphanistrum* (wild radish) seeds can remain viable in the soil for at least 6 years (Cheam & Code 1998). It has been suggested that seed dormancy in many Brassicaceae species, such as wild radish, is related to the presence of a silique (pod) that surrounds the seeds, physically inhibiting germination (e.g. (Mekenian & Willemsen 1975)). Once seed dormancy is released, germination requirements are flexible resulting in a number of germination events throughout a growing season. This is a selective trait that allows weeds to escape control events within a crop. Other weedy characteristics common to Brassicaceous species include the ability to produce large number of seeds quickly and strong competitive ability especially in crops.

Potential gene flow data between *B. napus* and other Brassicaceae species has been reviewed by (Salisbury 1991), (Scheffler & Dale 1994a), (Salisbury & Wratten 1997) and (Rieger et al. 1999) and summarised below in Table 2. Hybrids have been produced between *B. napus* and a Brassicaceae species in Category III and IV following hand pollination and the use of embryo rescue methods. However the hybridisation data from these sophisticated experiments gives no measure of the likelihood of successful hybridisation in the field (Scheffler & Dale 1994a). No field hybrids have been reported for any Brassicaceae species from Category III to VI and as such, regarded as extremely unlikely to be hybridise naturally with canola. Category I species (e.g. *B. rapa* and *B. juncea*) have the highest sexual compatibility with *B. napus* with a high probability of hybridisation and gene introgression occurring between these species and *B. napus*. Hybrids between *B. napus* and Category II species (e.g. *Raphanus raphanistrum*) have been recorded in the field, although successful gene introgression has not been measured.

Table 2. Potential gene flow between canola (*B. napus*) & Australian *Brassicaceae* species

Category	I	II	III	IV	V	VI
Tribe	*Brassicaceae*	*Brassicaceae*	*Brassicaceae*	*Brassicaceae*	*Brassicaceae*	Other
Glasshouse 'rescued' hybrids	Yes	Yes	Yes	Yes	No	No
Glasshouse hand hybrids	Yes	Yes	Yes	No	No	No
Field hybrids	Yes	Yes[2]	Not reported	Not reported		
Gene introgression	Yes/Likely[1]	Not reported				
Weeds	*Brassica rapa* *Brassica juncea*[1]	*Raphanus raphanistrum* *Hirschfeldia incana* *Sinapis arvensis*	*Brassica fruticulosa* *Brassica nigra* *Brassica tournefortii* *Diplotaxis muralis* *Diplotaxis tenuifolia* *Rapistrum rugosum*	*Brassica oxyrrhina* *Diplotaxis tenuisiliqua*	Conringia orientalis Carrichtera annua **Cakile maritima**	Capsella bursapastoris Cardaria draba Lepidium sp. Myagrum perfoliatum Sisymbrium orientale Sisymbrium irio Sisymbrium erysimoides *Sisymbrium officinale*
Condiment, fodder & vegetable species	Forage *B. napus*[1] *B. napus* vegetables[1] *B. rapa* vegetables[1] Condiment *B. juncea*[1]		*Brassica alboglabra*[3] *Brassica chinensis*[4] *Brassica nigra* *Brassica oleracea* *Brassica pekinensis*[4] *Raphanus sativus* *Sinapis alba*			

® DECREASING SEXUAL COMPATIBILITY ®

[1] Considered likely to happen over a period of time **if** the species are in physical proximity and have flowering synchrony.
[2] Frequency of interspecific hybrids approx. 10^{-4} to 10^{-8}. Likelihood of subsequent introgression or formation of fertile amphidiploids significantly less again.
[3] This species is sometimes considered to be a subspecies of *B. oleracea*.
[4] These species have sometimes been considered to be subspecies of *B. rapa*.

Table 2. Potential gene flow between canola (*B. napus*) & Australian *Brassicaceae* species (cont.)

Category	I	II	III	IV	V	VI
Tribe	*Brassicaceae*	*Brassicaceae*	*Brassicaceae*	*Brassicaceae*	*Brassicaceae*	Other
Glasshouse 'rescued' hybrids	Yes	Yes	Yes	Yes	No	No
Glasshouse hand hybrids	Yes	Yes	Yes	No	No	No
Field hybrids	Yes	Yes	Not reported	Not reported		
Gene introgression	Yes/Likely#	Not reported*				
Native species						*Arabidella* (6 *sp.*) *Balbaretinia* (1 *sp.*) *Barbarea* (2 *sp.)* *Blennodia* (25 *sp.*) *Cardomine* (5 *sp.*) *Carinavalva* (1 *sp.*) *Cheesemania* (1 *sp.*) *Cuphonotus* (2 *sp.*) *Geococcus* (1 *sp.*) *Harmsiodoxa* (3 *sp.*) *Irenepharsus* (3 *sp.*) *Lepidium* (35 *sp.*) *Menkea* (*6 sp.*) *Microlepidium* (*2 sp.*) *Pachymitus* (*1 sp.*) *Phlegmatospermum* (*4 sp.*) *Rorippa* (*4 sp.*) *Scambopus* (*1 sp.*) *Stenopetalum* (*9 sp.*)

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Gene movement between canola and other members of the Brassicaceae family occurs at extremely low levels in nature. Factors which may influence the rate of outcrossing include environmental conditions, genotypes and experimental design (e.g. size of the donor and recipient populations). Large sources of pollen may have a considerable effect on a small population of compatible plants (Ellstrand et al. 1989); (Klinger et al. 1991). The flowering periods of many weedy Brassicaceae species overlap with canola. Depending on the season and region, the synchrony of flowering between species can also influence the rate of outcrossing in the field. Generally, canola can flower from September to January while many weedy Brassicaceae species begin flowering around August. However this will vary with environmental conditions and under ideal growing conditions, some weedy species may flower at any time during the year (Rieger et al. 1999).

Naturally occurring hybrids in the field between canola and Brassicaceae species have been reported for three economically important weed species in Australia: *Raphanus raphanistrum* (wild radish), *Hirschfeldia incana* (Buchan weed) and *Sinapis arvensis* (charlock). The potential for outcrossing and gene introgression in these three species are discussed in more detail below.

Canola - Raphanus raphanistrum (wild radish)

Raphanus raphanistrum (wild radish) occurs in Queensland, New South Wales, Victoria, Tasmania, South Australia and Western Australia (Groves et al. 2000). It is a major weed of cropping regions, particularly in southern Australia. Large numbers of wild radish can also occur along roads and railway lines in and around canola growing areas in Australia (Agrisearch 2001); (Dignam 2001). However, it has never been reported as a weed in natural habitats (e.g. National Parks) in Australia (Dignam 2001).

Wild radish has been shown to hybridise with *B. napus* under both laboratory and field conditions (Chevre et al. 1997); (Darmency et al. 1998); (Eber et al. 1994); (Mikkelsen et al. 1996a); (Rieger et al. 1999), (Rieger et al. 2001). Differences in the frequency of outcrossing between these species depend on the direction of pollination. Studies suggest that hybridisation into canola (canola pollinated by wild radish) is more likely to occur. However, under Australian field conditions, the outcrossing rate from wild radish to canola (i.e. canola pollinated by wild radish) was still very low (Rieger et al. 1999); (Rieger et al. 2001). Two hybrids were located in 52×10^6 canola seedlings (an outcrossing rate of 1 in 26×10^6). In comparison, French studies estimate that the frequency of hybrids from wild radish pollinating canola, was $5 \times 10^{-4} - 2 \times 10^{-5}$ (Chevre et al. 2000). In the other direction (canola pollen fertilising wild radish), hybridisation occurred at 3×10^{-5} to 10^{-7} (Chevre et al. 1999); (Chevre et al. 2000). In the Australian study by Rieger et al. (2001) no hybrids were detected amongst 25,000 wild radish seedlings.

Hybrid seeds (canola x wild radish) although rare in proportion to the total soil seedbank (0.1 seeds per m^2), can survive for similar periods as canola seeds, decreasing to 1 % and less after 3 years (Chadoeuf et al. 1998). Fertility was improved in subsequent backcross generations with wild radish (Chevre et al. 1997); (Chevre et al. 1998).

Since hybridisation into canola is more likely, hybrid individuals are mostly expected to occur in the crop. A large proportion of the seed is expected to be harvested and removed from the site (Rieger et al. 2001). However due to pod shattering or spillages at harvest, a proportion of the original hybrid seed will remain on site. Consequently, hybrid individuals are expected to occur intermingled with canola volunteers in the following year. Cultivation and a number of alternative herbicides are available to control both types of volunteers (Rieger et al. 2001).

Gene escape from canola to other plant species will only occur as a result of hybridisation and introgression of genes into the wild population. Introgression involves the transfer of segments of a genome and genes between hybrid back-crossing with the wild population. While hybridisation between canola and wild radish has been documented (see above), successful introgression of genes from canola into wild radish has not been documented beyond 4 generations ((Chevre et al. 1997),(Chevre et al. 1998), (Chevre et al. 1999)).

Canola – Hirschfeldia incana (Buchan weed)

Hirschfeldia incana (Buchan weed) occurs in Queensland, New South Wales, Victoria, Tasmania, South Australia and Western Australia. It is a minor problem in agricultural areas of Queensland and New South Wales (Groves et al. 2000). Buchan weed will also invade disturbed native vegetation. It can also occur in large numbers along railways and roadsides in canola growing regions in Australia (Dignam 2001).

Spontaneous hybridisation with canola is known to occur in this species, although where Buchan weed is fertilised by canola pollen, the frequency of hybridisation is low. When canola was mixed with Buchan weed at a density of 1:625, Buchan weed: canola, 1.5 % of the Buchan weed seeds were hybrids (Lefol et al. 1996b). In the other direction, where canola is pollinated by Buchan weed, in a 1:1 ratio, hybridisation frequency increased and 70 % of seeds were hybrids (Lefol et al. 1996b).

Hybrids were as vigorous as, if not more competitive than the weed although fertility was low at an average of 0.2 seeds per plant (Chevre et al. 1999). Hybrids produced only 0.5 seeds per plant after spontaneous backcrossing with Buchan weed (Lefol et al. 1996b). Survival of hybrid seed in undisturbed soil was intermediate between the parents, with nearly 15 % survival the first year which decreased to complete extinction at 41 months (Chadoeuf et al. 1998).

While hybridisation between canola and Buchan weed has been documented (see above), gene escape from canola to other *H. incana* is extremely unlikely since there have are no documented occurrences of introgression of genes into the wild population.

Canola – Sinapis arvensis (charlock)

Sinapis arvensis (charlock) occurs in Queensland, New South Wales, Victoria, Tasmania, South Australia and Western Australia. For the most part, charlock is a problem in agricultural areas and is particularly serious weed in cropping regions of New South Wales (Groves et al. 2000). It can also occur in disturbed sites along roadsides and railways in canola growing regions in Australia (Dignam 2001).

Charlock and canola are generally not considered to be sexually compatible. The majority of studies have found embryo rescue or ovule culture the only methods for successfully effecting hybridisation. Under open pollination conditions and using charlock as the pollinator, six hybrids were obtained from 50 000 flowers (Lefol et al. 1996a) and 0.18 hybrid seeds obtained per 100 flowers (Chevre et al. 1996). Hybridisation was not detected with canola as the pollinator (Lefol et al. 1996a); (Downey 1999).

Hybridisation between canola and charlock has not been documented under field conditions and as a consequence, gene escape from canola to charlock through gene introgression is extremely unlikely to occur.

TO OTHER PLANT SPECIES

Another mechanism by which canola can transfer genetic material to sexually non-compatible plants is through 'bridging'. Bridging is defined as 'a mating made between two incompatible species'. Such a possibility of the 'bridging' phenomenon may occur with *B. juncea* acting as the intermediate species. The occurrence of hybrids between canola and *B. juncea* is rare, and moreover, the hybrids do not persist long enough in the environment due to poor fertility, poor germination, and high seedling mortality, to serve as a bridge species (APHIS 1998).

Furthermore, crosses between *B. juncea* and *B. nigra* are not fully compatible and it follows that crosses between *B. napus* hybrids, and *B. nigra* would be even less compatible. Another genetic barrier for gene transfer is that it has to take place by chromosomal crossing over in the canola and *B. juncea* hybrid to be stably introduced into *B. nigra* (Scheffler & Dale 1994b).

Brassica rapa may also act as an intermediate species or bridging species between canola and incompatible species. Canola and *B. rapa* can produce hybrids and introgress into successive generations (Metz et al. 1997). Gene transfer between canola x *B. rapa* hybrids and species unrelated to canola, may occur therefore between those species related to *B. rapa*. However the stability of gene introgression has not been measured. Furthermore, the persistence of hybrids formed in crosses of canola x *B. rapa* is considerably less than *B. rapa* due to lowered fertility and reduced dormancy (Bing et al. 1991);(Jorgensen et al. 1999) which may lower the probability of *B. rapa* acting as a bridging species.

TO OTHER ORGANISMS

No published evidence was found from comparative analyses of numerous gene sequences from microorganisms and plants to suggest evidence of strong inter-kingdom gene homologies that would be indicative of recent or frequent gene

exchanges between plants and microorganisms, with the exception of T-DNA of the Ti-plasmid of *Agrobacterium*.

Phylogenetic comparison of the sequences of plant and bacterial genes suggests that horizontal gene transfer from plants to bacteria during evolutionary history has been extremely rare, if occurring at all (Doolittle 1999). Horizontal gene transfer from plants to bacteria has not been demonstrated experimentally under natural conditions (Syvanen 1999). Even if a rare plant-to-microbe gene transfer were to take place, there is no reason to believe that such a transfer of any of the sequences would pose any plant pest risk. However, the level of risk would depend on the protein/s encoded by the sequence. For instance, herbicide tolerant genes (e.g. *bar* gene for glufosinate-ammonium tolerance and CP4-EPSPS gene for glyphosate tolerance) are derived from common soil bacterium, so an effect from a transfer of sequences would not be expected. The evidence suggest DNA transfer from canola to microorganisms is highly unlikely if not altogether impossible.

POTENTIAL FOR DISSEMINATION OF CANOLA VIA SEED MOVEMENT

Seed movement can be an important factor in overall spatial and temporal gene movement. Seed mediated gene flow is typically over shorter distances although occasionally over very long distances and with the added condition that establishment of a plant containing the gene in question is more likely (Raybould & Gray 1993). Dormancy is a mechanism for dispersing seed temporally.

WIND

The small size of canola seeds and their high numbers on post harvest fields may facilitate some dispersal by wind (Lutman 1993). Furthermore, anecdotal evidence exists to indicate strong winds or wind storms can move entire pods from harvested material in windrows. The dispersal distance will depend on the wind strength, the amount of trash on the ground and the moisture content of the material. It is reasonable to expect, that seeds and pods of low moisture content, may be transported within the field, to adjacent fields or outside agricultural areas.

WATER

No data exists on seed transport rates by water of canola and other *Brassica* species. However it is probable that because the specific gravity of bare seed is slightly higher than that of water, most of the seed would tend to sink, especially after soaking. It is likely however that seeds would be transported relatively easily as bed load sediment in rivers and creeks. There is a high likelihood that the majority of seed being transported by water would be carried to positions unfavourable for establishment.

HUMANS (CLOTHING, VEHICLES)

The greatest potential for the movement of canola seeds is from post harvest spillage by agricultural machinery or during transportation away from the production areas. Seed from the trial sites may also be moved on electrostatically charged containers (e.g. tarpaulins and bags) in storage bins. Observations by OGTR monitoring officers and industry personnel have identified small amounts of seed leaving trial sites on or in clothing and boots of workers. The environmental risk posed by small amount of

seed leaving sites in workers' boots is negligible (probably less than 1 in a million chance of GM canola establishing and crossing with wild weedy relatives).

ANIMALS

Individual canola seeds are released from the plant as the seed pods dry out and open (dehisce or shatter). Due to the significant number and small size of canola seed, individual seeds may be transferred by small to large vectors. (e.g. ants, birds, grazing animals or humans). Birds, such as cockatoos and sparrows can shred or remove pods during development and maturity (Stanley & Marcroft 1999). Mice can also climb plants and feed on the seeds and pods, or feed on ungerminated seed sown close to the surface.

The viability of canola seed after passing through the digestive gut of animals is poorly understood. Anecdotal evidence from Canada suggests canola seed (Roundup Resistant® canola), mixed with wheat, remained viable and subsequently emerged after being fed to chickens and distributed as chicken manure spread on a field 12 months later (Martens 2001). This suggests that ingestion of canola seed by animals may have little effect on its viability. Further research is needed to elucidate this issue.

COMMERCIALLY RELEASED GM CANOLA

INTERNATIONALLY

Glufosinate tolerant hybrid canola, including the male sterile and fertility restorer parent lines:

Canada: The Plant Biosafety Office of the Canadian Food Inspection Agency approved these canola for environmental release in 1996, for use in feed in 1996 and in food in 1997;

The United States: The US Department of Agriculture and the Food and Drug Administration approved these canola for use in food and animal feed in 1998 and for environmental release in 1999;

Japan: The Japanese Ministries of Agriculture, Forestry and Fisheries, and Health and Welfare approved these canola for use in food in 1997, for use in feed and for environmental release in 1998;

Australia: The Australian and New Zealand Food Authority approved the use of hybrid glufosinate tolerant canola for use in food in 2001;

Europe: The environmental safety and food safety of glufosinate tolerant male sterile, fertility restorer and the hybrid lines are currently being assessed for by regulators in Europe.

Glyphosate tolerant (Roundup Ready® canola)

Canada: Health Canada approved this canola for use in food in 1994 and the Plant Biosafety Office of the Canadian Food Inspection Agency approved this canola for environmental release and use in feed in 1995;

The United States: The US Department of Agriculture and the Food and Drug Administration approved this canola for use in food and animal feed in 1995 and for environmental release in 1999;

Japan: The Japanese Ministries of Agriculture, Forestry and Fisheries and Health and Welfare approved this canola for environmental release and for use in food and feed in 1996;

Mexico: The Mexican Secretary for Agriculture, Livestock and Rural Development and Secretary for Health approved this canola for use in feed and food in 1996;

UK: The Ministry of Agriculture, Forestry and Fisheries approved this canola for oil in 1996;

Australia: The Australian and New Zealand Food Authority have recently approved the use of Roundup Ready® canola for use in food in 2000.

Europe: The environmental safety and food safety of glyphosate tolerant canola is currently being assessed for by regulators in Europe.

Bromoxynil tolerant canola

Canada: The Plant Biosafety Office of the Canadian Food Inspection Agency approved this canola for environmental release and for use in feed in 1997, for use in food in 1997;.

Japan: The Ministry of Health, Labour and Welfare approved this canola for environmental release in 1997, for use in feed in 1999 and for use in food in 2001;

Australia: The Australian and New Zealand Food Authority is currently assessing the use of this canola for food.

COMMERCIALLY RELEASED NON-GM HERBICIDE TOLERANT CANOLA

AUSTRALIA

Triazine tolerant canola

Despite penalties associated with lower grain yield and oil content, lower resistance to blackleg and persistence of triazine herbicides in the soil, there has been rapid adoption of triazine-tolerant (TT) canola in Australia. Triazine resistance in canola was bred by classical means by crossing a triazine resistant weedy relative (*B. rapa*) into canola (Beversdorf et al. 1980) and was commercially released into Australia in 1993. In 1999, triazine-tolerant (TT) canola accounted for almost 50 % of the

Australian crop. In the majority of cases, TT canola is chosen because the weeds (particularly Brassicaceae species) present cannot be controlled in the conventional varieties. In 1998, it was estimated that the areas of TT canola grown were 90 % in Western Australia because *R. raphanistrum* is a major problem, and 25 - 30 % in South Australia, Victoria and New South Wales (Lemerle et al. 1999).

TT canola represents a challenge to integrated weed control, especially in Western Australia, where there are concerns about widespread use of triazine (Group C) herbicides. Parts of Western Australia have a long history of triazine herbicide use, particularly in lupins and there is already evidence of atrazine resistant annual ryegrass (*Lolium rigidum*) and triazine resistance in wild radish.

Imidizalinone tolerant canola

Imidizalinone tolerant (IT) canola was first commercially available in Australia in 2000. IT canola has been conventionally bred and has been developed through somaclonal variation. These varieties are marketed along with an imidazolinone herbicide mix (Group B) called 'On Duty', which has a wide spectrum of activity. Unlike the TT varieties, the IT varieties carry no yield or oil penalties. Group B herbicides are frequently used in cropping sequences and are high risk in terms of development of herbicide resistance. Group B resistance has emerged in some areas of Western Australia so that the use of IT canola varieties is limited.

INTERNATIONALLY

Triazine tolerant canola

Triazine tolerant canola is commercially grown in Canada and U.S.A.

Imidizolinone tolerant canola

Imidizolinone tolerant canola is commercially grown in Canada and U.S.A.

SUMMARY

Brassica napus is an exotic species, originally cultivated in Asia and the Mediterranean. Canola refers to *B. napus* varieties which have been breed for low levels of two toxicants, erucic acid and glucosinates. It is cultivated for its high quality edible oil used in many foods (e.g. margarines) and seed meal used as animal feed.

Canola has been grown in Australia since 1969. Canola is grown throughout the grain belt of southern Australia. Production has significantly increased in Australia over the last decade and 1.4 Mha were sown to canola in 2000.

Spring varieties of canola are sown in Australia. These varieties have a shorter growing season than winter varieties which are grown in Europe. Spring varieties do not have a vernalisation requirement. Most canola crops in Australia are grown over winter and harvested in late spring. A small area of canola is grown over summer and harvested in early autumn. These areas experience milder temperatures and reliable rainfall (or irrigated) over summer.

Canola is one of the most profitable crops available to grain growers in southern Australia. It provides a valuable disease break between cereal crops and also has biofumigation properties against soil fungi. Canola has a range of pests in Australia including insects, pathogens and viruses and weeds. Brassicaceous weeds are particularly deleterious as there are limited herbicide options to control these plants within a canola crop. There has been a high rate of adoption by farmers of non-GM herbicide tolerant canola varieties to address this problem, despite yield and oil content penalties.

Despite a high level of self fertilisation, outcrossing rates between 12 – 47 % can also occur. Canola pollen can be dispersed by both wind and insects. The majority of wind dispersed pollen travels less than 10 m but longer distance transfer can occur under strong winds. Honeybees, where they occur, play a major role in the transfer of pollen over long distances. While most foraging occurs between 1 and 5 m, bees can move pollen up to 4 km. The frequency of outcrossing from canola to other related species decreases as distance increases. In Australia, outcrossing rates of 0.15 % to less than 0.01 % have been detected between commercial canola crops from 0 to 3 km. These distances are variable depending on local conditions (e.g. wind direction, wind speed), experimental design (e.g. size and orientation of plots) and insect movements.

Large seedbanks of canola seed can persist for many years due to high amounts of seed losses before and during harvest. Canola seeds become dormant after long exposure to darkness and water stress. Seeds can persist in undisturbed soils for up to 10 years or more, and up to 5 years in disturbed soils.

In Australia and Canada, canola is a problem weed in agricultural areas due to high seed losses and a persistent seedbank. Canola is also a plant which occurs in disturbed habitats such as roadsides, railway verges and field margins in areas where canola is grown. However canola does not occur as a weed in undisturbed natural habitats.

Outcrossing levels of 10-50 % can occur between canola crops. Outcrossing between canola and other *Brassica* species (e.g. *B. rapa*) can occur but at a lower frequency due to lesser genetic compatibility. *Brassica rapa*, is an obligate outcrosser and as a result, the frequency of outcrossing increases with smaller numbers of *B. rapa* plants in canola crops.

The frequency of outcrossing between important Brassicaceous weeds in Australia, *Raphanus raphanistrum* (wild radish), *Hirschfeldia incana* (Buchan weed) and *Sinapis arvensis* (charlock) is extremely low. Outcrossing rates vary but generally the frequency of outcrossing between canola and wild radish is greater from wild radish to canola. While a proportion of this seed will be collected at harvest, a proportion will remain on site and emerge in the following crop. Routine farm management practices will effectively control these plants. Hybrids between Buchan weed and canola are extremely low and hybridisation between canola and charlock has not been detected with canola as the pollinator. There have been no documented occurrences of gene introgression between canola and the Brassicaceous weeds *H. incana* and *S.*

arvensis, and no gene introgression has been measured beyond 4 generations between canola and *R. raphanistrum*.

Seed movement is also an important factor in spatial and temporal gene movement. Possible mechanisms of seed transport include, wind, water, vehicles, humans and animals.

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