Effect of *Brassica* genotype on the infestation by cabbage stem weevil *Ceutorhynchus pallidactylus* (Mrsh.) (Col.: Curculionidae) and the parasitism of stem weevil larvae

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„Für euch, Kinder der Wissenschaft und der Weisheit, haben wir dieses geschrieben. Erforschet das Buch und suchet euch unsere Ansicht zusammen, die wir verstreut und an mehreren Orten dargetan haben; was euch an einem Orte verborgen bleibt, das haben wir an einem anderen offen gelegt, damit es fassbar werde für eure Weisheit.“

Heinrich Cornelius Agrippa von Nettesheim

De occulta philosophia 3, 65

English translation: “We have written this for you, children of science and wisdom. Explore the sermons and search for our minds we have expounded in many places inside. What is hidden from you in one passage, we have offered you in another, to conceive it for your wisdom“.

Dedicated to my great-grandmother, Alwine...

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Chapter I - General introduction

Oilseed rape (*Brassica napus* L. *var. oleifera* Metzg.) (*Brassicaceae*) is the third most widely grown crop in the European Union (FAO 2005). In Germany the acreage grown in 2008/09 was ca. 1.42 Mill. ha (UFOP 2008). The area of oilseed rape continues to increase due to the increasing demand for plant oils in food (e.g. edible oil) and non-food sectors (e.g. lubrication-purpose, bio energy and bio fuels) (Winter 2004). With the recent extension of the planted area in the last twenty years, damage by diseases and pests of oilseed rape has become more severe (Lamb 1989; Alford *et al*. 2003). One of the most important limiting factors in the production of *Brassica* oilseeds is the complex of insect pests associated with these plants (Ekbom 1995). A succession of pests attacks the crop from its early seedling stage in autumn to the beginning of pod ripening in the following summer. Winfield (1992) has specified two important groups of insect pests infesting in winter oilseed rape in European countries, namely (i) pests attacking the crop in its vegetative phase, i.e. slugs, cabbage root fly, cabbage stem flea beetle, several species of stem miners and wood pigeons, and (ii) pests attacking the flower buds and seed pods, i.e. pollen beetle, cabbage seedpod weevil and brassica pod midge. Damage of each specialist pest species is mostly restricted to a particular plant organ of oilseed rape (Kirk 1992). Among pests infesting the vegetative growth stage of oilseed rape, the cabbage stem weevil, *Ceutorhynchus pallidactylus* (Marsham 1802), syn: *C. quadridens* (Coleoptera: Curculionidae)) is widely distributed on crops of oilseed rape in Central and Northern Europe. In many countries, *C. pallidactylus* is estimated to be an important pest on oilseed rape, e.g. Austria (Berger 1991), Czech Republic (Kazda 1957; Šedevý & Vašák 2002), Finland (Kangas 1976), France (Lerin 1995), Poland (Dmoch 1958; Pałosz 1978; Kelm & Walczak 1998), Slovakian Republic (Holecová *et al.* 2005), Switzerland (Büchi 1990) and the United Kingdom (Winfield 1961a; Graham & Gould 1980; Ferguson *et al.* 2006).

The main objectives of this study are:

1. to develop laboratory screening methods for identification and quantification of host plant quality and resistance in brassicaceous species to cabbage stem weevil (*Ceutorhynchus pallidactylus*).

2. to screen a large assortment of cultivars of oilseed rape, breeding lines, resynthesized oilseed rape lines and other species of Brassicaceae under controlled conditions in growth chambers
3. to validate the resistance of selected genotypes under field conditions
4. to investigate the parasitism of stem-boring ceutorynchid larvae in various cultivars of *Brassica* spp.

The following chapter will present an overview about the state of the art.

Host selection of oilseed rape pests

Herbivorous insect species commonly accept only a limited number of plant species as hosts (van Loon *et al*. 1992), and location and selection of host plants is an elementary step in the insects` life cycle. Many studies were carried out to identify cues involved in host finding of specialist and generalist insects (Singh & Ellis 1993). Schoonhoven *et al*. (1998) have divided the host selection behaviour into five specific phases: searching, selection, acceptance, preference and finally recognition. Two major groups of stimulants are important for host finding: visual and olfactory cues (Hawkes *et al*. 1978; Schoonhoven *et al*. 1998). Visual cues are related to plant morphology, colour, leaf structure, while chemical cues like plant volatiles or phagostimulants can be influenced by various factors, e.g. wounding or physiological status of the plant (Blaakmeer *et al*. 1994; Schoonhoven *et al*. 1998). Plant metabolites are widely considered to be mediators in insect orientation to host plants (Rask *et al*. 2000; Pontoppidan *et al*. 2003), and their acceptance as a resource for feeding or oviposition (Lamb 1989; Schoonhoven *et al*. 1998).

For host finding and acceptance, the specialist insect pests of *Brassica* oilseeds rely on chemical compounds, characteristic for the family *Brassicaceae*: the glucosinolates (GSLs) (Ekbom 1995). Approximately 120 different GSLs have been detected so far (Fahey 2001). They are stored in plant tissue and hydrolyzed particularly by the enzyme myrosinase into breakdown products like isothiocyanate, hydrogen sulphate and D-glucose (Ekbom 1995).

The total amount and composition of GSLs within plants depends on several factors: genetic, environmental, agronomic or on influence of insect attack (Fenwick 1983; Koritsas *et al*. 1991; Bartlet *et al*. 1999a; Wallsgrove *et al*. 1999). The total amount of

GSL is usually not closely related to single GSL compounds or their distribution within plant organs (Fieldsend & Milford 1994; Yungchang *et al.* 1999). Further, in many studies no correlation was found between the GSL content in the seeds and in the vegetative tissue (Wallsgrove *et al*. 1999).

The GSL profile within plants can have substantial impact on host plant quality for specialist insects. This highly specific system is generally accepted to be a part of the plant's defence against generalist pests. GSLs are known as anti-feedants for many polyphagous herbivores. In contrast, insect species, which are adapted on Brassicaceae, use GSLs or their fission products as feeding stimulants or attractants (Lamb 1989; Pontoppidan *et al*. 2003), which is an effect of co-evolution between host plant and insect (Rask *et al.* 2000).

Interactions between the content of GSLs in Brassicaceae and the behaviour of pests have been studied extensively in recent years (Rask *et al.* 2000; Liblikas *et al.* 2003) with particular emphasis on insect attraction and feeding. In plants injured by the cabbage stem flea beetle, *Psylliodes chrysocephala* L., a significant increase of the indolic GSL glucobrassicin and neoglucobrassicin was demonstrated (Koritsas *et al*. 1991). Both GSLs have been identified as an attractant for *P. chrysocephala* (Wallsgrove *et al*. 1999). In addition, GSLs also act as feeding stimulants (Giamoustaris & Mithen 1995). Feeding by *P. chrysocephala* increased with increasing levels of GSLs. The large white butterfly, *Pieris brassicae* L., uses the GSL gluconasturtiin as a recognition cue for location of host plants (Huang & Renwick 1994) and feeding of larvae (Miles *et al*. 2005). Further, glucobrassicin serves as an attractant for oviposition (Van Loon *et al*. 1992). GSLs provide also important cues for flea beetles on *Brassica* spp., *Phyllotreta cruciferae* (Goeze) and *P. chrysocephala*, (Lamdon *et al*. 1998; Liblikas *et al*. 2003; Henderson *et al*. 2004), cabbage root fly, *Delia radicum* (L.), (Hardman & Ellis 1978), cabbage seedpod weevil, *Ceutorhynchus obstrictus* (Marsham), (McCaffrey *et al.* 1999; Ulmer & Dosdall 2006) and brassica pod midge, *Dasineura brassica* Winn., (Bartlett *et al.* 1999b).

The aim of various studies was to alter the host-herbivore-interaction by modifying the GSL content in host plants (Åhman 1982; Larsen *et al*. 1983). However, insect feeding responses to cultivars of low- and high-GSL content did not show consistent differences (Weber *et al*. 1986; Mithen 1992; Giamoustaris & Mithen 1995; Bartlet 1996). In laboratory experiments, pollen beetle, *Meligethes aeneus* (Fabr.), was less attracted to low-GSL cultivars compared with cultivars containing a high content of GSL (Cook *et al.* 2003).

Mechanisms of host plant resistance to oilseed rape pests

Host plant resistance to pest insects has been generally sub-divided in antixenosis and antibiosis (Painter 1951; Kogan & Ortmann 1978). 'Antixenosis' refers to the inability of a plant to be an attractant host for herbivorous insects, influencing them to select other plants (Kogan & Ortmann 1978). 'Antibiosis' is characterized as plant factors affecting the insect life history and performance negatively (survival, growth or fecundity). Both, antixenosis and antibiosis are mainly caused by morphological or chemical plant parameters (Kogan & Ortmann 1978). In the past, resistant cultivars have been developed in various crop species against various pests, e.g. aphids (Ellis & Farrell 1995; Messina & Bloxham 2004), stem and root borers (Reay-Jones *et al.* 2003; Murray *et al.* 2007) and lepidopteran species (Stoner 1990; Sarfraz *et al.* 2007).

Brassica plants have developed different resistant reactions to insect attack, which can be constitutive or induced by the insect itself (Meiners & Hilker 2002). Constitutive defense mechanisms are often caused by plant morphology (Way & Murdie 1965; Schoonhoven *et al.* 1998; Stoner 1990) or secondary metabolite compounds (Giamoustaris & Mithen 1997; Lambdon *et al.* 1998). Induced plant resistance is commonly based on different mechanisms. A specific defensive reaction of *Brassica* host plants has been reported with cabbage stem weevil (Pałosz 1980; Nuss 2004). As a response to oviposition into petioles, extensive wound callus tissue grows under the egg batch, thereby pushing the eggs to the surface (Broschewitz 1985). The percentage of egg batches destroyed by callus formation has been reported to vary between 40% (Broschewitz & Daebler 1987) and more than 70% (Pałosz 1980). Kazda (1953) presumed that the production of callus tissue could be increased by oviposition during early growth stages and high humidity conditions. The role of feeding or semiochemicals in activating this physiological reaction is still unknown.

Isothiocyanates have been determined as attractants and feeding stimulants for many brassicaceous pests (Koritsas *et al.* 1991; Giamoustaris & Mithen 1995). The development of cultivars that produce only low amounts of specific isothiocyanates might change the attraction of specialist insects to their host plants (Cook *et al.* 2003; Williams 2004). The stem weevils, *C. pallidactylus* and *C. napi*, were attracted to yellow water traps at significantly higher numbers when 2-phenylethyl isothiocyanate was used as a bait substance (Walczak *et al*. 1998). Specific parasitoids were attracted by this isothiocyanate as well (Bradburne & Mithen 2000; Ulber & Wedemeyer 2006).

Screening methods for host plant resistance

A large number of laboratory screening methods have been developed for testing the susceptibility of different host plants to pest species (Farrell 1977; Harmon & McCaffrey 1997; Ulmer *et al*. 2001). The basis for the identification of (partial) resistance in plants is the screening of a large assortment of genetically diverse genotypes in bioassays. The identification of resistant genotypes requires simple, rapid and reliable techniques for the evaluation of pest performance (Ellis & Kift 2003). Consequently, the selection of parameters which can be used as indicators for the host quality of the plant is the first step of developing a new screening method (Farrell 1977). Screening methods should allow preferably discrimination between different types of resistance, namely antixenosis and antibiosis.

The standardization of plant material grown under greenhouse conditions rather than the standardization of test insects is of great importance in achieving reliable results (Harmon & McCaffrey 1997). Pests (e.g. aphids or mites) are often reared in the laboratory quite successfully, so the standardization of larval instar or nutritional status is possible. Specialist pests which prefer single plant organs or growth stages for reproduction are often more reluctant to permanent rearing in the laboratory, especially when they are univoltine or rely on winter diapauses (Broschewitz 1985; Barari *et al*. 2006).

In general, there are two basic methods that can be applied to test host plant quality for herbivorous insects: (i) multi-choice tests and (ii) no-choice tests (Farrell 1977; Ulmer *et al*. 2001). In no-choice tests, only one individual plant or detached plant part is exposed to the insects, whereas in choice-tests the insects can select between two or more different plant genotypes. Every method has it specifications. Multiple-choice tests allow better comparisons between different genotypes at the same time (Bartlet & Williams 1991), but they do not represent the field situation, where only one cultivar of a crop species is available to colonization by the pest. No-choice tests are often easier to manage (Risch 1985), but the exclusion of choice-factors can deform results in a way, that only extreme responses of the insect can be detected (Farell 1977). Furthermore, no-choice test can customize insects to a plant, which may not be a common host. The design of a screening method may have significant effects on results (Risch 1985). For example, screening by using detached leaves or racemes of brassicaceous plant species induces breakdown of GSLs into various products (Blaakmeer *et al*. 1994), which might act as stimulant cues for specialist pests (Pontoppidan *et al*. 2003). Generalists are considered to be less affected by the screening design than highly specialist species (Risch 1985). Biochemical assays can apply for detecting alterations of primary or secondary plant metabolites (Farell 1977).

Results from laboratory screening experiments need to be validated under field conditions for confirmation. Chemical cues and stimulants which can influence many phases in host searching and host acceptance, especially when volatile odors are involved, might be inhibited by the screening design in the laboratory.

In the past, various screening methods have been developed for testing plant resistance to pest insects of *Brassica* spp. in the laboratory (Bartlett & Williams 1991; Harmon & McCaffrey 1997; Sarfraz *et al*. 2007). However, methods for screening the resistance to cabbage stem weevil, *C. pallidactylus*, were not available at the beginning of this research work.

***Life cycle and phenology of* C. pallidactylus**

C. pallidactylus is an univoltine stem miner about 3 mm long and brown to black in colour with a patch of white scales in the centre of the elytra (Fig 1). Adult *C. pallidactylus* migrate from their overwintering habitats (under leaf litter at the edges of woods or shrubs) into crops of oilseed in March/April (Günthart 1949; Alford *et al.* 2003). First flights commonly occur at temperatures > 12 °C and low wind velocity < 3m/s (Friesland 1990; Kjær-Pedersen 1992; Johnen & Meier 2000). Females feed on leaves and shoots up to 12 days for ovary maturation (Broschewitz 1985). During March to May they deposit batches of 4-6 translucent eggs (0.4mm by 0.6mm) (Winfield 1992) into small pockets bitten into the parenchyma of petioles and midribs of leaves (Ferguson *et al.* 2003; Barari *et al.* 2005).

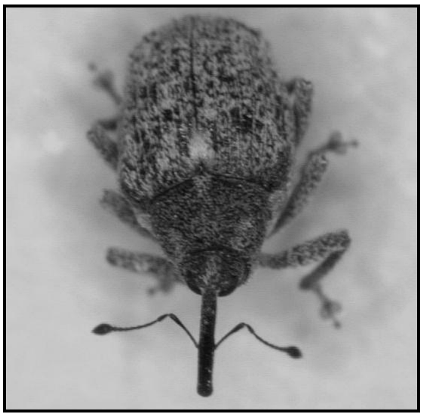

Fig 1: Adult cabbage stem weevil (courtesy by the author)

The total number of eggs laid per female was estimated at 60-70 eggs in the field (Broschewitz 1985) and up to 280 eggs in the laboratory (Körting 1942). Larvae are coloured white to yellowish and have a brown head capsule (Fig 2). During the first and second instar, they feed within the pith of petioles. Third instar larvae bore into the main stems of plants (Broschewitz 1985; Barari *et al*. 2005).

Fig 2: Larva of cabbage stem weevil (courtesy by the author)

In June/July, mature third instar larvae leave the stem through exit holes, to pupate 2-3cm deep in the soil (Broschewitz & Daebler 1987; Barari *et al*. 2005). Depending on weather conditions pupation takes approximately 20-30 days (Broschewitz 1985). Adult weevils emerge from soil at the end of June or beginning of July. After feeding on wild and cultivated Brassicaceae or perennial oilseed rape for four weeks, they migrate to their overwintering sites (Broschewitz & Daebler 1987; Williams 2004) (Fig 3).

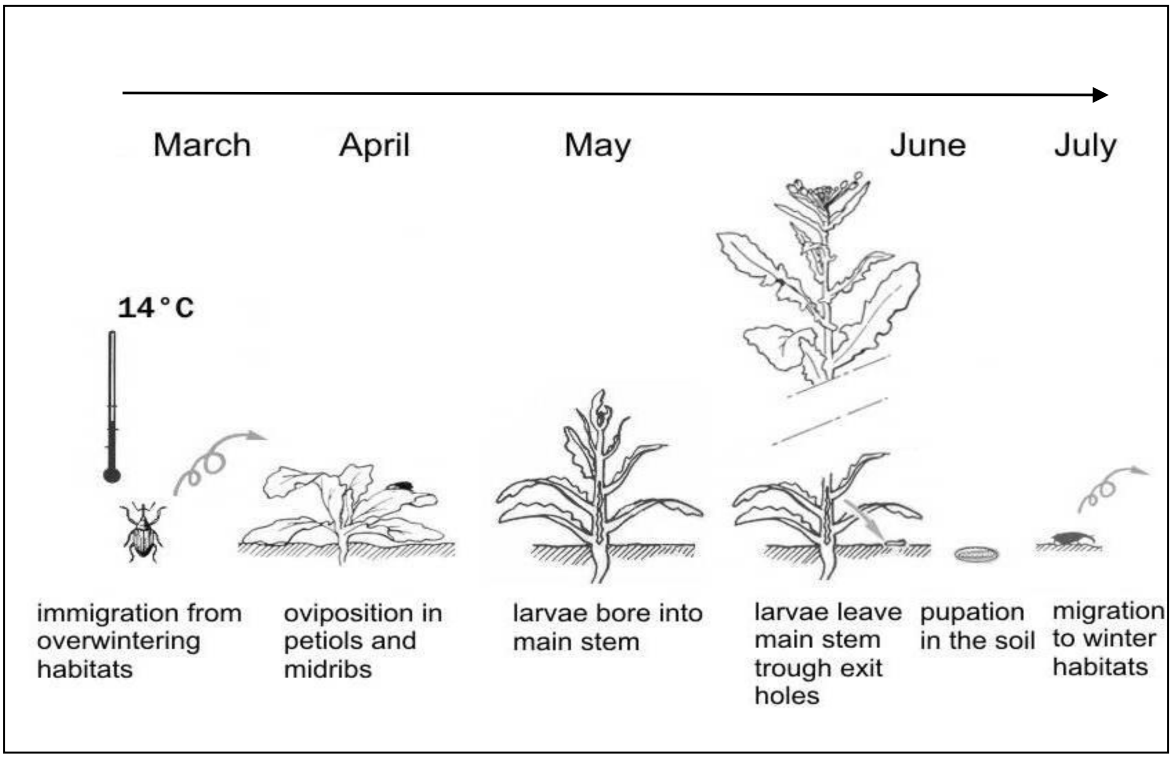

Fig 2: Lifecycle of *C. pallidactylus* (modified after CETIOM 1985)

Host plant range and damage potential of *C. pallidactylus*

The cabbage stem weevil can exploit a large number of host plants of Brassicaceae (Speyer 1921; Günthart 1949; Tommes 1998). Adults have been reported to feed on cultivated and wild relatives of oilseed rape, *B. napus* L. *var. oleifera* Metzg., turnip rape, *Brassica rapa* L., cabbage, *Brassica oleracea* L., black mustard, *B. nigra* (L.), Indian mustard, *B. juncea* (L.), radish, *Raphanus* spp., white mustard, *Sinapis alba* (L.), sea-kale, *Crambe maritime* L., and several wild brassicaceous species.

The pest status and plant damage of *C. pallidactylus* has been studied extensively in spring and winter oilseed rape and vegetable *Brassicas* (Günthart 1949; Winfield 1961a; Broschewitz 1985). The damage potential in oilseed rape has been assessed controversially in the literature. Winfield (1961a) and Graham & Gould (1980) supposed that yield losses by *C. pallidactylus* in the UK only occur in spring rape and came to the conclusion, that *C. pallidactylus* is unimportant for winter oilseed rape. Yield reductions by *C. pallidactylus* on winter oilseed rape were also not detected by Lehman (1965). In Poland, economic damage by *C. pallidactylus* only occurred in winter crops of low density with weak and smaller plants (Pałosz 1980). In contrast, Broschewitz (1985) found that winter rape crops of high plant densities are more susceptible and suffer from increased larval infestation, due to thinner main stems and a higher fecundity of females. At low plant density and especially with cultivars producing a high biomass, the infested plants have higher compensation ability (Broschewitz & Daebler 1987). In Slesvig-Holstein, Landschreiber (2005) reported on yield losses up to 20%. Kelm & Walzak (1998) found a positive correlation between the length of the larval feeding tunnel and shedding of pods; they assumed that dry stress of plants might increase damage by *C. pallidactylus*. Yield effects by feeding of larvae can be compensated by oilseed rape plants at early growth stages (Kelm & Klukowski 2000). Damage by adult cabbage stem weevils feeding on leaves has never been observed (Broschewitz 1985).

While the direct damage of *C. pallidactylus* larvae by feeding within petioles and stems varies between fields and locations, indirect damage of larvae by causing wounds and ports for entry stem canker (*Leptosphaeria maculans* (Desm.) Ces. and Not. [anamorph: *Phoma lingam* (Tode ex Fr. Desm.]) is more important; it increases early colonization and fast growth of the pathogen within stems (Lücke & Pluschkell 1982; Broschewitz *et al*. 1993). Pałosz *et al*. (1994) found a positive correlation between infestation by *C. pallidactylus* and the severity stem cancer. Insecticide applications for control of cabbage stem weevil reduced the incidence of *Leptosphaeria* stem cancer and increased yield (Hartleb *et al*. 1998). Interactions between *C. pallidactylus* and other pest species using the same ecological niche have been reported for rape stem weevil, *Ceutorhynchus napi*, (Dechert & Ulber 2004), and cabbage stem flee beetle, *Psylliodes chrysocephala* (Ferguson *et al*. 2006).

Strategies for control of *C. pallidactylus*

Application of insecticides is the most commonly used measure for control of *C. pallidactylus* as other important pests on oilseed rape (Hartleb *et al*. 1998; Buntin 1999; Williams 2004; Seta & Wolski 2006). With two insecticide applications in spring, percentage infestation of cabbage stem weevil significantly decreased (Wahmhoff 2000). As the pest invades the oilseed rape crop in irregular succession, exact timing is crucial and single applications are often little effective (Free & Williams 1979). Yellow water traps monitor migration flights of adults in spring (Garbe *et al*. 1996) and help to detect the ideal date for application. Spraying thresholds are based today on yellow water trap catches (Williams 2004). Computer-based decision support systems, e.g. PRO_PLANT, are used to forecast the phenology in the crop (Johnen & Meier 2000).

Synthetic pyrethroids are the predominant insecticides applied on oilseed rape. However, this group of insecticides is non-selective for non-target insects, such as predators or parasitoids (Ekbom 1995; Williams 2004), being a natural source of potential benefit. Further frequent usage of pyrethroids can result in insecticide resistance. Resistance of pollen beetle against pyrethroids have been reported all over Europe (Hansen 2003; Derron *et al.* 2004; Heimbach *et al*. 2006; Wegorek *et al.* 2006); there are first indications for similar effects on *C. pallidactylus*. The risk of insecticide resistance places greater emphasis on searching for alternative control methods.

An extensive overview on non-chemical methods of pest management in oilseed rape is given by Williams (2004). Several cultivation techniques can contribute to Integrated Pest Management (IPM) (Williams 2004). A significantly higher reproduction rate of *C. pallidactylus* was found in conventional farmed, ploughed fields than in organic conservation tillage farming (Büchs & Katzur 2004). Only insignificant reductions of the larval abundance of *C. pallidactylus* were obtained by using mixed cropping systems of oilseed rape with cereals or legumes (Paulsen *et al.* 2006). A higher numbers of parasitoids in fields with non-inversion tillage is reported (Nitzsche & Ulber 1998). Plant architecture and development can influence pest abundance (Büchs & Katzur 2004; Williams 2004). Infestation by cabbage stem weevil increases as plant density decreases because a higher number of leaves and larger leaf size at low plant density increases oviposition (Nuss & Ulber 2007). Generally, hybrid cultivars are considered to have higher compensation ability to pest damage by growing more vigorously (Lamb 1989).

Conservation biological control of *C. pallidactylus* can provide substantial natural control. The larvae are attacked by five endoparasitoids (Alford *et al*. 2003). In Germany and other Central European countries, the most common species is *Tersilochus obscurator* Aubert (Hym.; Ichneumonidae) (Klingenberg & Ulber 1994, Ulber & Nitzsche 2006). Females parasitize host larvae, while these are feeding within petioles. They oviposite through the plant tissue and lay a single egg into pest larvae. The level of parasitism of *C. pallidactylus* by *T. obscurator* ranges between 18 and 52% (Nissen 1997, Ulber 2003); it depends on several external factors, e.g. climate (Johnen *et al*. 2006), stem-base diameter (Ulber & Fischer 2006), plant volatiles (Jönsson *et al.*

2005) and crop management practices (Klingenberg & Ulber 1994; Nitzsche & Ulber 1998). Further, predators like Carabidae and Staphylinidae can play an important role (Piper & Williams 2004; Schlein & Büchs 2004; Büchs *et al*. 2006). They feed on *C. pallidactylus* larvae when these are leaving the plant for pupation in soil. In addition, entomophagous larvae of the muscid fly, *Phaenoia trimaculata* Bouche, feed on the larvae within stems (Fritzsche 1955; Lehmann 1965). Female *P. trimaculata* invades the split stems of oilseed rape to oviposite into the damaged pith. The abundance of stem mining larvae can be reduced up to 55% (Fritzsche 1955).

More recently, the so-called 'Push-Pull-strategy' has been developed in the UK as a new component of IPM in oilseed rape (Cook *et al*. 2003; Williams 2004). It utilises the behavioural responses of pests and parasitoids to plant- and insect-derived semiochemicals (Cook *et al*. 2007a). Repellent compounds and/or resistant cultivars are used to 'push' the pests from the main crops, and attractants or more attractive plants are used to 'pull' insects to trap crop areas, e.g. turnip rape (Potting *et al*. 2005), where they can be killed selectively by insecticides (Barari *et al*. 2005) or entomopathogenic fungi (Butt *et al*. 1998). At present, the development of the 'Push-Pull- strategy' is in progress (Cook *et al.* 2007b).

A new approach to IPM is the development of genetically modified organisms. Two types of transgenic insect-resistant *Brassica* have been developed (Earle *et al*. 2004): expressing *Bacillus thuringiensis* genes (Xue-bao *et al*. 1995) and the expression of proteinase inhibitor genes (Girard *et al*. 1998). The impact of these unspecific methods on *C. pallidactylus* has not been evaluated, so far. Additionally, a new method had been established in the laboratory for controlling pollen beetle by transforming oilseed rape with a pea lectin gene (Lehrmann 2007). The usage of transgenic insect-resistant *Brassica* crops as an instrument of IPM is not possible in Europe because of low public acceptance and legal guidelines (Davis 2006).

Host plant resistance to insect pests is an underutilized strategy in IPM (Eigenbrode & Trumble 1994). Classical crop breeding can play an important role for developing oilseed rape cultivars being resistant to insect pests, which could reduce the need for insecticide application (Williams 2004). Resistant lines have been developed so far against important diseases, e.g. *Phoma lingam, Alternaria brassicae* and *Sclerotinia sclerotiorum* (Manthey 2005). Screening for resistance in common breeding lines and cultivars has been initiated for various pests in other countries, e.g. aphids, *Lipaphis* *erysimi* Kalt., in Indian Mustard (Abraham & Bhatia 1994), root maggots, *Delia* ssp., (Dosdall *et al*. 1994; Jyoti *et al*. 2001) and for cabbage flea beetles, *Phyllotreta* sp., (Henderson *et al*. 2004). Resistant breeding lines against cabbage seedpod weevil have been developed by introgression (*S. alba* x *B. napus*) in Canadian oilseed rape (Kalischuk & Dosdall 2004; Dosdall & Kott 2006).

Modern cultivars and breeding lines of *Brassica napus* are characterized by a limited genetic diversity (Seyis *et al*. 2000) and they have little potential to provide sources of resistance for practical exploitation in breeding programs (Ellis 1992). Two genetic resources are of importance to enlarge the genetic variability: wild brassicaceous species and resynthesized oilseed rape ('resyntheses').

Wild species of *Brassica* have been screened extensively, resulting in specific resistance to specialist pests: *D. radicum* (Ellis *et al*. 1999; Jensen *et al*. 2002; Felkl *et al*. 2005), cabbage aphid, *Brevicoryne brassicae* L. (Cole 1994; Cole 1997), and cabbage white fly, *Aleyrodes proletella* L. (Ellis *et al.* 1996). The mechanisms of resistance are often unknown. Cole (1994) presumed a hyper-sensitive reaction in resistant plants to *B. brassicae*, causing a blockage within the stylet canal.

A large assortment of species belonging to the subtribe Brassicinae has been identified as potential sources for antixenosis, antibiosis or mixtures of both to pest insects, particularly *Brassica incana* Tenore, *B. spinescens* Pomel and *B. fruticulosa* Cyrillo (Cole 1997). However, because of the different genomes within the genus *Brassica*, hybridization between wild brassicaceous species is crucial (Ellis *et al*. 1999; Pink *et al*. 2003).

Recently, the development of resynthesized lines of oilseed rape has become a promising approach to enlarge the genetic divergence in plant breeding (Becker *et al*. 1995; Seyis *et al*. 2000; Becker 2000). The main objective is to re-design the ancestral combination between *Brassica oleracea* x *Brassica rapa* (U 1935) by using biological varieties of both diploid parent species (Becker *et al*. 1995). Resynthesized lines of oilseed rape represent a genetic resource with high diversity for breeding hybrid cultivars (Girke 2002). They provide a source for disease resistance, e.g. against *Phoma lingam* (Crouch *et al*. 1994), *Plasmodiophora brassica* (Diederichsen & Sacristan 1996) and *Verticillium longisporum* (Rygulla *et al.* 2007). Increased levels of resistance to oilseed rape pests can be expected as well (Seyis *et al*. 2000). However, negative interactions of crossing high yield cultivars with wild brassicaceous species are possible, e.g. yield reduction (Allard 1996).

So far there are very few in-depth studies to evaluate the level of resistance to *C. pallidactylus* in brassicaceous plants. Winfield (1961b) reported on reduced susceptibility of *Sinapis alba* L., and Way & Murdie (1965) found a higher infestation in cultivars of Brussels sprouts without waxy surface on the leaves. In field observations, single low cultivars of oilseed rape (high GSL content, no erucic acid in seeds) were preferred by *C. pallidactylus* (Mrówczyński 1992). Mrówczyński (1998) found significant differences between the infestation levels of modern double low lines (no erucic acid and low glucosinolate content in the seeds) in field trials. An extensive field screening of brassicaceous crops to pests was the EU-Projekt ‘RESGEN’ (Lühs *et al*. 2003), which tested the susceptibility of approximately 200 *Brassica* genotypes against cabbage stem flea beetle, rape stem weevil, and cabbage stem weevil in a one-year field experiment.

In this study, potential characteristics of resistance to cabbage stem weevil (*Ceutorhynchus pallidactylus*) have been evaluated for a wide spectrum of Brassicaceae (cultivars, breeding lines, varieties, resynthesized rapeseeds, other *Brassica* spp.), for the first time. For the screening of a large assortment with a wide genetic variability, new methods have been developed which enable a rapid and reliable selection of genotypes with (partial) resistance in climate chambers. In field trials, the susceptibility of *Brassica* genotypes to *C. pallidactylus* has to be evaluated to confirm results from laboratory experiments. Finally the study shall be a basis for introduction of oilseed rape with insect resistance in national breeding programs and utilization in agriculture practice.

The following chapters have been published or will be submitted under following titles:

🕮 Eickermann M & B Ulber (2010): Screening of oilseed rape and other brassicaceous genotypes for susceptibility to *Ceutorhynchus pallidactylus* (Mrsh.). Journal of Applied Entomology **134**, 542-550.

🕮 Eickermann M, Ulber B & S Vidal (2010): Resynthesized lines and cultivars of *Brassica napus* L. provide sources of resistance to the cabbage stem weevil

(*Ceutorhynchus pallidactylus* (Mrsh.)). Bulletin of Entomological Research. In press. DOI: 10.1017 / S0007485310000489. Published online: 24 November 2010.

🕮 Influence of different *Brassica* host plants to parasitism of ceutorhynchid stem weevils

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References

Abraham V & CR Bhatia (1994): Testing for tolerance to aphids in Indian mustard Brassica juncea (L.) Czern and Coss. Plant Breeding **112**, 260-263.

Allard, RW (1996): Genetic basis of the evolution of adaptedness in plants. Euphytica **92**, 1-11.

Åhman I (1982): A comparison between high and low glucosinolate cultivars of summer oilseed rape (*Brassica napus* L.) with regard to their levels of infestation by the brassica pod midge (*Dasineura brassicae* Winn.). Journal of Applied Entomology **94**, 103-109.

Alford DV, Nilsson C & B Ulber (2003): Insect pests of oilseed rape crops. In: DV Alford (ed), *Biocontrol of Oilseed Rape Pests*. Blackwell Science, Oxford, 9-41.

Barari H, Cook SM, Clark SJ & IH Williams (2005): Effect of a turnip rape (*Brassica rapa*) trap crop on stem-mining pests and their parasitoids in winter oilseed rape (*Brassica napus*). Biological Control **50**, 69-86.

Barari H, Cook SM & IH Williams (2006): Rearing and identification of the larval parasitoids of *Psylliodes chrysocephala* and *Ceutorhynchus pallidactylus* from field-colleted specimens. IOBC/wprs Bulletin **29** (7), 215-223.

Bartlet E & IH Williams (1991): Factors restricting the feeding of the cabbage stem flea beetle (*Psylliodes chrysocephala*). Entomologia Experimentalis et Applicata **60**, 233-238.

Bartlet E (1996): Chemical cues to host-plant selection by insect pests of oilseed rape. Agricultural Zoology Reviews **7**, 89-116.

Bartlet E, Williams IH & JA Pickett (1999a): The ideal glucosinolate profile for pest resistance in oilseed rape. IOBC/wprs Bulletin **22** (10), 13-17.

Bartlet E, Kiddle G, Williams I & R Wallsgrove (1999b): Wound-induced increases in the glucosinolate content of oilseed rape and their effect on subsequent herbivory by a crucifer specialist. Entomologia Experimentalis et Applicata **91**, 163-167.

Becker HC, Enqvist GM & B Karlsson (1995): Comparison of rapeseed cultivars and resynthesized lines based on allozyme and RFLP markers. Theoretic and Applied Genetics **91**, 62-67.

Becker HC (2000): Nutzung genetischer Ressourcen in der Pflanzenzüchtung. In: Hammer, K & T Gladis (eds), *Nutzung genetischer Ressourcen – ökologischer Wert der Biodiversität*. Schriften zu Genetischen Ressourcen **16**. ZADI, Bonn, 79-90.

Berger HK (1991): Oilseed rape in Austria – pest problems. IOBC/wprs Bulletin **14** (6), 14-20.

Blaakmeer A, Geervliet JBF, van Loon JJA, Posthumus MA, van Beek TA & Æ de Groot (1994): Comparative headspace analysis of cabbage plants damaged by two species of *Pieris* caterpillars: consequences for inflight host location by *Cotesia* parasitoids. Entomologia Experimentalis et Applicata **73**, 175-182.

Bradburne RP & R Mithen (2000): Glucosinolate genetics and the attraction of the aphid parasitoids *Diaeretiella rapae* to Brassica. Proceedings of the Royal Society, London B **267**, 89-95.

Broschewitz B (1985): Untersuchungen zur Biologie und Schadwirkung des Gefleckten Kohltriebrüsslers (*Ceutorhynchus quadridens* PANZER) am Winterraps (*Brassica napus* L. *var. oleifera* Metzg.). PhD-thesis, Wilhelm-Pieck-Universität, Rostock.

Broschewitz B & F Daebler (1987): Beitrag zur Biologie und Schadwirkung des Gefleckten Kohltriebrüsslers (*Ceutorhynchus quadridens* PANZ.) an Winterraps. Nachrichtenblatt für den Pflanzenschutzdienst in der DDR **41**, 34-37.

Broschewitz B, Steinbach P & S Goltermann (1993): Einfluss stängelbewohnender tierischer Schaderreger auf den Befall von Winterraps mit *Phoma lingam* und *Botrytis cinerea*. Gesunde Pflanzen **45**, 106-110.

Büchi R (1990): Investigations on the use of turnip rape as trap crop plant to control oilseed rape pests. IOBC/wprs Bulletin **13** (4), 32-39.

Büchs W & K Katzur (2004): Means to control pests in organic oilseed rape production. IOBC/wprs Bulletin **27** (10), 225-238.

Büchs W, Felsmann DS, Klukowski Z, Luik A, Nilsson C, Ahmed B, Ulber B & IH Williams (2006): Key predator species in oilseed rape crops - a review of literature and MASTER results. CD-Rom Proceedings of the International Symposium "Integrated Pest Management in Oilseed Rape Pests", Goettingen, Germany, 3-5 April 2006.

Buntin GD (1999): Damage loss assessment and control of the cabbage seedpod weevil (Coleoptera: Curculionidae) in winter canola using insecticides. Journal of Economic Entomology **92**, 220-227.

Butt TM, Carreck NL, Ibrahim L & IH Williams (1998): Honeybee-mediated infection of pollen beetle (*Meligethes aeneus* Fab.) by the insect-pathogenic fungus, *Metarhizium anisopliae*. Biochemical Science and Technology **8**, 533-538.

CETIOM (1985): Cahiers techniques. Insectes et autres ravageurs – Colza d'hiver. Creation CETIOM, Paris.

Cole RA (1994): Locating a resistance mechanism to the cabbage aphid in two wild Brassicas. Entomologia Experimentalis et Applicata **71**, 23-31.

Cole RA (1997): The relative importance of glucosinolates and amino acids to the development of two aphid pests Brevicoryne brassicae and Myzus persicae on wild and cultivated brassica species. Entomologia Experimentalis et Applicata **85**, 121-133.

Cook SM, Smart LE, Rasmussen HB, Bartlet E, Martin JL, Murray DA, Watts NP and IH Williams (2003): Push-pull strategies to reduce insecticide input to oilseed rape (*Brassica napus*): Potential of low alkenyl glucosinolate oilseed rape varieties (push!), and turnip rape (*Brassica rapa*) trap crops (pull!). Proceedings of the 11th International Rapeseed Congress, Copenhagen, 6-10 July 2003, **3**, 1018-1021.

Cook SM, Khan, ZR & JA Pickett (2007a): The use of push-pull strategies in integrated pest management. Annual Review of Entomology **52**, 375-400.

Cook SM, Jönsson M, Skellern MP, Murray DA, Anderson P & W Powell (2007b): Responses of *Phradis* parasitoids to volatiles of lavender, *Lavendula angustifolia* — a possible repellent for their host, *Meligethes aeneus*. Biological Control **52**, 591-598.

Crouch JH, Lewis BG & RF Mithen (1994): The effect of a genom substitution on the resistance of *Brassica napus* to infection by *Leptosphaeria maculans*. Plant Breeding **112**, 265-278.

Davis LC (2006): Genetic engineering, ecosystem change, and agriculture: an update. Biotechnology and Molecular Biology Review **1**, 87-102.

Dechert G & B Ulber (2004): Interactions between the stem-mining weevils Ceutorhynchus napi Gyll. and Ceutorhynchus pallidactylus (Mrsh.) (Coleoptera: Curculionidae) in oilseed rape. Agricultural and Forest Entomology **6**, 193-198.

Derron JO, LeClech E, Bezençon N & G Goy (2004): Résistance des méligéthes du colza aux pyréthrinoïdes dans le basin lémanique. Revue Suisee d' Agriculture **36**, 237-242.

Diederichsen E & MD Sacristan (1996): Disease response of resynthesized *Brassica napus* L. lines carrying different combinations of resistance to *Plasmodiophora brassicae* Wor.. Plant Breeding **115**, 5-10.

Dmoch J (1958): Wstępne obserwacje nad biologią chowaczy: czterozębnego Ceutorhynchus quadridens Panz. i pdobnika C. assimilis Payk.. Polskie Pismo Entomologiczne 3, Seria B, Zeszyt **1-2** (9-10), 15-20.

Dosdall LM, Herbut MJ & NT Cowle (1994): Susceptibilities of species and cultivars of canola and mustard to infestation by root maggots (*Delia* ssp.) (Diptera: Anthomyiidae). The Canadian Entomologist **126**, 251-260.

Dosdall LM & LS Kott (2006): Introgression of resistance to cabbage seedpod weevil to canola from yellow mustard. Crop Science **46**, 2437-2445.

Earle ED, Cao J & AM Shelton (2004): Insect-resistant transgenic Brassicas. In: Pua EC & CJ Douglas (ed), *Biotechnology in Agriculture and Forestry – Brassica* **54**. Springer, Berlin.

Eigenbrode SD & AT Trumble (1994): Host plant resistance to insects in integrated pest management in vegetable crops. Journal of Agricultural Entomology **11**, 201-224.

Ekbom B (1995): Insect Pests. In: Kimber DS & DJ McGregor (ed), Brassica *oilseeds, Production and Utilization*. CAB International, Wallingford, 141-152.

Ellis PR (1992): Sources of resistance in Brassicas to cabbage root fly. IOBC/wprs Bulletin **15** (4), 94-101.

Ellis PR & JA Farrell (1995): Resistance to cabbage aphid (*Brevicoryne brassicae*) in six brassica accessions in New Zealand. New Zealand Journal of Crop and Horticultural Science **23**, 25-29.

Ellis PR, Ramsey AD, Singh R & DAC Pink (1996): Wild Brassica species as sources of resistance to *Brevicoryne brassicae* and *Aleyrodes proletella*. IOBC/wprs Bulletin **19**, 1-7.

Ellis PR, Pink DAC, Barber NE & A Mead (1999): Identification of high levels of resistance to cabbage root fly, *Delia radicum*, in wild *Brassica* species. Euphytica **110**, 207-214.

Ellis PR & NB Kift (2003): The exploitation of plant resistance in controlling insect pests of vegetable crops. IOBC/wprs Bulletin **26** (3), 47-55.

FAO (2005): http//apps.fao.org/page/collections?subset=agriculture

Fahey JW, Zalcmann AT & P Talalay (2001): The chemical diversity and distribution of glucosinolates and isothiocyanates among plants. Phytochemistry **56**, 5-51.

Farrell JAK (1977): Plant resistance to insects and the selection of resistant lines. The New Zealand Entomogist **6**, 244-261.

Felkl G, Jensen EB, Kristiansen K & SB Andersen (2005): Tolerance and antibiosis resistance to cabbage root fly in vegetable *Brassica* species. Entomologia Experimentalis et Applicata **116**, 65-71.

Fenwick GR, Heaney RK & WJ Mullin (1983): Glucosinolates and their breakdown products in food and food plants. CRC - Critical reviews in Food Science and Nutrition **128**, 123-201.

Ferguson AW, Klukowski Z, Walczak B, Clark SJ, Mugglestone MA, Perry JN & IH Williams (2003): Spatial distribution of pest insects in oilseed rape: implications for integrated pest management. Agriculture, Ecosystems and Environment **95**, 509-521.

Ferguson AW, Barari H, Warner DJ, Campbell JM, Smith ET, Nigel P, Watts NP & IH Williams (2006): Distributions and interactions of the stem miners *Psylliodes chrysocephala* and *Ceutorhynchus pallidactylus* and their parasitoids in a crop of winter oilseed rape (*Brassica napus*). Entomologia Experimentalis et Applicata **119**, 81–92.

Fieldsend J & GFJ Milford (1994): Changes in glucosinolates during crop development in single- and double-low genotypes of winter oilseed rape (Brassica napus): II. Profiles and tissue-water concentration in vegetative tissues and developing pods. Annals of Applied Biology **124**, 543-555.

Free JB & IH Williams (1979): The infestation of crops of oil-seed rape (*Brassica napus* L.) by insect pests. Journal of Agricultural Science **92**, 203-2018.

Friesland H (1990): Ein meteorologisch begründetes Prognoseverfahren für den Kohltriebrüssler in Winterraps. Landwirtschaftliches Jahrbuch **67**, Sonderheft.

Fritzsche R (1955): Phaeonia trimaculata Bouché als Parasit des Großen Kohltriebrüßlers Ceutorrhynchus napi Gyll. und des Gefleckten Kohltriebrüßlers Ceutorrhynchus quadridens Panz.. Sonderdruck aus Nachrichtenblatt für den Deutschen Pflanzenschutzdienst **9**, 35-36.

Garbe V, Broschewitz B, Erichsen E, Hoßfeld R, Lauenstein G, Steinbach P & B Ulber (1996): Schadschwellen bei Rapsschädlingen. Instrumente einer wirtschaftlichen Winterrapsproduktion. RAPS **14**, 58-63.

Giamoustaris A & RF Mithen (1995): The effect of modifying the glucosinolate content of leaves of oilseed rape (*Brassica napus ssp. oleifera*) on its interaction with specialist and generalist pests. Annals of Applied Biology **126**, 347-363.

Giamoustaris A & RF Mithen (1997): Glucosinolates and disease resistance in oilseed rape (*Brassica napus ssp. oleifera*). Plant Pathology **46**, 271-275.

Girard C, Le Metayer M, Zaccomer B, Bartlet E, Williams I, Bonade-Bottino M, Pham-Delegue M & L Jouanin (1998): Growth stimulation of beetle larvae reared on a transgenic oilseed rape expressing a cysteine proteinase inhibitor. Journal of Insect Physiology **44**, 263-270.

Girke A (2002): Neue Genpools aus resynthetisiertem Raps (*Brassica napus* L.) für die Hybridzüchtung. PhD-thesis, Georg-August-Universität, Goettingen.

Graham CW & HJ Gould (1980): Cabbage stem weevil (*Ceutorhynchus quadridens*) on spring oilseed rape in Southern England and its control. Annals of Applied Biology **95**, 1-10.

Günthart E (1949): Beiträge zur Lebensweise und Bekämpfung von *Ceutorhynchus quadridens* PANZ. und *Ceutorhynchus napi* Gyll. mit vielen Beobachtungen an weiteren Kohl- und Rapschädlingen. Mitteilungen der Schweizerischen Entomologischen Gesellschaft **23**, 441-591.

Hanson LM (2003): Insecticide-resistant pollen beetles (Meligethes aeneus F.) found in Danish oilseed rape (Brassica napus L.) fields. Pest Management Science **59**, 1057-1059.

Hardman JA & PR Ellis (1978): Host plant factors influence the susceptibility of cruciferous crops to cabbage root fly attack. Entomologia Experimentalis et Applicata **24**, 193-197.

Harmon BL & JP McCaffrey (1997): Laboratory bioassay to assess Brassica spp. germplasm for resistance to the cabbage seedpod weevil (Coleoptera: Curculionidae). Journal of Economic Entomology **90**, 1392-1399.

Hartleb H, Wolff C, Kufeld W & H Briswitz (1998): Phoma-Spätbefall durch Triebrüsslerbekämpfung reduzieren. RAPS **16**, 26-31.

Hawkes C, Patton S & TH Coaker (1978): Mechanisms of host plant finding in adult cabbage root fly, *Delia brassicae*. Entomologia Experimentalis et Applicata **24**, 219-227.

Heimbach U, Müller A & T Thieme (2006): First steps to analyse pyrethroid resistance of different oilseed rape pests in Germany. Nachrichtenblatt des Deutschen Pflanzenschutzdienstes **58**, 1-5.

Henderson AE, Hallett RH & JJ Soroka (2004): Prefeeding behavior of the crucifer flea beetle, *Phyllotreta cruciferae,* on host and nonhost crucifers. Journal of Insect Behavior **17**, 17-39.

Holecová M, Némethová D & M Kúdela (2005): Structure and function of weevil assemblages (Coleoptera, Curculionidae) in epigeon of oak-hornbeam forests in SW Slovakia. Ekológia **2**, 179-204.

Huang XP & JAA Renwick (1994): Relative activities of glucosinolates as oviposition stimulants for *Pieris rapae* and *Pieris napi oleracea*. Journal of Economic Entomology **20**, 1025-1037.

Jensen EB, Felkl G, Kristiansen K & SB Andersen (2002): Resistance to the cabbage root fly, *Delia radicum*, within *Brassica fruticulosa*. Eupyhtica **124**, 379-386.

Johnen, A & H Meier (2000): A weather-based decision support system for managing oilseed rape pests. Proceedings of British Crop Protection Council (BCPC) Conference Pests & Diseases 2000, Brighton, UK, 793-800.

Johnen A, Ulber B, Williams IH, Klukowski Z & C Nielsson (2006): Prüfung der Vorhersagegenauigkeit der proPlant-Phänologiemodelle für wichtige Schädlinge im Winterraps unter heterogenen klimatischen Bedingungen in Europa. Mitteilungen der Biologischen Bundesanstalt für Land- und Forstwirtschaft **400**, 277.

Jönsson M, Lindkvist A & P Anderson (2005): Behavioural responses in three ichneumonid pollen beetle parasitoids to volatiles emitted from different phenological stages of oilseed rape. Entomologia Experimentalis et Applicata **111**, 363-369.

Jyoti JL, Shelton AM & ED Earle (2001): Identifying sources and mechanisms of resistance in crucifers for control of cabbage maggot (Diptera: Anthomyiidae). Journal of Economic Entomology **94**, 942-949.

Kalischuk AR and LM Dosdall (2004): Susceptibilities of seven Brassicaceae species infestation by the cabbage seedpod weevil (Coleoptera: Curculionidae). The Canadian Entomologist **136**, 265-276.

Kangas E (1976): Über die an Cruciferae lebenden Ceutorhynchus-Arten (Col., Curculionidae) Finnlands. Annales Agriculturae Fenniae **15**, 72-80.

Kazda V (1953): Bionomic a hospodarsky vyznam krytonosce ctyrzubeho, Ceutorhynchus quadridens (Panz.), v strednich cechach. Zoologice a entomologice listy **2**, 231-240.

Kazda V (1957): Die wichtigsten Rapsschädlinge in der Tschechoslowakei und ein Versuch der Prognosestellung des Schadauftretens von *Ceutorhynchus napi* GYLL.. 4. Internationaler Pflanzenschutzkongress Hamburg **1**, 729-732.

Kelm M & B Walczak (1998): The relationship between the stem weevil (*Ceutorhynchus pallidactylus* Marsh.) injury and losses of the flower buds. IOBC/wprs Bulletin **21** (5), 147-151.

Kelm M & Z Klukowski (2000): The effect of stem weevil (*Ceutorhynchus pallidactylus* Marsh.) infestation on oilseed rape yield. IOBC/wprs Bulletin **23** (6), 125-130.

Kirk WDJ (1992): Insects on cabbages and oilseed rape. Naturalists' Handbooks 18, Richmond Publishing, Slough.

Kjaer-Pedersen C (1992): Dispersive flight of the cabbage stem weevil. Proceedings of the 8th International Symposium on Insect Plant Relationship Wageningen, Netherlands, 9-13 March 1992.

Klingenberg A & B Ulber (1994): Untersuchungen zum Auftreten der Tersilochinae (Hym., Ichneumonidae) als Larvenparasitoide einiger Rapsschädlinge im Raum Göttingen 1990 und 1991 und zu deren Schlupfabundanz nach unterschiedlicher Bodenbearbeitung. Journal of Applied Entomology **117**, 220-227.

Kogan M & EE Ortmann (1978): Antixenosis – a new term proposed to replace Painter's 'non-preference' modality of resistance. Bulletin of the Entomological Society of America **24**, 175-176.

Koritsas VM, Lewis JA & GR Fenwick (1991): Glucosinolate responses of oilseed rape, mustard and kale to mechanical wounding and infestation by cabbage stem flea beetle (*Psylliodes chrysocephala*). Annuals of Applied Biology **118**, 209-221.

Körting A (1942): Über die Lebensweise des Gefleckten Kohltriebrüßlers (*Ceutorhynchus quadridens* Panz.) und seine Bedeutung als Ölfruchtschädling. Arbeiten über physiologische und angewandte Entomologie aus Berlin Dahlem **9**, 207-237.

Lamb RJ (1989): Entomology of oilseed *Brassica* crops. Annual Review of Entomology **43**, 211-229.

Lambdon PW, Hassel M & R Mithen (1998): Feeding preference of woodpigeons and flea-beetles for oilseed rape and turnip rape. Annals of Applied Biology **133**, 313-328.

Landschreiber M (2005): Die Vorblütenschädlinge sind weiter auf dem Vormarsch. RAPS **23**, 4-9.

Larsen LM, Lind F, Nielsen JK, Olsen O, Pedersen L & H Sorensen (1983): Studies of insects associated with crucifers: Host plant selection as a function of allelochemicals present in the plants. Problems related to rape containing high and low glucosinolates. Proceedings of the 6th International Rapeseed Conference **1**, 270-275.

Lehmann W (1965): Einfluss chemischer Bekämpfungsmaßnahmen auf einige Rapsschädlinge und ihre Parasiten – Teil II: Knospen- und Stängelschädlinge. Archiv für Phytopathologie und Pflanzenschutz **1**, 209-219.

Lehrmann A (2007): Oilseed rape transformed with a pea lectin gene. PhD-thesis, Swedish University of Agricultural Science, Uppsala.

Lerin J (1995): Assessment of yield losses caused by insects in winter oilseed rape, a critical review. IOBC/wprs Bulletin **18** (4), 95-101.

Liblikas I, Mõttus E, Borg-Karlson AK, Kusik S, Ojarand A, Kännaste A & J Tanilsoo (2003): Flea bettle (Coleoptera: Chrysomelidae) response to alkyl thiocyanates and alkyl isothiocyanates. Agronomy Research **1**, 175-184.

Lücke W & H-J Pluschkell (1982): Erfahrungen und Schlussfolgerungen zur gezielten Bekämpfung der Rapsschädlinge im Bezirk Rostock. Nachrichtenblatt für den Pflanzenschutzdienst in der DDR **36**, 158-161.

Lühs W, Ulber B, Seyis F, Frauen M, Busch H, Friedt W, Gustafsson M & G Poulsen (2003): Evaluation of a Brassica napus core collection and prospects of its exploitation through rapeseed breeding. 11th International Rapeseed Congress Copenhagen, Denmark, July 2003, 2, 371-374.

Manthey R (2005): Aktueller Leistungsvergleich der in Deutschland zugelassenen und untersuchten Winterrapssorten. RAPS **23**, 134-137.

McCaffrey JP, Harmon BL, Brown J, Brown AP & JB Davis (1999): Assessment of *Sinapis alba*, *Brassica napus* and *S. alba* x *B. napus* hybrids (Coleoptera: Curculionidae). Journal of Agricultural Science **132**, 289-295.

Meiners T & M Hilker (2002): Plant's defensive responses towards insect oviposition. IOBC/wprs **25** (6), 165-168.

Messina FJ & AJ Bloxham (2004): Plant resistance to the Russian wheat aphid: effects on a nontarget aphid and the role of induction. The Canadian Entomologist **136**, 129-137.

Miles CL, del Campo ML & JAA Renwick (2005): Behavioural and chemosensory responses to a host recognition cue by larvae of *Pieris rapae*. Journal of Comparative Physiology A **191**, 147-155.

Mithen R (1992): Leaf glucosinolate profiles and their relationship to pest and disease resistance in oilseed rape. Euphytica **63**, 71-83.

Mrówczyński M (1992): Injuries of winter rapeseed variety types by pests. Part II. Stem Pests. Prace Naukowe IOR Poznan – **XXXIV** (1/2).

Mrówczyński M (1998): Studies on pest damage of cultivars and lines of winter oilseed rape. IOBC/wprs Bulletin **21** (5), 153-154.

Murray PJ, Cheung AKM & MT Abberton (2007): Intraspecific variation in *Trifolium pratense*: impact on feeding and host location by *Sitona lepidus* (Coleoptera, Curculionidae). Journal of Pest Science **80**, 51-57.

Nissen U (1997): Ökologische Studien zum Auftreten von Schadinsekten und ihren Parasitoiden an Winterraps norddeutscher Anbaugebiete. PhD-thesis, Christian-Albrechts-Universität, Kiel.

Nitzsche O & B Ulber (1998): Einfluß der Bodenbearbeitung nach Winterraps auf die Schlupfabundanz der Parasitoiden von *Ceutorhynchus napi*, *Ceutorhynchus pallidactylus* und *Meligethes aeneus*. Mitteilungen der Biologischen Bundesanstalt **357**, 62-63.

Nuss H (2004): Effect of plant density and plant architecture on the abundance and within-plant distribution of stem borers of winter oilseed rape. PhD-thesis, Georg-August-Universität, Goettingen.

Nuss H & B Ulber (2007): Effect of sowing rate on the infestation of winter oilseed rape by stem-boring pests and on yields. 12th International Rapeseed Congress, March 26-30, 2007 (Wuhan, China), IV (Plant Protection), 290-293.

Painter RH (1951): Insect resistance in crop plants. Macmillan. New York.

Pałosz T (1978): Ceutorhynchus quadridens PANZ. (Col., Curculionidae) as a pest of winter rape in Poland. Polskie Pismo Entomologiczne **48**, 653-663.

Pałosz T (1980): Einschätzung des Schadenskoeffizienten einiger Rapsschädlinge. Tagungsbericht Nr. 181 der Akademie der Landwirtschaftswissenschaften der Deutschen Demokratischen Republik - Sonderdruck.

Pałosz T, Sieńkowski A & B Grala (1994): Statistische Beziehungen zwischen einigen Rapsanbaufaktoren. Archiv für Phytopathologie und Pflanzenschutz **29**, 101-106.

Paulsen HM, Schochow M, Ulber B, Kühne S & G Rahmann (2006): Mixed cropping systems for biological control of weeds and pests in organic oilseed crops. Aspects of Applied Biology **79**, 215-220.

Pink DAC, Kift NB, Ellis PR, McClement SJ, Lynn J & GM Tatchell (2003): Genetic control of resistance to the aphid Brevicoryne brassicae in the wild species Brassica fruticulosa. Plant Breeding **122**, 24-29.

Piper R & I Williams (2004): Incidence and feeding activity of epigeic, predatory invertebrates within winter oilseed rape in the UK with comparisons between integrated and conventional crop management. IOBC/wprs Bulletin **27** (10), 281-288.

Pontoppidan B, Hopkins R, Rask L & J Meijer (2003): Infestation by cabbage aphid (*Brevicoryne brassicae*) on oilseed rape (*Brassica napus*) causes a long lasting induction of the myrosinase system. Entomologia Experimentalis et Applicata **109**, 55-62.

Potting RPJ, Perry JN & W Powell (2005): Insect behavioural ecology and other factors affecting the control efficacy of agro-ecosystern diversification strategies. Ecological Modelling **182**, 199-216.

Rask L, Andréasson E, Ekbom B, Eriksson S, Pontoppidan B & J Meijer (2000): Myrosinase: gene family evolution and herbivore defense in Brassicaceae. Plant Molecular Biology **42**, 93-113.

Reay-Jones FPF, Way MO, Sétamou M, Legrende BL & TE Reagan (2003): Resistance to the Mexican rice borer (Lepidoptera: Crambidae) among Louisiana and Texas sugarcane cultivars. Journal of Economic Entomology **96**, 1929-1934.

Risch SJ (1985): Effects of induced chemical changes on interpretation of feeding preference tests. Entomologia Experimentalis et Applicata **39**, 81-84.

Rygulla W, Snowdon RJ, Eynck C, Koopmann B, von Tiedemann A, Lühs W & W Friedt (2007): Broadening the genetic basis of *Verticillium longisporum* resistance in *Brassica napus* by interspecific hybridization. Phytopathology **97**, 1391-1396.

Sarfraz M, Dosdall LM & BA Keddie (2007): Resistance of some cultivated Brassicaceae to infestations by Plutella xylostella (Lepidoptera: Plutellidae). Journal of Economic Entomology **100**, 215-224.

Schlein O & W Büchs (2004): Approaches to assess the importance of carnivorous beetles as predators of oilseed rape pests. IOBC/wprs Bulletin **27** (10), 289-292.

Schoonhoven LM, Jermy T & JJA van Loon (1998): Host plant selection: how to find a host plant. In: *Insect-Plant Biology – From physiology to evolution*. Chapman & Hall, London, 121-154.

Šedevý J & J Vašák (2002): Differences in flight activity of pests on winter and spring oilseed rape. Plant Protection Science **38**, 139-144.

Seta G & A Wolski (2006): Trial of qualification of harmfulness and effectiveness of *Meligethes aeneus* F. and *Ceutorhynchus pallidactylus* Marsh. on winter oilseed rape control in dependence of air temperature in spring time. Progress in Plant Protection **46**, 390-394.

Seyis F, Friedt W & W Lühs (2000): Resynthese-Raps (*Brassica napus* L.) als genetische Ressource für die Qualitäts- und Ertragszüchtung. In: Hammer K & T Gladis (eds), *Nutzung genetischer Ressourcen – ökologischer Wert der Biodiversität*. Schriften zu Genetischen Ressourcen **16**. ZADI, Bonn, 91-112.

Singh R & PR Ellis (1993): Sources, mechanisms and bases of resistance in cruciferae to the cabbage aphid, *Brevicoryne brassicae*. IOBC/wprs Bulletin **16** (5), 21-35.

Speyer W (1921): Beitrag zur Biologie des Gefleckten Kohltriebrüsslers (*Ceutorrynchus quadridens* Panz.). Entomologische Blätter **17**, 118-124.

Stoner KA (1990): Glossy leaf wax and plant resistance to insects in *Brassica oleracea* under natural infestation. Environmental Entomology **19**, 730-739.

Tommes S (1998): Herbivor-Parasitoid-Gesellschaften an Kreuzblütlern (Brassicaceae): Artenreichtum und Interaktionen. Master-thesis, Georg-August-Universität, Goettingen.

U N (1935): Genom analysis in Brassica with special reference to the experimental formation of B. napus and peculiar mode of fertilization. Japanese Journal of Botany 7, 389-452.

UFOP (2008): Online: http://www.ufop.de/2165.php

Ulber B (2003): Parasitoids of ceutorynchid stem weevils. In: DV Alford (ed), *Biocontrol of Oilseed Rape Pests*. Blackwell Science, Oxford, 87-95.

Ulber B & K Fischer (2006): Effect of plant density and host plant architecture on the abundance and within-plant distribution of stem-mining pests and the level of parasitism. CD-Rom Proceedings of the International Symposium "Integrated Pest Management in Oilseed Rape Pests", Goettingen, Germany, 3-5 April 2006.

Ulber B & O Nitzsche (2006): Phenology of parasitoids (Hym., *Ichneumonidae*, *Tersilochinae*) of oilseed rape pests in northern Germany in 1995-1997. IOBC/wprs Bulletin **29** (7), 173-179.

Ulber B & R Wedemeyer (2006): Responses of *Tersilochus microgaster* und *Tersilochus obscurator* (Hymenoptera: Ichneumonidae) to volatile 2-phenylethyl-isothiocyanate. CD-Rom Proceedings of the International Symposium "Integrated Pest Management in Oilseed Rape Pests", Goettingen, Germany, 3-5 April 2006.

Ulmer BJ, Gillot C & M Erlandson (2001): Feeding preferences, growth, and development of *Mamestra configurata* (Lepidoptera: Noctuidae) on Brassicaceae. The Canadian Entomologist **133**, 509-519.

Ulmer, BJ & LM Dosdall (2006): Glucosinolate profile and oviposition behaviour in relation to the susceptibilities of Brassicaceae to the cabbage seedpod weevil. Entomologia Experimentalis et Applicata **121**, 203-213.

van Loon JJA, Blaakmeer A, Griepink FC, Van Beek TA, Schoonhoven LM & A de Grot (1992): Leaf surface compound from *Brassica oleraceae* (Cruciferae) induces oviposition by *Pieris brassicae* (Lepidoptera: Pieridae). Chemoecology **3**, 39-44.

Wahmhoff W (2000): Integrierter Rapsanbau. Untersuchungen zur Entwicklung integrierter Produktionsverfahren am Beispiel des Winterrapses (*Brassica napus* L.). Erich Schmidt Verlag, Berlin.

Walczak B, Kelm M, Klukowski Z, Smart LE, Ferguson AW & IH Williams (1998): The effect of trap design and 2-phenylethyl isothiocyanate on catches of stem weevils (*Ceutorhynchus pallidactylus* Marsh. and *C. napi* Gyll.) in winter oilseed rape. IOBC/wprs Bulletin **21** (5), 141-146.

Wallsgrove R, Bennett R, Kiddle G, Bartlet E & J Ludwig-Mueller (1999): Glucosinolate biosynthesis and pest/disease interactions. Proceedings of the 10th International Rapeseed Congress, Canberra, Australia, 1999, 104-107.

Way MJ & G Murdie (1965): An example of varietal variations in resistance of Brussels sprouts. Annuals of Applied Biology **56**, 326-328.

Weber G, Oswald S & U Zöllner (1986): Die Wirtseignung von Rapssorten unterschiedlichen Glucosinolatgehaltes für *Brevicoryne brassicae* (L.) und *Myzus persicae* (Sulzer) (Hemiptera, Aphididae). Journal of Plant Diseases and Protection **93**, 113-124.

Wegorek P, Obrepalska-Steplowska A, Zamoyska J & Nowaczyk (2006): Resistance of pollen beetle (*Meligethes aeneus* F.) in Poland. Resistant Pest Management Newsletter **16**, 28-29.

Williams IH (2004): Advances in Insect Pest Management of Oilseed Rape in Europe. In: AR Horwitz (ed), *Insect Pest Management*. Springer Verlag, Berlin, 181-208.

Winfield AL (1961a): Observations on the biology and control of the cabbage stem weevil, *Ceutorhynchus quadridens* Panz. on throwse mustard (*Brassica juncea*). Bulletin of Entomological Research **52**, 589-600.

Winfield AL (1961b): Studies on the relationship between three species of Coleoptera and certain species of annual mustard and rape. Entomologia Experimentalis et Applicata **4**, 123-132.

Winfield AL (1992): Management of oilseed rape pests in Europe. Agricultural Zoology Reviews **5**, 51-95.

Winter H (2004): Untersuchungen zur Introgression von Resistenzen gegen die Wurzelhals- und Stengelfäule [*Leptophaeria maculans* (Desm.) Ces. et De Not.] aus verwandten Arten in den Raps (*Brassica napus* L.). PhD-thesis, Freie Universität, Berlin.

Yungchang L, Kiddle G, Bennett R, Doughty K & R Wallsgrove (1999): Variation in the glucosinolate content of vegetative tissue of Chinese lines of Brassica napus L..Annals of Applied Biology **134**, 131-136.

Xue-bao L, Hui-Zhu M & B Yong-Yan (1995): Transgenic plants of rutabaga (*Brassica napobrassica*) tolerant to pest insects. Plant Cell Reports **15**, 97-101.

Chapter II - Screening of oilseed rape and other brassicaceous genotypes for susceptibility to cabbage stem weevil (*Ceutorhynchus pallidactylus* (Mrsh.))

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Abstract

Production of oilseed rape, *Brassica napus* L., is affected by various insect pests. The cabbage stem weevil, *Ceutorhynchus pallidactylus* (Mrsh.) (Col.: Curculionidae), is one of the most damaging pests in Northern and Central Europe that requires regular control measures. Host plant resistance is a key factor in integrated pest management systems. To evaluate a large number of genotypes for their susceptibility to infestation by *C. pallidactylus,* new screening techniques were developed for testing both, the amount of feeding and the number of eggs deposited by adult *C. pallidactylus* on accessions of Brassicaceae under controlled conditions.

In no-choice screening tests, the leaf area consumed by adult cabbage stem weevil was quantified on a wide spectrum of 107 brassicaceous genotypes (*B. napus*, *B. rapa* L. and *B. oleracea* L. cultivars, breeding lines, resynthesized rapeseed lines and wild *Brassicaceae*). In comparison to feeding on the standard cultivar 'Express', the average leaf area consumed by *C. pallidactylus* on nine oilseed rape cultivars, four resynthesized rapeseed lines and five other accessions [*B. oleracea*, *Camelina alyssum* (Mill.) and *Lunaria annua* L.] was significantly reduced by 44 – 90%. In dual-choice screening tests for the evaluation of oviposition preferences on 42 genotypes, female *C. pallidactylus* laid significantly fewer eggs into plants of two oilseed rape cultivars, five resynthesized rapeseeds and three accessions of *B. oleracea* and *B. fruticulosa* Cyrillo, respectively, than into plants of the standard cv 'Express'. Results of both laboratory screening test were confirmed by results of additional field testing.

Keywords: *Brassica*; cabbage stem weevil; dual-choice oviposition test; no-choice feeding test; oilseed rape; plant resistance

Chapter III - Resynthesized lines and cultivars of *Brassica napus* L. as sources of resistance to cabbage stem weevil (*Ceutorhynchus pallidactylus* (Mrsh.))

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Abstract

The cabbage stem weevil (*Ceutorhynchus pallidactylus* (Mrsh.)) (Col., Curculionidae) is a serious pest of winter oilseed rape (*Brassica napus* L. *var. oleifera* Metzg.) in central and northern Europe. Although host-plant resistance is a key tool in integrated pest management systems, resistant genotypes are not yet available for this species. Resynthesized rapeseed lines (*B. oleracea* L.×*B. rapa* L.) are broadening the genetic diversity and might have potential as sources of resistance to pest insects. The host quality, of nine resynthesized rapeseed lines and six genotypes of *B. napus* to cabbage stem weevil, was evaluated in laboratory screening tests and in a semi-field experiment. In dual-choice oviposition tests, female *C. pallidactylus* laid significantly fewer eggs on five resyntheses and on swede cv 'Devon Champion' than on the moderately susceptible oilseed rape cv 'Express', indicating a lower host quality of these genotypes. Results of laboratory screenings were confirmed in a semi-field experiment, in which twelve genotypes were exposed to *C. pallidactylus* females. The number of larvae was significantly lower in two resyntheses and in cv 'Devon Champion' than in oilseed rape cv WVB 9. The total, as well as individual, glucosinolate (GSL) content in the leaves differed substantially among the genotypes tested. The amount of feeding by larvae of *C. pallidactylus*, as measured by a stem-injury coefficient, was positively correlated with the indolyl GSL compounds 3-indolylmethyl and 4-methoxy-3-indolylmethyl, and with the aromatic GSL 2-phenylethyl, whereas it was negatively correlated with 4-hydroxy-3-indolylmethyl. Thus, the composition and concentration of GSL compounds within the plant tissue might be a key factor in breeding for pest resistance in oilseed rape.

Keywords: glucosinolates; larval performance; oilseed rape, resynthesis; semi-field trial; stem injury coefficient

Chapter IV - Influence of different *Brassica* host plants on parasitism of ceutorynchid stem weevils

Abstract

The rape stem weevil (*Ceutorhynchus napi*) and the cabbage stem weevil (*C. pallidactylus*) are major pests of winter oilseed rape (*Brassica napus* L. *var. oleifera* Metzg.) in Europe. Their stem-mining larvae are parasitized by the specialist larval endoparasitoids, *Tersilochus fulvipes* and *T. obscurator*, respectively. In three-year field experiments, the effect of host plant genotype on the level of parasitism of the ceutorynchid larvae by T. *fulvipes* and *T. obscurator* was determined on a large assortment of *Brassica* genotypes (*B. napus*, *B. rapa*, *B. oleracea* and *B. juncea*). The species of *Brassica* plant had a significant effect on larval parasitism. In three cultivars of turnip rape, *B. rapa*, the level of parasitism of *C. napi* by *T. fulvipes* was significantly reduced compared to cultivars of winter oilseed rape, *B. napus*. Based on data pooled over three years, the rate of parasitism on *C. pallidactylus* by *T. obscurator* was significantly reduced in cv 'Wotan' in comparison to other accessions, whereas parasitism of larvae on fodder rape 'Emerald' (*B. napus*) was significantly increased in two of three years. The level of parasitism of *C. pallidactylus* and *C. napi* by *T. obscurator* and *T. fulvipes*, respectively, was not significantly correlated with the number of host larvae/m^2. The length of main stem and number of racemes/plant had a significantly negative effect on parasitism by *T. fulvipes* in 2004. However, weak correlations of parasitism rates with plant growth parameters indicate that other cues have major importance for the parasitism of ceutorynchid host larvae.

Introduction

Multitrophic interactions between host plants, pests and parasitoids have been reported several times in literature (Vet & Dicke 1992; Fox *et al.* 1996; Lill *et al.* 2002). Host plant quality and genotype have potential to affect the infestation by herbivorous insects as well as the level of biocontrol by natural enemies. Cabbage stem weevil, *C. pallidactylus* (Mrsh.), and rape stem weevil, *C. napi* Gyll., (Coleoptera: Curculionidae) are major pests of winter oilseed rape, *Brassica napus* L. *var. oleifera* Metzg. in Central and Northern Europe (Alford *et al.* 2003). Both species are univoltine with three larval instars. Adult stem weevils migrate from their overwintering habitats to crops of oilseed rape in March/April. Female *C. napi* lay their eggs singly into the top of the main stem, whereas female *C. pallidactylus* lay egg batches into petioles and mid-ribs of leaves. The larvae of either species mine within the pith of host plants. Mature larvae migrate to soil for pupation. On crops of winter oilseed rape, the univoltine larval endoparasitoids *Tersilochus fulvipes* Gravenhorst and *T. obscurator* Aubert (Hymenoptera: Ichneumonidae) have been identified as key parasitoids of rape stem weevil and cabbage stem weevil, respectively (Klingenberg & Ulber 1994; Ulber 2003; Ulber & Nitzsche 2006). Both parasitoid species are assumed to be host-specific (Ulber 2003). Natural control of host populations by these parasitoid species has been reported from various European countries, e.g. Austria (Kraus & Kromp 2002), Czech Republic (Sedivy 1983), France (Jourdheuil 1960), Germany (Klingenberg & Ulber 1994), Switzerland (Büchi & Roos-Humbel 1991), and the UK (Barari *et al.* 2005).

Female parasitoids lay their eggs singly into host larvae while these are feeding concealed within petioles (*T. obscurator*) or stems (*T. fulvipes*). Parasitoid larvae hatch within hosts; however, development of second and third instar larvae is delayed until the full-grown host larvae have migrated to soil for pupation (koinobiont). The adult wasps diapause inside the pupal cocoon until emergence in the following spring (Ulber 2003). The multitrophic system of host plant (oilseed rape), host larva (ceutorynchid stem weevil) and parasitoid (*Tersilochus* spp.) is affected by various factors, e.g. climate (Johnen *et al.* 2006), plant architecture (Ulber & Fischer 2006), plant volatiles (Jönsson *et al.* 2005), and farming practices, like plant density (Ulber & Fischer 2006), tillage (Klingenberg & Ulber 1994; Nitzsche & Ulber 1998), and insecticide application (Klukowski 2006; Neumann & Ulber 2009, in prep.). The objectives of this research were to investigate influences of host plant species and cultivars on the level of infestation and larval parasitism of *C. napi* and *C. pallidactylus* by *T. fulvipes* and *T. obscurator*, respectively. Three field plot experiments were conducted in 2004, 2005 and 2006 at Goettingen (Northern Germany) to study the level of parasitism in various *Brassica* genotypes and the effect of plant architecture on parasitism by *T. obscurator* and *T. fulvipes*.

Materials and Methods

Each year, a broad spectrum of *Brassica* species and cultivars was grown in a randomized block design with four replicated plots (12m * 2.5m; 30m^2 per plot) (Table 1). Selection of cultivars was aimed to cover a broad host plant spectrum for stem weevils.

Table 1: *Brassica* species and cultivars included in field experiments

No. of cultivars	Species (Crop type)	Cultivar	Source *
8	*Brassica napus* (open pollinated oilseed rape)	‘Express’, ‘Bristol’, ‘Idol’, ’Lisek’, ‘Prince’, ‘Smart’, ‘Viking’, ‘Wotan’	A, B, C
5	*Brassica napus* (hybrid oilseed rape)	‘Artus’, ‘Elan’, ‘Extra’, ‘Talent’, WRH 242	A, B
1	*Brassica napus* (fodder rape)	‘Emerald’	F
3	*Brassica rapa* (turnip rape)	‘Malvira’, ‘Perko’, ‘Salut’	A, D, E
2	*Brassica oleracea* (fodder cabbage)	‘Markola’, ‘Gruener Angeliter’	A, F
1	*Brassica juncea* (Indian mustard)	‘Vitasso’	G

* A: Norddeutsche Pflanzenzucht Hans-Georg Lembke KG (NPZ); B: Deutsche Saatveredelung Lippstadt-Bremen GmbH (DSV); C: Syngenta Seeds GmbH; D: KWS Saat AG; E: Semundo Saatzucht GmbH; F: Raps GbR; G: Lochow-Petkus.

Four cultivars of *B. napus* were examined in all three years (‘Express’, ‘Prince’, ‘Wotan’ and fodder rape ‘Emerald’) while five cultivars were studied in two years and eleven cultivars only in one year. The standard cv ‘Express’ was grown around the experiment to avoid edge effects. Plant densities were monitored at the beginning of March. Plant growth (length of main stem, stem-base diameter, number of leaves and lateral racemes on main stem) was assessed from samples of 20 plants/plot at the end of April.

The effect of host plant genotype on the abundance of host larvae and the level of parasitism was determined from samples of 20 plants/plot collected in mid May, before larvae started to migrate to soil for pupation. Stems and lateral racemes were cut longitudinal to collect larvae of stem weevils. They were stored in 70% Ethanol and later dissected under a microscope (sample size: 40 – 250 larvae/genotype). The identity of *T. fulvipes* and *T. obscurator* was confirmed by rearing out adult parasitoids from subsamples of host larvae. Full-grown host larvae were transferred to boxes containing moist soil substrate; after ten weeks, parasitoid cocoons were isolated from soil and adult parasitoids identified. All data were subjected to ANOVA; differences between means ($p \leq 0.05$) were evaluated by Tukey-HSD test. Percentages were transformed by arc-sin-transformation. Regression analyses were conducted by using Sigma Plot Version 10.

Results

Abundance of stem weevils and overall level of parasitism in 2004-2006

In all years, the number of stem weevil larvae per plant was relatively high, particularly in 2005 (*C. napi*) and in 2006 (*C. pallidactylus and C. napi*) (Table 2). The level of parasitism of *C. pallidactylus* larvae was higher than parasitism of *C. napi* and increased over years. Only specimen of *T. fulvipes* and *T. obscurator* were obtained by rearing adult parasitoids from host larvae.

Table 2. Mean number (± SEM) of ceutorynchid larvae and mean percentage (± SEM) of parasitism in field experiments at Goettingen in 2004 – 2006

	C. pallidactylus		***C. napi***	
	Larvae/plant	**% parasitism**	**Larvae/plant**	**% parasitism**
2004	3.4 ± 0.5	17.3 ± 1.9	5.4 ± 0.5	10.8 ± 2.4
2005	3.6 ± 0.3	20.8 ± 1.5	9.8 ± 0.7	15.2 ± 1.9
2006	11.5 ± 2.8	27.6 ± 2.5	8.8 ± 1.7	15.1 ± 2.7

Larval abundance and parasitism of C. pallidactylus

Significant differences were detected between the number of host larvae on 16 different cultivars tested in 2004 (Fig 1). Plant infestation by *C. pallidactylus* was significantly reduced in the accession of *B. oleracea* ‘Markola’ and ‘Gruener Angeliter’ compared to *B. rapa* cultivars ‘Malvira’ and ‘Perko’. No significant difference was found between the number of larvae in 10 cultivars of *B. napus*. Parasitism of *C. pallidactylus* larvae by *T. obscurator* was not significantly affected by *Brassica* genotypes in 2004 (Fig 1).

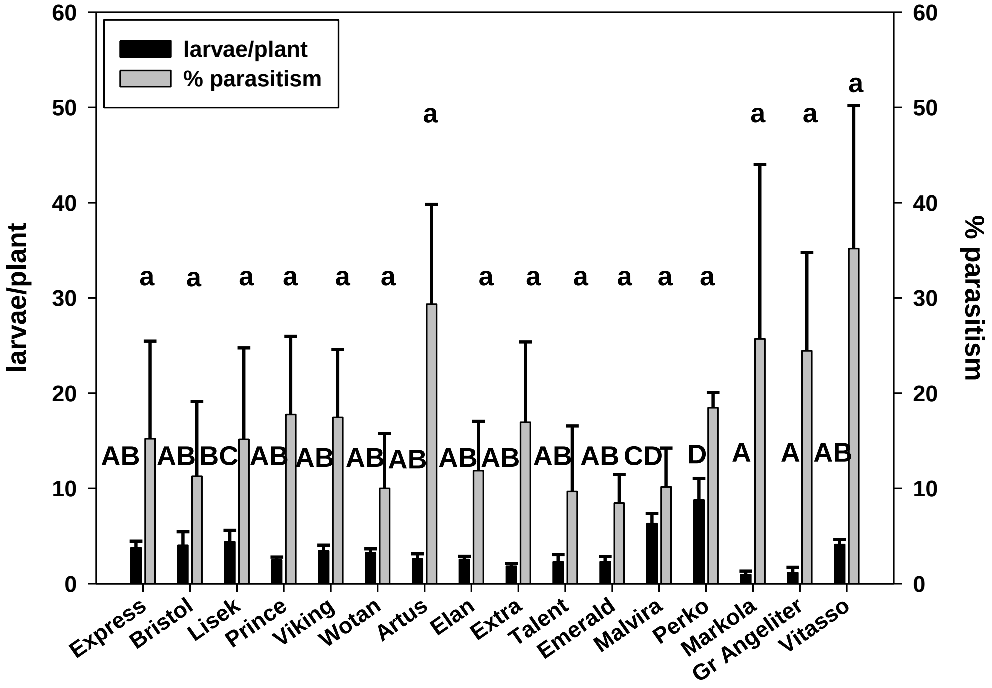

Fig 1: Mean level of infestation (+ SEM) by *C. pallidactylus* and mean level of parasitism (+ SEM) on different *Brassica* genotypes in 2004. Larvae/plant: ANOVA; Tukey-HSD; n = 64; F = 4.72; $p \leq 0.0001$. Parasitism: ANOVA; Tukey-HSD; $p = 0.9555$

In 2005 no significant difference occurred between the level of infestation by cabbage stem weevil in 12 tested genotypes (Fig 2). However, numbers of larvae in the hybrid cultivar ‘Extra’ and the turnip rape ‘Salut’ were very low. The level of parasitism by *T. obscurator* differed significantly between the *B. napus* cultivars ‘Wotan’ (10%) versus ‘Express’, ‘Smart’, ‘Viking’ and ‘Extra’ (25-27%). In all other genotypes, the level of parasitism ranged between 15 and 23%.

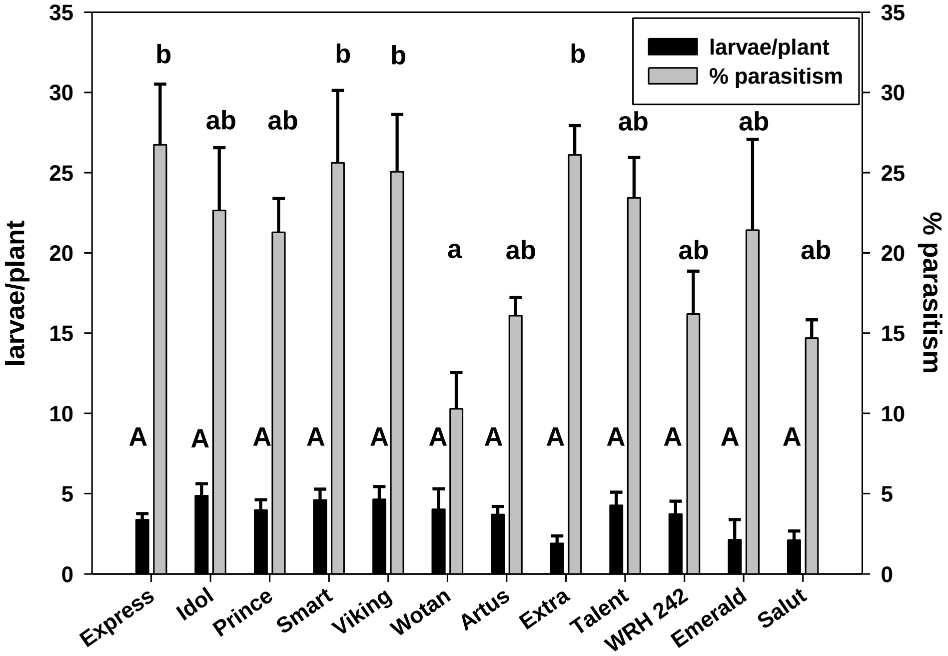

Fig 2: Mean level of infestation (+ SEM) by *C. pallidactylus* and mean level of parasitism (+ SEM) on different *Brassica* genotypes in 2005. Larvae/plant: ANOVA; Tukey-HSD; p = 0.107. Parasitism: ANOVA; Tukey-HSD; n = 48; F = 3.10; p = 0.005

In 2006 only five cultivars were included in the field experiment (Fig 3). The abundance of *C. pallidactylus* was significantly reduced in fodder rape ‘Emerald’ compared to the open pollinating oilseed rape cultivars ‘Prince’ und ‘Wotan’. Parasitism of *C. pallidactylus* in ‘Wotan’ and turnip rape ‘Salut’ was significantly lower than in ‘Express’. Pooled data of the years 2004, 2005 and 2006 showed that the level of parasitism in ‘Wotan’ was relatively low, although but not statistically significant. In contrast, a relative high level of parasitism by *T. obscurator* was observed in all years in the accessions ‘Express’, ‘Prince’ and fodder rape ‘Emerald’ (only 2005 and 2006).

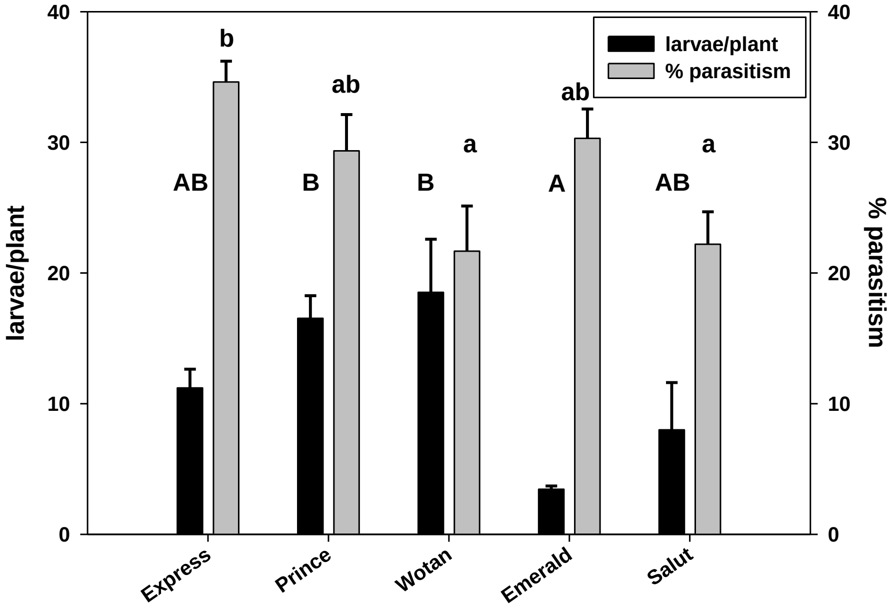

Fig 3: Mean level of infestation (+ SEM) by *C. pallidactylus* and mean level of parasitism (+ SEM) on different *Brassica* genotypes in 2006. Larvae/plant: ANOVA; Tukey-HSD; n = 20; F = 5.47; p = 0.0064. Parasitism: ANOVA; Tukey-HSD; n = 20; F = 4.37; p = 0.0153

Larval abundance and parasitism of C. napi

In 2004, the abundance of *C. napi* larvae was significantly reduced in *B. juncea* 'Vitasso' and *B. oleracea* 'Markola' in comparison to *B. napus* fodder rape 'Emerald' (Fig 4). No significant difference occurred between cultivars of oilseed rape and turnip rape. No parasitized host larvae were found in *B. rapa* cv 'Malvira' (Fig 4), and a significantly lower rate of parasitism was determined in *B. rapa* cv 'Perko' in comparison to *B. oleracea* 'Markola'.

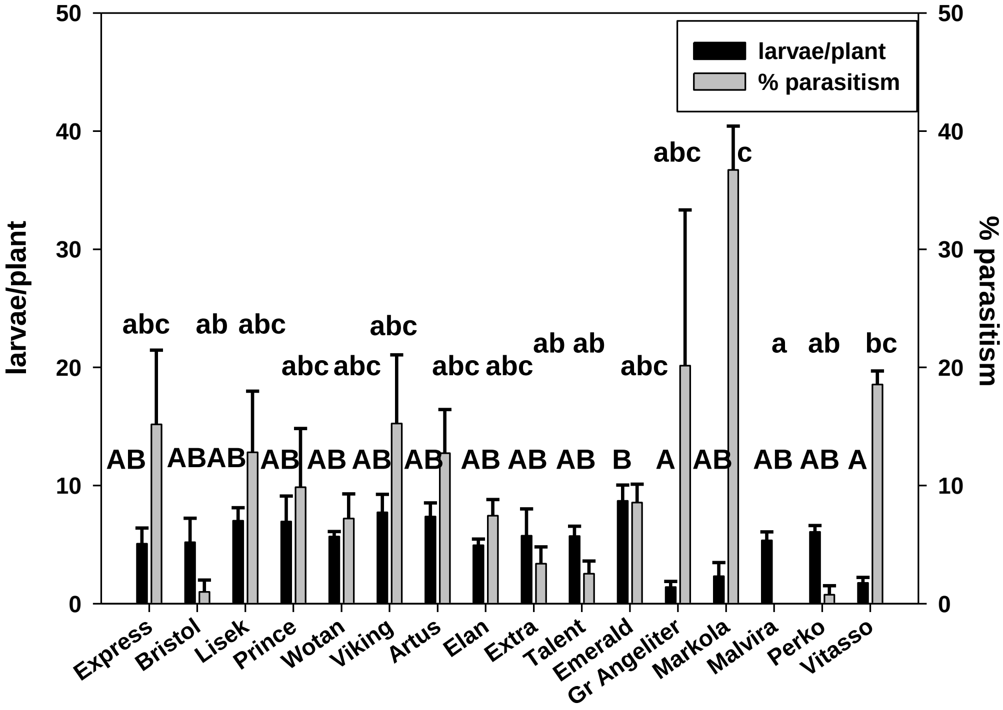

Fig 4: Mean level of infestation (+ SEM) by *C. napi* and mean level of parasitism (+ SEM) on different *Brassica* genotypes in 2004. Larvae/plant: ANOVA; Tukey-HSD; n = 64; F = 2.67; $p \leq 0.005$. Parasitism: ANOVA; Tukey-HSD; n = 64; F = 4.31; $p \leq 0.0001$

In 2005 the level of infestation by *C. napi* was significantly lower on 'Salut', 'Extra' and 'Viking' compared to 'Wotan', fodder rape 'Emerald' and WRH 242 (Fig 5). The level of parasitism of *C. napi* by *T. fulvipes* on *B. rapa* cv 'Salut' (4%) was significantly lower than on all other accession except 'Emerald'. 'Idol' showed an exceptionally high rate of parasitism in 2005 (27%).

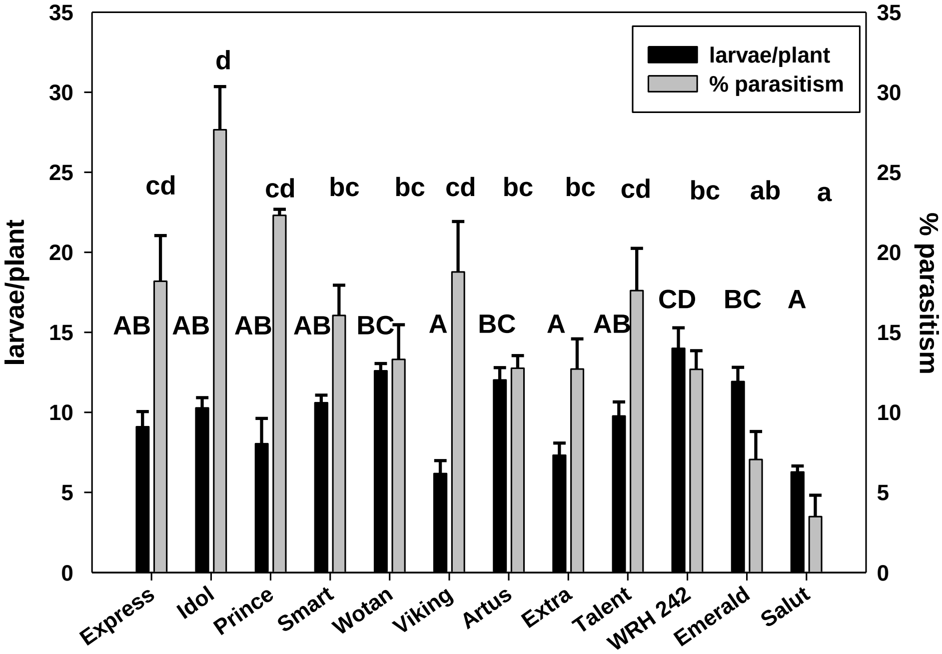

Fig 5: Mean level of infestation (+ SEM) by *C. napi* and mean level of parasitism (+ SEM) on different *Brassica* genotypes in 2005. Larvae/plant: ANOVA; Tukey-HSD; n = 48; F = 8.19; p ≤ 0.0001. Parasitism: ANOVA; Tukey-HSD; n = 48; F = 10.12; p ≤ 0.0001

In 2006 the abundance of *C. napi* larvae was significantly lower in *B. napus* cv 'Express', 'Prince' and B. rapa cv 'Salut' than in cv 'Wotan' and fodder rape cv 'Emerald' (Fig 6). Parasitism of *C. napi* in *B. rapa* cv 'Salut' was significantly lower than in *B. napus* cv 'Prince'.

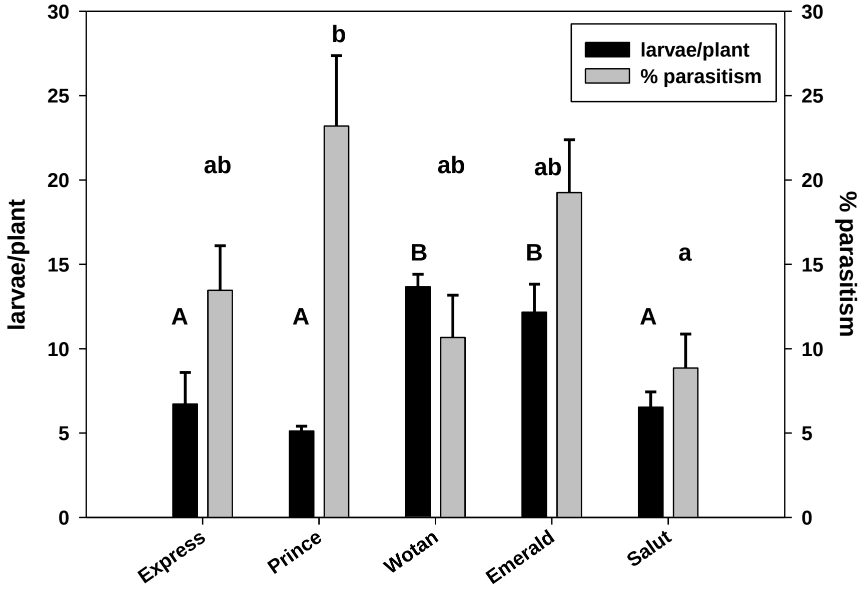

Fig 6: Mean level of infestation (+ SEM) by *C. napi* and mean level of parasitism (+ SEM) on different *Brassica* genotypes in 2006. Larvae/plant: ANOVA; Tukey-HSD; n = 20; F = 9.40; p = 0.0005. Parasitism: ANOVA; Tukey-HSD; n = 20; F = 3.88; p = 0.0233

Relationship between density of ceutorynchid host larvae and parasitism

In all years, the number of *T. obscurator* per m^2 was significantly correlated to the number of *C. pallidactylus* larvae per m^2 (Table 3). Similarly, the number of *T. fulvipes* per m^2 was significantly correlated to the number of *C. napi* per m^2 (Table 3). However, percentage parasitism of *C. napi* and *C. pallidactylus* by *T. fulvipes* and *T. obscurator*, respectively, showed no significant relationship to host abundance in all years (Table 4).

Table 3. Effect of abundance of ceutorynchid host larvae per m^2 on density of *T. obscurator* and *T. fulvipes* per m^2 on different *Brassica* species and cultivars at Goettingen in 2004 – 2006

	Year	**n**	**F**	**r^2**	**y**	**$p \leq 0.05$**
C. pallidactylus	2004	64	25.97	0.295	3.44 + 0.14 x	*
C. pallidactylus	2005	48	100.34	0.686	-2.43 + 0.23 x	*
C. pallidactylus	2006	20	74.69	0.806	1.78 + 0.26 x	*
C. napi	2004	64	28.08	0.312	-13.18 + 0.13 x	*
C. napi	2005	48	17.42	0.275	15.99 + 0.11 x	*
C. napi	2006	20	6.95	0.279	5.44 + 0.08 x	*

* - $p \leq 0.05$

Table 4. Effect of abundance of ceutorynchid host larvae per m^2 on level of parasitism (%) of *T. obscurator* and *T. fulvipes* on different *Brassica* species and cultivars at Goettingen in 2004 – 2006

	Year	**n**	**F**	**r^2**	**y**	**$p \leq 0.05$**
C. pallidactylus	2004	64	0.61	0.01	22.30 – 0.02 x	ns
C. pallidactylus	2005	48	1.96	0.04	24.59 + 0.02 x	ns
C. pallidactylus	2006	20	0.29	0.02	32.39 + 0.01 x	ns
C. napi	2004	64	2.01	0.03	18.86 - 0.01 x	ns
C. napi	2005	48	0.02	0.00	21.82 + 0.00 x	ns
C. napi	2006	20	1.86	0.09	25.57 – 0.04 x	ns

ns - not significant

Effect of plant architecture on parasitism

The relationship between plant growth parameters (diameter and length of main stem, number of leaves and number of lateral racemes per main stem) and the rate of parasitism was analysed in order to estimate the role of plant architecture on host finding and host accession by the parasitoids *T. fulvipes* and *T. obscurator*.

The parasitism of *T. obscurator* was not significantly affected by plant architecture during the experimental years. For *T. fulvipes* an effect of plant parameters was only found in 2004 (Table 5). Regression analysis of parasitism rates of *C. napi* showed significant relationships to plant growth parameters only in one of three experimental years. In 2004 both, stem length and the number of racemes per main stem were significantly negative related to parasitism. The low coefficients of determination (r^2) indicate that other factors than plant architecture may have higher impact on the level of parasitism within plants.

Table 5. Relationship between plant architecture and level of parasitism of *C. napi* by *T. fulvipes* at Goettingen in 2004

Year	**Plant parameter**	**F**	**r^2**	**y**	**p**
2004	Stem length	22.87	0.27	31.94 - 0.29x	< 0.0001
n = 64	Number of racemes	5.59	0.08	23.73 – 1.31x	0.021

Discussion

In this study the relationship between the *Brassica* host plant and the rate of larval parasitism of the ceutorynchid stem weevils *C. napi* and *C. pallidactylus* by the parasitoids *T. fulvipes* and *T. obscurator*, respectively, was investigated.

The level of infestation by ceutorynchid host larvae was significantly affected by *Brassica* host plant. *C. napi* and *C. pallidactylus* have a wide range of brassicaceous host plants (Alford *et al.* 2003). Different susceptibilities of oilseed rape cultivars, turnip rape and brassicaceous species were reported by Eickermann & Ulber (2009a). The overall abundance level of *C. pallidactylus* increased from 2004 and 2005 (3 larvae/plant) to 2006 (12 larvae/plant), whereas peak abundance of *C. napi* occurred in 2005 (10 larvae/plant). There was a slight tendency of increasing levels of parasitism with increasing host density, but parasitism did not show a significant density-dependant response in all years.

During three experimental years, the mean level of parasitism of *C. pallidactylus* by *T. obscurator* was higher than parasitism of *C. napi* by *T. fulvipes*. This is in agreement with reports in literature for this region (Ulber 2003): Parasitism of *C. pallidactylus* was found to vary between 14% and 52% at Goettingen (Ulber 2003; Ulber & Fischer 2006) and between 40% and 70% in Austria (Kraus & Kromp 2002). However, a comparison between the years is biased by the change of cultivars.

The results of this study showed that the three-trophic-level system can be significantly affected by the *Brassica* host plant. Similar effects have been found for other specialist or generalist pest species and their parasitoids (Vet & Dicke 1992; Fox *et al.* 1996).

The species of *Brassica* host plant had a significant effect on larval parasitism of stem weevils. For *T. obscurator* high levels of parasitism were found on oilseed rape cultivars 'Express', 'Prince', fodder rape 'Emerald' and in 2004 in cultivars of *B. oleracea* and *B. juncea*. In contrast, low levels of parasitism were detected in the oilseed rape cv 'Wotan' and the turnip rape cv 'Salut'. Similarly, cultivars of turnip rape showed low rates of parasitism by *T. fulvipes*. There was no consistent difference between the parasitism rates of *C. pallidactylus* and *C. napi* in open pollinating and hybrid cultivars of oilseed rape.

Our study for the first time has determined a multitrophic interaction between *Brassica* host plant, stem-boring pests and parasitoids on crops of oilseed rape. Close interactions within multitrophic systems of *Brassica* crops have been shown for the leaf feeding diamondback moth, *Plutella xylostella* (L.), and its parasitoid *Diadegma insulare* (Cresson) (Sarfraz *et al.* 2008). The parasitism by larval endoparasitoid, *Phradis morionellus* Holmgr., of pollen beetle, *Meligethes aeneus* (Fabr.), was not significantly different between cultivars of *Sinapis alba* and *B. napus* (Billqvist & Ekbom 2001a). In contrast, parasitism of pollen beetle, by the parasitoid *Diospilus capito* Nees, was significantly increased on host plants of *S. alba* compared to *B. napus* (Billqvist & Ekbom 2001b).

Host location and host acceptance of parasitoids on *Brassica* are mainly affected by two cues: plant architecture and plant volatiles (Schoonhoven *et al.* 1998). In our study several factors of plant architecture were assessed for possible effects on parasitism: A significantly negative influence of stem length and number of lateral racemes on parasitism by *T. fulvipes* was only determined in one year. On larger stems and increased number of racemes, host location by *T. fulvipes* might have biased due to larger areas of stem that females have when searching for host larvae. Additionally, host larvae can use more space and refuges for dispersal within the pith of larger stems.

No significant effect of plant architecture on *T. obscurator* was detected. Plant architecture can vary widely between cultivars (Eickermann & Ulber 2009b) and is influenced by environmental conditions. This might be the reason why parasitism rate was not significantly related to parameters of plant growth in all years. A close interaction between plant development, host larval abundance and migration of Tersilochinae to crops of oilseed rape had been shown by Ulber & Nitzsche (2006).

In our study a high level of parasitism by *T. obscurator* and *T. fulvipes* was detected in *B. juncea* and in cultivars of *B. oleracea*, which began to flower three weeks later than cultivars of oilseed rape. Late flowering plants might have attracted adults of Tersilochinae much longer for feeding on pollen. A high density of food source is assumed to retain parasitoids and increase parasitism (Lewis *et al.* 1998). In contrast, very low levels of parasitism on the three cultivars of turnip rape in all years might have resulted from the very early flowering time of turnip rape. In this case, flowering was completed too early to provide pollen for the attraction of adult parasitoids. In trap crop experiments, *T. obscurator* was more abundant in field plots of oilseed rape than in plots of turnip rape (Barari *et al.* 2005). Additionally, distribution of parasitoids within the field can be affected by external factors like wind direction (Williams *et al.* 2007) and inter-specific interaction (Ferguson *et al.* 2006).

An impact of secondary plant chemicals to herbivores and their natural enemies has been reported several times (Pickett *et al.* 1999; Sznajder & Harvey 2003; Gols *et al.* 2007). The most important secondary plant metabolites of *Brassica* species, the glucosinolates and their breakdown products, play an important role in mediating the plant-pest-interaction (Rask *et al.* 2000). Significant effects of glucosinolates in tritrophic systems have been shown in field trials with the parasitoid *Diaeretiella rapae* (McIntosh) of *Brevicoryne brassicae* (L.) on *B. oleracea* cultivars (Bradburne & Mithen 2000). Enhanced production of but-3-enyl isothiocyanate within plants increased attraction of the parasitoid.

Further, orientation of *D. rapae* to the aphid species, *Lipaphis erysimi* (Kalt.), was increased by but-3-enyl isothiocyanate as well (Blande *et al.* 2007). This indicates that host location of the parasitoid is mainly mediated by plant volatiles, namely the isothiocyanates. *T. obscurator* and *T. fulvipes* have been shown to be strongly attracted by 2-phenylethyl isothiocyanate (Ulber & Wedemeyer 2006). Adults *C. pallidactylus* and *C. napi* were also attracted by this isothiocyanate (Walczak *et al.* 1998). The release of volatiles from plants of *Brassica* can be affected by the phenological stage, thereby influencing host finding of parasitoids (Jönsson *et al.* 2005). In our study, observations were focused only on the effect of host plant genotype on the level of parasitism, isothiocyanates and other plant volatiles have not been analyzed. Other influences of host plants on the performance, sex ratio and biomass of parasitoids have been shown in other tritrophic systems, e.g. for the parasitoid *Diadegma semiclausum* Hellen of *P. xylostella* on *B. oleracea* and *S. alba* (Gols *et al.* 2007) or for the predator *Chrysoperla carnea* (Stephens) on different accessions of *B. oleracea* (Reddy *et al.* 2004).

Further studies are needed to determine the mechanisms, how host plant traits affect parasitism of stem-mining weevils in crops of *Brassica*. The manipulation of plant attributes to promote the level of parasitism, in combination with plant resistance to pests, could play an important role in integrated pest management strategies.

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References

Alford DV, Nilsson C & B Ulber (2003): Insect pests of oilseed rape crops. In: DV Alford (ed), *Biocontrol of Oilseed Rape Pests*. Blackwell Science, Oxford, 9-41.

Barari H, Cook SM, Clark SJ & IH Williams (2005): Effect of a turnip rape (*Brassica rapa*) trap crop on stem-mining pests and their parasitoids in winter oilseed rape (*Brassica napus*). Biological Control **50**, 69-86.

Billqvist A & B Ekbom (2001a): The influence of host plant species on parasitism of pollen beetles (*Meligethes* ssp.) by *Phradis morionellus*. Entomologia Experimentalis et Applicata **98**, 81–92.

Billqvist A & B Ekbom (2001b): Effects of host plant species on the interaction between the parasitic wasp *Diospilus capito* and pollen beetle (*Meligethes* spp.). Agricultural and Forest Entomology **3**, 147-152.

Blande JD, Pickett JA & GM Poppy (2007): A comparison of semiochemically mediated interactions involving specialist and generalist Brassica-feeding aphids and the braconid parasitoid Diaeretiella rapae. Journal of Chemical Ecology **33**, 767-779.

Bradburne RP & R Mithen (2000): Glucosinolate genetics and the attraction of the aphid parasitoid *Diaeretiella rapae* to *Brassica*. Proceedings of the Royal Society **267**, 89-95.

Büchi R & S Roos-Humbel (1991): Nützlinge reduzieren die Zahl der Rapsschädlinge. Landwirtschaft Schweiz **4**, 69-73.

Eickermann M & B Ulber (2009a): Screening of oilseed rape and other brassicaceous genotypes for susceptibility to cabbage stem weevil (*Ceutorhynchus pallidactylus* (Marsh.). Submitted.

Eickermann M & B Ulber (2009b): Resynthesized lines and cultivars of *Brassica napus* L. as sources of resistance to cabbage stem weevil (*Ceutorhynchus pallidactylus* (Mrsh.)). Submitted.

Ferguson AW, Barari H, Warner DJ, Campbell JM, Smith ET, Nigel P, Watts NP & IH Williams (2006): Distributions and interactions of the stem miners *Psylliodes chrysocephala* and *Ceutorhynchus pallidactylus* and their parasitoids in a crop of winter oilseed rape (*Brassica napus*). Entomologia Experimentalis et Applicata **119**, 81–92.

Fox LR, Kester KM & J Eisenbach (1996): Direct and indirect responses of parasitoids to plants: sex ratio, plant quality and herbivore diet breadth. Entomologia Experimentalis et Applicata 80, 289-292.

Gols R, Raaijmakers CE, van Dam NM, Dicke M, Bukovinszky T & JA Harvey (2007): Temporal changes affect plant chemistry and tritrophic interactions. Basic and Applied Ecology **8**, 421-433.

Jourdheuil P (1960): Influence de quelques facteurs écologiques sur les fluctuations de population d`une biocénose parasitaire: étude relative à quelques Hyménoptères

(Ophioninae, Diospilinae, Euphorinae) parasites de divers Coléoptères inféodés aux Crucifères. Annales des Épiphyties **11**, 445-658.

Johnen A, Ulber B, Williams IH, Klukowski Z & C Nielsson (2006): Prüfung der Vorhersagegenauigkeit der proPlant-Phänologiemodelle für wichtige Schädlinge im Winterraps unter heterogenen klimatischen Bedingungen in Europa. Mitteilungen der Biologischen Bundesanstalt für Land- und Forstwirtschaft **400**, 277.

Jönsson M, Lindkvist A & P Anderson (2005): Behavioural responses in three ichneumonid pollen beetle parasitoids to volatiles emitted from different phenological stages of oilseed rape. Entomologia Experimentalis et Applicata **111**, 363-369.

Klingenberg A & B Ulber (1994): Untersuchungen zum Auftreten der Tersilochinae (Hym., Ichneumonidae) als Larvenparasitoide einiger Rapsschädlinge im Raum Göttingen 1990 und 1991 und zu deren Schlupfabundanz nach unterschiedlicher Bodenbearbeitung. Journal of Applied Entomology **117**, 220-227.

Klukowski K (2006): The impact of insecticide application and timing on parasitoid activity and levels of parasitism. CD-Rom Proceedings of the International Symposium „Integrated Pest Management in Oilseed Rape Pests“, Goettingen, Germany, 3-5 April 2006.

Kraus P & B Kromp (2002): Parasitization rate of the oil seed rape pests Ceutorhynchus napi, Ceutorhynchus pallidactylus (Coeloptera, Curculionidae) and Meligethes aeneus (Coeleoptera, Nitidulidae) by Ichneumonids in several localities of Eastern Austria. IOBC/wprs Bulletin **25** (2), 117-122.

Lewis WJ, Stapel JO, Cortesero AM & K Takasu (1998): Understanding how parasitoids balance food and host needs: Importance to biological control. Biological Control **11**, 175-183.

Lill JT, Marquis RJ & RE Ricklefs (2002): Host plants influence parasitism of forest caterpillars. Nature **417**, 170-173.

Neumann N & B Ulber (2009): Influence of insecticide application on host finding of *Tersilochus obscurator* (Hym.: Ichneumonidae). IOBC/wprs Bulletin, in prep.

Nitzsche O & B Ulber (1998): Einfluß der Bodenbearbeitung nach Winterraps auf die Schlupfabundanz der Parasitoiden von *Ceutorhynchus napi*, *Ceutorhynchus pallidactylus* und *Meligethes aeneus*. Mitteilungen der Biologischen Bundesanstalt **357**, 62-63.

Pickett JA, Smiley DWM & CM Woodcock (1999): Secondary metabolites in plant-insect-interactions: Dynamic systems of induced and adaptive responses. In: JA Callow (ed), *Advances in Botanical Research* **30**. Academic Press, Oxford, 91-115.

Rask L, Andréasson E, Ekbom B, Eriksson S, Pontoppidan B & J Meijer (2000): Myrosinase: gene family evolution and herbivore defense in Brassicaceae. Plant Molecular Biology **42**, 93-113.

Reddy GVP, Tabone E & MT Smith (2004): Mediation of host selection and oviposition behavior in the diamondback moth *Plutella xylostella* and its predator *Chrysoperla carnea* by chemical cues from cole crops. Biological Control **29**, 270-277.

Sarfraz M, Dosdall LM & BA Keddie (2008): Host plant genotype of the herbivore *Plutella xylostella* (Lepidoptera: Plutellidae) affects the performance of its parasitoid *Diadegma insulare* (Hymneoptera: Ichneumonidae). Biological Control **44**, 42-51.

Schoonhoven LM, Jermy T & JJA van Loon (1998): Host plant selection: how to find a host plant. In: *Insect-Plant Biology – From physiology to evolution*. Chapman & Hall, London, 121-154.

Sedivy J (1983): Tersilochinae as parasitoids of insect pests of winter rape (Hymenoptera: Ichneumonidae). Contributions of the American Entomological Institute **20**, 266-276.

Sznajder B & JA Harvey (2003): Second and third trophic level effects of differences in plant species reflect dietary specialisation of herbivores and their endoparasitoids. Entomologia Experimentalis et Applicata **109**, 73-82.

Ulber B (2003): Parasitoids of ceutorynchid stem weevils. In: DV Alford (ed), *Biocontrol of Oilseed Rape Pests*. Blackwell Science, Oxford, 87-95.

Ulber B & K Fischer (2006): Effect of plant density and host plant architecture on the abundance and with-in plant distribution of stem-mining pests and the level of parasitism. CD-Rom Proceedings of the International Symposium „Integrated Pest Management in Oilseed Rape Pests“, Goettingen, Germany, 3-5 April 2006.

Ulber B & O Nitzsche (2006): Phenology of parasitoids (Hym., Ichneumonidae, Tersilochinae) of oilseed rape pests in northern Germany in 1995-1997. IOBC/wprs Bulletin **29** (7), 173-179.

Ulber B & R Wedemeyer (2006): Responses of *Tersilochus microgaster* and *Tersilochus obscurator* (Hymenoptera: Ichneumonidae) to volatile 2-phenylethyl-isothiocyanate. CD-Rom Proceedings of the International Symposium „Integrated Pest Management in Oilseed Rape Pests“, Goettingen, Germany, 3-5 April 2006.

Vet LEM & M Dicke (1992): Ecology of infochemical use by natural enemies in a tritrophic context. Annual review of Entomology **37**, 141-172.

Walczak B, Kelm M, Klukowski Z, Smart LE, Ferguson AW & IH Williams (1998): The effect of trap design and 2-phenylethyl isothiocyanate on catches of stem weevils (*Ceutorhynchus pallidactylus* Marsh. and *C. napi* Gyll.) in winter oilseed rape. IOBC/wprs Bulletin **21** (5), 141-146.

Williams IH, Frearson DJT, Barari H & A McCartney (2007): First field evidence that parasitoids use upwind anemotaxis for host-habitat location. Entomologia Experimentalis et Applicata **123**, 299-307.

Chapter V - General discussion

The cabbage stem weevil, *Ceutorhynchus pallidactylus* (Mrsh.) (Coleoptera: Curculionidae), is a major pest of oilseed rape in Central Europe (Alford *et al.* 2003). It is adapted to a wide range of wild and cultivated brassicaceous plant species, such as *B. napus* (Broschewitz 1985; Mrówczyński 1998), *B. rapa* (Büchi 1990), *B. nigra* (L.), Indian mustard, *B. juncea* (L.), radish, *Raphanus* spp., white mustard, *Sinapis alba* (L.) (Winfield 1961), sea-kale, *Crambe maritime* L. and field penny-cress, *Thlaspi arvense* L. (Speyer 1921; Günthart 1949). Resistant cultivars of oilseed rape are highly demanded for minimizing the number of insecticide applications in integrated pest management systems (Williams 2004). The development of plant resistance to insect pests in winter oilseed rape (*Brassica napus* L. *var. oleifera* Metzg.) is still in the early stages. So far, only a few studies have been conducted to identify sources of resistance against *C. pallidactylus* in brassicaceous plants (Winfield 1961; Mrówczyński 1998; Lühs *et al*. 2003; Mrówczyński *et al.* 2006), showing only minor differences of susceptibility. In addition, the potential mechanisms of resistance are still unknown.

The main objectives of this study were as follows:

1. to develop laboratory screening methods for identification and quantification of resistance to cabbage stem weevil (*Ceutorhynchus pallidactylus*) in brassicaceous species
2. to screen a large assortment of cultivars of oilseed rape, breeding lines, resynthesized rapeseed lines and other species of Brassicaceae under controlled conditions in growth chambers
3. to validate the resistance of selected genotypes under field conditions
4. to evaluate the influence of host plant genotype on parasitism of ceutorynchid stem weevils

Screening methods

Screening tests based on leaf feeding by herbivorous insect species have been applied successfully for testing the susceptibility of plants to various pests (Risch 1985), e.g. for bean leaf beetle, *Cerotoma trifurcate* (Forster) (Bradshaw *et al.* 2007), *Helicoverpa armigera* (Hübner) (Sharma *et al.* 2005), *Mamestra configurata* Walker (Ulmer *et al.* 2001) and cabbage stem flea beetle, *Psylliodes chrysocephala* L. (Bartlet & Williams

1991). For quantification of the leaf area removed by herbivores, techniques like computer scanning or computer-aided video image analysis can be used (O`Neal *et al.* 2002).

Screening methods based on oviposition in dual-choice tests are able to detect host finding and host acceptance of adult pest species and performance of their larvae as well. They have been applied successfully for testing the susceptibility of plants to root maggots, *Delia* spp. (Dosdall *et al.* 1994), pollen beetle, *Meligethes aeneus* (Fabr.) (Ekbom & Borg 1996) and diamondback moth, *Plutella xylostella* (L.) (Sarfraz *et al.* 2007). The screening method and the way the test plants are offered to the insects can have significant effects on the results. Generalists are less attracted by specific volatiles emitted from plants than specialist herbivores (Risch 1985). Using detached leaves or wounded plants in screening tests can affect the cues responsible for attraction of specialist species (Farrell 1977). While no-choice tests are only able to discriminate between plants of remarkably differing host plant quality (Risch 1985), multi-choice tests which include cues relevant for host location and selection are more favourable for detecting minor differences between plant susceptibility (Bartlet & Williams 1991).

Two new screening methods have been developed in this study which can be used to quantify the amount of leaf feeding of *C. pallidactylus* in no-choice tests and the oviposition preference in dual-choice tests, respectively. Both methods are suitable for rapid screening of a large assortment of genotypes and for assessment of significant differences between host suitability. A comparison between the results of both screenings methods showed a significant positive relationship. However, both methods address only the antixenosis effect of plant genotypes. To characterize antibiosis effects, parameters of larval performance such as larval mortality or larval weight need to be determined in greater detail.

Validation of the results of laboratory screening methods under field conditions showed significant correlations. The coefficient of determination differed between both screening methods. The results of the dual-choice oviposition screening test were more consistent with results of field studies (r = 66%), as this test method is probably better adapted to the process of host finding and host selection in the field. Relevant cues like plant morphology or plant volatiles are included. Thus, the oviposition preference test appears to be superior to the feeding test for estimating the level of host suitability.

Host plant susceptibility

Research on host plant resistance in plant genotypes relies mainly on laboratory assays, with adult stages of pest insects. Screening commonly includes at first commercial varieties of the crop plant, followed by breeding lines, land races and wild genotypes (Ellis & Kift 2003). A wide spectrum of brassicaceous plant species was tested in this study for susceptibility to cabbage stem weevil. In dual-choice and no-choice tests various cultivars or lines of oilseed rape, resynthesized rapeseed lines, cultivars of *B. oleracea* and wild species of Brassicaceae showed significantly less susceptibility to *C. pallidactylus*. Considerable differences were detected not only between the host quality of genotypes of oilseed rape vs. other species and genera of Brassicaceae, but also between different cultivars of oilseed rape, which have only a low genetic variability (Becker *et al*. 1995). In studies comparing the leaf feeding of pests on modern lines of oilseed rape significant differences between these lines were also observed with cabbage flea beetle, *Phyllotreta cruciferae* (Goeze) (Bodnaryk & Palaniswamy 1990) and diamondback moth (Sarfraz *et al.* 2007). The leaf area consumed by *C. pallidactylus* on different genotypes of *B. oleracea* tested showed significant differences. Similarly, the host of aphids for various accessions of *B. oleracea* differed significantly (Singh & Ellis 1993; Cole 1997).

Among the other species of *Brassica* tested for their susceptibility to *C. pallidactylus* in the laboratory, only three genotypes of *Brassica fruticulosa* Cyrillo showed a significant reduction of oviposition compared to ‘Express’. *B. fruticulosa* had been selected as a source of resistance to cabbage aphid, *Brevicoryne brassicae* L., cabbage root fly, *Delia radicum* (L.) and other pest species of brassicaceous plants (Cole 1997; Jensen *et al.* 2002; Pink *et al.* 2003; Felkl *et al*. 2005). So far, no resistance loci have been identified, but a polygenic resistance is assumed (Pink *et al.* 2003). *Crambe abyssinica* Hochst. ex R.E. Frees was found to be little attractive to pollen beetle, *Meligethes aeneus* (Fabr.) (Ekbom & Borg 1996), cabbage seedpod weevil, *Ceutorhynchus obstrictus* (Marsham) (Kalischuk & Dosdall 2004; Cárcamo *et al.* 2007) and cabbage flea beetles, *Phyllotreta* spp. (Henderson *et al.* 2004), respectively. The factors causing this non-preference trait have not been yet determined. For pollen beetle the form of flower buds is supposed to effect oviposition preferences (Ekbom & Borg 1996). Chemical cues (Palaniswamy & Lamb 1992) and plant morphology (Kalischuk & Dosdall 2004) are known to affect the host preference of flea beetles and cabbage seedpod weevil, respectively. Plant architecture is significantly positive correlated with oviposition of *C. pallidactylus*, i.e. length of main stem (Ferguson *et al.* 2003) and the number of lateral racemes and leaves on main stems (Eickermann & Ulber 2009, in prep.). However, plant architecture appears not to be the most important factor in the plant-weevil-interaction, and other cues like plant volatiles may have higher impact.

The gene pool utilized for elite oilseed rape breeding has been eroded as major emphasis was given to specific oil and seed quality, but not traits like resistance (Seyis *et al*. 2000). As a consequence, genetic variability within modern cultivars is highly reduced. Resynthesized oilseed rape has potential to enhance the genetic diversity significantly (Becker *et al*. 1995); they were found to provide new sources of resistance to plant diseases (Diederichsen & Sacristan 1996; Rygulla *et al.* 2007) and possibly to pest species as well (Seyis *et al*. 2000). In our study, we have proved for the first time that some resyntheses exhibit a significantly lower susceptibility to *C. pallidactylus* in laboratory and field experiments. Characteristics of resyntheses are based on their progenitors (Girke 2002). Different cultivars of Chinese cabbage, *B. rapa ssp. pekinensis*, were used as progenitor of resyntheses H 226, L 16 and L 122, which were significantly less susceptible to cabbage stem weevil. However, further experiments are required to determine potential resistance factors of *B. rapa ssp. pekinensis* in detail.

Influence of glucosinolates

Plant species belonging to the family Brassicaceae are able to biosynthesize glucosinolates (GSLs). GSLs and their breakdown products are considered to play an important role in plant-insect-interactions (Mithen 1992; Wallsgrove *et al*. 1999). The content and profile of GSLs within plants depend on several factors, such as genetic, environmental, agronomic and attack by herbivores or diseases (Bartlet *et al.* 1999; Wallsgrove *et al*. 1999; Textor & Gershenzon 2009). GSLs are subdivided into three different major chemical classes: aliphatic, aromatic and indolic. Their relative concentration can differ significantly between plant species, cultivars and even ecotypes and also interact differently with herbivore species (Textor & Gershenzon 2009). The reduction of the total GSL content in seeds by breeding selection often resulted in a reduction of aliphatic GSLs, whereas the level of indolic GSLs remained approximately constant (Weber *et al.* 1984; Mithen 1992). In general, the majority of specialist pests on oilseed rape were found to be attracted more strongly by indolic GSLs. Only

C. obstrictus responded more clearly to aliphatic GSLs of host plants, e.g. gluconapin and glucobrassicanapin (McCaffrey *et al.* 1999; Ulmer & Dosdall 2006). Additionally, the aromatic GSL sinalbin is assumed to have an antixenotic effect on the larvae of *C. obstrictus* (Dosdall & Kott 2006). In recent studies (Cárcamo *et al.* 2007), an antibiotic effect of aliphatic GSL sinigrin on larvae of cabbage seedpod weevil was detected as well.

In our semi-field experiments, a significant effect of single GSL compounds in *Brassica* cultivars and resynthesized rapeseed lines on the stem injury coefficient by *C. pallidactylus* was found. In resyntheses that were significantly less infested by *C. pallidactylus* larvae the GSL 4-methoxyglucobrassicin was completely missing, whereas the aromatic GSL gluconasturtiin and the indolic GSL glucobrassicin were found only at low or medium concentrations compared to other genotypes tested. Glucobrassicin is the dominant GSL in the leaves during early vegetative growth stages of oilseed rape (Rothe *et al.* 2004). The indolic GSL 4-hydroxyglucobrassicin occurred at a medium concentration in resyntheses L 16, L 122 and H 226. This indicates that host preference of *C. pallidactylus* is affected by a certain balance between single GSL compounds, rather than individual GSLs.

Genotypes of Brassicaceae were found to be significantly less susceptible to *C. pallidactylus* in the laboratory screenings, e.g. honesty, *Lunaria annua* L., false flax, *Camelina alyssum* (L.) and *B. fruticulosa* Cyrillo. These species produce only very low amounts of glucobrassicin and neoglucobrassicin (Schraudolf 1968; Cole 1997). However, *L. annua* also contains alkaloids and saponins which have been described as feeding deterrents for cabbage flea beetle, *Phyllotreta nemorum* L. (Nielsen 1989). The leaves of *C. alyssum* contain the flavonoid quercetin (Onyilagha *et al.* 2003), but the role of these phytochemicals in host finding and host acception of *C. pallidactylus* has not been examined in our experiment. In addition to secondary plant metabolites like GSLs, proteins and carbohydrates can affect host plant quality as well (Gols *et al.* 2007). The specific relation between these plant substances needs to be determined in further experiments.

Multitrophic interactions

The promotion of the parasitism of pests by host plant cues has been discussed in literature (Bottrell *et al.* 1998; Gols & Harvey 2009). So far, this strategy has failed because knowledge on such highly specific interactions is insufficient. However, various cues have been identified to assist host finding of parasitoids like plant architecture (Wang *et al.* 1997), plant growth stages (Gols *et al.* 2007) and allelochemicals (Reddy *et al.* 2004). A significant effect of host plant species on parasitism had been detected so far only with parasitism of pollen beetle by *Diospilus capito* Nees on oilseed rape (Billqvist & Ekbom 2001). *C. pallidactylus* can be attacked by five larval endoparasitoids in Europe (Alford *et al*. 2003) all belonging to the family Ichneumonidae. *Tersilochus obscurator* Aubert is the most important parasitoid in Germany (Ulber 2003, Ulber & Nitzsche 2006). The level of parasitism depends on several external factors such as weather conditions (Johnen *et al*. 2006), plant architecture (Ulber & Fischer 2006), plant volatiles (Jönsson *et al*. 2005) and crop management (Nitzsche & Ulber 1998; Ulber & Fischer 2006). In three-years field experiments, the level of parasitism of *C. napi* by *T. fulvipes* was significantly lower in cultivars of *B. rapa* than in cultivars of oilseed rape. Further, parasitism of cabbage stem weevil by *T. obscurator* was significantly reduced in oilseed rape cv ‘Wotan’. The late flowering period of *B. oleracea* and *B. juncea* cv ‘Vitasso’ compared to oilseed rape might have provided food resources to adult Tersilochinae and increased parasitism of larvae. Late flowering period can affect contents and profiles of GSLs in this specific growth stage that can influence host recognition of parasitoids (Jönsson *et al*. 2005; Gols *et al*. 2007). Interaction between host larvae and parasitoids can differ widely between cultivated and wild Brassicaceae (Kahuthia-Gathu *et al.* 2008).

A significant influence of single GSL compounds, especially indolic and aromatic GSLs, on interaction between host plant and specialist pests have been shown in our study. Similarly, single GSLs seem to play also an important role as cues for host finding of parasitoids as well. 3-butenyl had a direct effect on parasitism of aphids by *Diaeretiella rapae* (McIntosh) (Bradburne & Mithen 2000). Gols *et al.* (2007) found only a weak effect of the GSL content within leaves of *S. alba* and *B. oleracea* on parasitism of *Plutella xylostella* L. by *Diadegma semiclausum* Hellen. The authors concluded that a complex of factors is influencing the tritrophic system. Further studies are needed to determine the role of GSL compounds for the parasitism of ceutorynchid stem miners in oilseed rape in greater detail.

Conclusion

More information is required to determine the potential of genotypes showing low susceptibility to cabbage stem weevil for breeding of modern cultivars of oilseed rape. Because species of the genus *Brassica* have different genomes, hybridization between wild species and *B. napus* is difficult (Ellis *et al*. 1999; Pink *et al*. 2003) and requires specific techniques like embryo rescue (Srivastava *et al*. 2004). Negative effects of crossing high yield cultivars with wild species on yield and quality traits are possible (Allard 1996). Whereas accessions of *B. fruticulosa* are genetically distant from modern oilseed rape, resyntheses promise to be an important source of resistance that can be more easily used for breeding of resistant oilseed rape in near future. Host plant resistance to pest insects is an underutilized strategy in IPM in oilseed rape. A better understanding of plant-insect interaction and the factors affecting host location and host acceptance are major points in breeding of (partly) resistant cultivars to cabbage stem weevil and other pest species of oilseed rape for the future.

References

Alford DV, Nilsson C & B Ulber (2003): Insect pests of oilseed rape crops. In: DV Alford (ed), *Biocontrol of Oilseed Rape Pests*. Blackwell Science, Oxford, 9-41.

Allard, RW (1996): Genetic basis of the evolution of adaptedness in plants. Euphytica **92**, 1-11.

Bartlet E, Kiddle G, Williams I & R Wallsgrove (1999): Wound-induced increases in the glucosinolate content of oilseed rape and their effect on subsequent herbivory by a crucifer specialist. Entomologia Experimentalis et Applicata **91**, 163-167.

Bartlet E & IH Williams (1991): Factors restricting the feeding of the cabbage stem flea beetle (*Psylliodes chrysocephala*). Entomologia Experimentalis et Applicata **60**, 233-238.

Becker HC, Enqvist GM & B Karlsson (1995): Comparison of rapeseed cultivars and resynthesized lines based on allozyme and RFLP markers. Theoretic and Applied Genetics **91**, 62-67.

Billqvist A & B Ekbom (2001): Effects of host plant species on the interaction between the parasitic wasp *Diospilus capito* and pollen beetle (*Meligethes* spp.). Agricultural and Forest Entomology **3**, 147-152.

Bodynaryk RP & P Palaniswamy (1990): Glucosinolate levels in cotyledons of mustard, *Brassica juncea* L. and rape, *B. napus* L. do not determine feeding rates of flea beetle, *Phyllotreta cruciferae* (Goeze). Journal of Chemical Ecology **16**, 2735-2746.

Bottrell DG, Barbosa P & F Gould (1998): Manipulating natural enemies by plant variety selection and modification: A realistic strategy? Annual Review of Entomology **43**, 347-367.

Bradburne RP & R Mithen (2000): Glucosinolate genetics and the attraction of the aphid parasitoid *Diaretiella rapae* to *Brassica*. Proceedings of the Royal Society **267**, 89-95.

Bradshaw JD, Rice MW & JH Hill (2007): No-choice preference of *Cerotoma trifurcata* (Coleoptera: Chrysomelidae) to potential host plants of Bean pod mottle virus (*Comoviridae*) in Iowa. Journal of Economic Entomology **100**, 804-814.

Broschewitz B (1985): Untersuchungen zur Biologie und Schadwirkung des Gefleckten Kohltriebrüsslers (*Ceutorhynchus quadridens* PANZER) am Winterraps (*Brassica napus* L. *var. oleifera* Metzg.). PhD-thesis, Wilhelm-Pieck-Universität, Rostock.

Büchi R (1990): Investigations on the use of turnip rape as trap crop plant to control oilseed rape pests. IOBC/wprs Bulletin **13** (4), 32-39.

Cárcamo H, Olfert O, Dosdall L, Herle C, Beres B & J Soroka (2007): Resistance to cabbage seedpod weevil among selected Brassicaceae germplasm. The Canadian Entomologist **139**, 658-669.

Cole RA (1997): The relative importance of glucosinolates and amino acids to the development of two aphid pests Brevicoryne brassicae and Myzus persicae on wild and cultivated Brassica species. Entomologia Experimentalis et Applicata **85**, 121-133.

Diederichsen E & MD Sacristan (1996): Disease response of resynthesized *Brassica napus* L. lines carrying different combinations of resistance to *Plasmodiophora brassicae* Wor.. Plant Breeding **115**, 5-10.

Dosdall LM, Herbut MJ & NT Cole (1994): Susceptibilities of species and cultivars of canola and mustard to infestation by root maggots (*Delia* spp.) (Diptera: Anthomyiidae). The Canadian Entomologist **126**, 251-260.

Dosdall LM & LS Kott (2006): Introgression of resistance to cabbage seedpod weevil to canola from yellow mustard. Crop Science **46**, 2437-2445.

Eickermann M & B Ulber (2009): Impact of *Brassica* genotypes on infestation and larval abundance of ceutorynchid stem miners (*Ceutorhynchus pallidactylus* (Mrsh.) and *Ceutorhynchus napi* Gyll.). In prep.

Ekbom B & A Borg (1996): Pollen beetle (*Meligethes aeneus*) oviposition and feeding preferences on different host plants species. Entomologia Experimentalis et Applicata **78**, 291-299.

Ellis PR, Pink DAC, Barber NE & A Mead (1999): Identification of high levels of resistance to cabbage root fly, *Delia radicum*, in wild *Brassica* species. Euphytica **110**, 207-214.

Ellis PR & NB Kift (2003): The exploitation of plant resistance in controlling insect pests of vegetable crops. IOBC/wprs Bulletin **26** (3), 47-55.

Farrell JAK (1977): Plant resistance to insects and the selection of resistant lines. The New Zealand Entomogist **6**, 244-261.

Felkl G, Jensen EB, Kristiansen K & SB Andersen (2005): Tolerance and antibiosis resistance to cabbage root fly in vegetable *Brassica* species. Entomologia Experimentalis et Applicata **116**, 65-71.

Ferguson AW, Klukowski Z, Walczak B, Clark SJ, Mugglestone MA, Perry JN & IH Williams (2003): Spatial distribution of pest insects in oilseed rape: implications for integrated pest management. Agriculture, Ecosystems and Environment **95**, 509-521.

Girke A (2002): Neue Genpools aus resynthetisiertem Raps (*Brassica napus* L.) für die Hybridzüchtung. PhD-thesis, Georg-August-Universität, Goettingen.

Gols R, Raaijmakers CE, van Dam NM, Dicke M, Bukovinzky & JA Harvey (2007): Temporal changes affect plant chemistry and tritrophic interactions. Basic and Applied Ecology **8**, 421-433.

Gols R & JA Harvey (2009): Plant-mediated effects in the Brassicacea on the performance and behaviour of parasitoids. Photochemistry Reviews **8**, 187-206.

Günthart E (1949): Beiträge zur Lebensweise und Bekämpfung von *Ceutorhynchus quadridens* PANZ. und *Ceutorhynchus napi* Gyll. mit vielen Beobachtungen an weiteren Kohl- und Rapschädlingen. Mitteilungen der Schweizerischen Entomologischen Gesellschaft **23**, 441-591.

Henderson AE, Hallett RH & JJ Soroka (2004): Prefeeding behavior of the crucifer flea beetle, *Phyllotreta cruciferae,* on host and nonhost crucifers. Journal of Insect Behavior **17**, 17-39.

Jensen EB, Felkl G, Kristiansen K & SB Andersen (2002): Resistance to the cabbage root fly, *Delia radicum*, within *Brassica fruticulosa*. Eupyhtica **124**, 379-386.

Johnen A, Ulber B, Williams IH, Klukowski Z & C Nielsson (2006): Prüfung der Vorhersagegenauigkeit der proPlant-Phänologiemodelle für wichtige Schädlinge im Winterraps unter heterogenen klimatischen Bedingungen in Europa. Mitteilungen der Biologischen Bundesanstalt für Land- und Forstwirtschaft **400**, 277.

Jönsson M, Lindkvist A & P Anderson (2005): Behavioural responses in three ichneumonid pollen beetle parasitoids to volatiles emitted from different phenological stages of oilseed rape. Entomologia Experimentalis et Applicata **111**, 363-369.

Kahuthia-Gathu R, Löhr B & HM Poehling (2008): Effect of common wild crucifer species of Kenya on fitness of two exotic diamondback moth parasitoids, *Cotesia plutellae* and *Diadegma semiclausum*. Crop Protection **27**, 1277-1484.

Kalischuk AR and LM Dosdall (2004): Susceptibilities of seven Brassicaceae species infestation by the cabbage seedpod weevil (Coleoptera: Curculionidae). The Canadian Entomologist **136**, 265-276.

Lühs W, Ulber B, Seyis F, Frauen M, Busch H, Friedt W, Gustafsson M & G Poulsen (2003): Evaluation of a Brassica napus core collection and prospects of its exploitation through rapeseed breeding. 11th International Rapeseed Congress Copenhagen, Denmark, July 2003, **2**, 371-374.

McCaffrey JP, Harmon BL, Brown J, Brown AP & JB Davis (1999): Assessment of *Sinapis alba*, *Brassica napus* and *S. alba* x *B. napus* hybrids (Coleoptera: Curculionidae). Journal of Agricultural Science **132**, 289-295.

Mithen R (1992): Leaf glucosinolate profiles and their relationship to pest and disease resistance in oilseed rape. Euphytica **63**, 71-83.

Mrówczyński M (1998): Studies on pest damage of cultivars and lines of winter oilseed rape. IOBC/wprs Bulletin **21** (5), 153-154.

Mrówczyński M, Wachowiak H, Muśnicki C, Jodłowski M & S Heimann (2006): Damage of registered Polish winter oilseed rape cultivars caused by pests. IOBC/wprs Bulletin **29** (7), 95-96.

Nielsen JK (1989): Host plant relationships of *Phyllotreta nemorum* L. (Coleoptera: Chrysomelidae). II. Various defensive mechanisms in plants and their role in determining the host plant range. Journal of Applied Entomology **107**, 193-202.

Nitzsche O & B Ulber (1998): Einfluß der Bodenbearbeitung nach Winterraps auf die Schlupfabundanz der Parasitoiden von *Ceutorhynchus napi*, *Ceutorhynchus pallidactylus* und *Meligethes aeneus*. Mitteilungen der Biologischen Bundesanstalt **357**, 62-63.

O'Neal ME, Landis DA & R Isaacs (2002): An inexpensive, accurate method for measuring leaf area and defoliation through digital image analysis. Journal of Economic Entomology **95**, 1190-1194.

Onyilagha J, Bala A, Hallett R, Gruber M, Soroka J & N Westcott (2003): Leaf flavonoids of the cruciferous species, Camelina sativa, Crambe spp., Thlaspi arvense and several other genera of the family Brassicaceae. Biochemical Systematics and Ecology **31**, 1309-1322.

Palaniswamy P & RJ Lamb (1992): Host preferences of the flea beetles *Phyllotreta cruciferae* and *P. striolata* (Coleoptera: Chrysomelidae) for crucifer seedlings. Journal of Economic Entomology **85**, 743-752.

Pink DAC, Kift NB, Ellis PR, McClement SJ, Lynn J & GM Tatchell (2003): Genetic control of resistance to the aphid Brevicoryne brassicae in the wild species Brassica fruticulosa. Plant Breeding **122**, 24-29.

Reddy GVP, Tabone E & MT Smith (2004): Mediation of host selection and oviposition behavior in the diamondback moth *Plutella xylostella* and its predator *Chrysoperla carnea* by chemical cues from cole crops. Biological Control **29**, 270-277.

Risch SJ (1985): Effects of induced chemical changes on interpretation of feeding preference tests. Entomologia Experimentalis et Applicata **39**, 81-84.

Rothe R, Hartung H, Marks G, Bergmann H, Götz R & F Schöne (2004): Glucosinolate contents in vegetative tissues of winter rape cultivars. Journal of Applied Botany **78**, 41-47.

Rygulla W, Snowdon RJ, Eynck C, Koopmann B, von Tiedemann A, Lühs W & W Friedt (2007): Broadening the genetic basis of *Verticillium longisporum* resistance in *Brassica napus* by interspecific hybridization. Phytopathology **97**, 1391-1396.

Sarfraz M, Dosdall LM & BA Keddie (2007): Resistance of some cultivated Brassicaceae to infestations by Plutella xylostella (Lepidoptera: Plutellidae). Journal of Economic Entomology **100**, 215-224.

Schraudolf H (1968): Untersuchungen zur Verbreitung von Indolglucosinolaten in Cruciferen. Experientia **24**, 434-435.

Seyis F, Friedt W & W Lühs (2000): Resynthese-Raps (*Brassica napus* L.) als genetische Ressource für die Qualitäts- und Ertragszüchtung. In: Hammer K & T Gladis (eds), *Nutzung genetischer Ressourcen – ökologischer Wert der Biodiversität*. Schriften zu Genetischen Ressourcen **16**. ZADI, Bonn, 91-112.

Sharma HC, Pampapathy G, Dhillon MK & JT Ridsdill-Smith (2005): Detached leaf assay to screen for host plant resistance to *Helicoverpa armigera*. Journal of Economic Entomology **98**, 556-576.

Singh R & PR Ellis (1993): Sources, mechanisms and bases of resistance in cruciferae to the cabbage aphid, *Brevicoryne brassicae*. IOBC/wprs Bulletin **16** (5), 21-35.

Speyer W (1921): Beitrag zur Biologie des Gefleckten Kohltriebrüsslers (*Ceutorrynchus quadridens* Panz.). Entomologische Blätter **17**, 118-124.

Srivastava A, Mukhopadhyay A, Arumugam N, Gupta V, Verma JK, Pental D & AK Pradhan (2004): Resythesis of *Brassica juncea* through interspecific crosses between *B. rapa* and *B. nigra*. Plant Breeding **123**, 204-206.

Textor S & J Gershenzon (2009): Herbivore induction of the glucosinolate-myrosinase defense system: major trend, biochemical bases and ecological significance. Photochemistry Review **8**, 149-170.

Ulber B (2003): Parasitoids of ceutorynchid stem weevils. In: DV Alford (ed), *Biocontrol of Oilseed Rape Pests*. Blackwell Science, Oxford, 87-95.

Ulber B & K Fischer (2006): Effect of plant density and host plant architecture on the abundance and with-in plant distribution of stem-mining pests and the level of parasitism. CD-Rom Proceedings of the International Symposium „Integrated Pest Management in Oilseed Rape Pests“, Goettingen, Germany, 3-5 April 2006.

Ulber B & O Nitzsche (2006): Phenology of parasitoids (Hym., Ichneumonidae, Tersilochinae) of oilseed rape pests in northern Germany in 1995-1997. IOBC/wprs Bulletin **29** (7), 173-179.

Ulmer BJ, Gillot C & M Erlandson (2001): Feeding preferences, growth, and development of *Mamestra configurata* (Lepidoptera: Noctuidae) on Brassicaceae. The Canadian Entomologist **133**, 509-519.

Ulmer, BJ & LM Dosdall (2006): Glucosinolate profile and oviposition behaviour in relation to the susceptibilities of Brassicaceae to the cabbage seedpod weevil. Entomologia Experimentalis et Applicata **121**, 203-213.

Wallsgrove R, Bennett R, Kiddle G, Bartlet E & J Ludwig-Mueller (1999): Glucosinolate biosynthesis and pest/disease interactions. Proceedings of the 10th International Rapeseed Congress, Canberra, Australia, 1999, 104-107.

Wang B, Ferro DN & DW Hosmer (1997): Importance of plant size, distribution of egg masses, and weather conditions on egg parasitism of the European corn borer, *Ostrinia nubilalis* by *Trichogramma ostriniae* in sweet corn. Entomologia Experimentalis et Applicata **83**, 337-345.

Weber G, Oswald S & U Zöllner (1986): Die Wirtseignung von Rapssorten unterschiedlichen Glucosinolatgehaltes für *Brevicoryne brassicae* (L.) und *Myzus persicae* (Sulzer) (Hemiptera, Aphididae). Journal of Plant Diseases and Protection **93**, 113-124.

Williams IH (2004): Advances in insect pest management of oilseed rape in Europe. In: AR Horwitz (ed), *Insect Pest Management*. Springer Verlag, Berlin, 181-208.

Winfield AL (1961): Observations on the biology and control of the cabbage stem weevil, *Ceutorhynchus quadridens* Panz. on throwse mustard (*Brassica juncea*). Bulletin of Entomological Research **52**, 589-600.

Summary

Effect of *Brassica* genotype on the infestation by cabbage stem weevil *Ceutorhynchus pallidactylus* (Mrsh.) (Col.: Curculionidae) and the parasitism of stem weevil larvae

The search for plant resistance to insect pests in oilseed rape (*Brassica napus* L. var. *oleifera* Metzg.) is still in an early stage of development. In this study, a wide spectrum of *Brassicaceae* representing a broad genetic variability (*B. napus* and *B. rapa* cultivars, breeding lines, resynthesized rapeseed lines, other *Brassica* spp.) was evaluated for resistance characteristics to cabbage stem weevil (*Ceutorhynchus pallidactylus*) for the first time. Two new methods were developed for the screening of host plant quality of a large assortment under controlled conditions in climate chambers to allow a rapid and reliable selection of genotypes with partial or full resistance.

For the quantification of leaf feeding by adult beetles, 106 genotypes were tested in no-choice screening tests for their susceptibility. In comparison to the standard cultivar 'Express', the average leaf area consumed by *C. pallidactylus* was reduced significantly by more than 40% on ten oilseed rape cultivars, four resynthesized rapeseed lines and five other accessions. In dual-choice screening tests for the evaluation of oviposition preferences, female *C. pallidactylus* laid significantly fewer eggs into plants of two oilseed rape cultivars, five resynthesized rapeseed lines and eight accessions of *B. oleracea* and *B. fruticulosa*, respectively, than into plants of the standard cultivar 'Express'.

Within three years, field experiments were carried out to evaluate the susceptibility of 42 *Brassica* genotypes to infestation by *C. pallidactylus* under field conditions. A significant correlation was found between the results obtained by laboratory screening tests and by field experiments, thereby confirming the reliability of the new laboratory tests for predicting the susceptibility of *Brassica* genotypes to *C. pallidactylus* under field conditions.

In a semi-field trial, no significant relationship was found between the total leaf glucosinolate content and the intensity of attack by *C. pallidactylus* in 12 genotypes tested. However, the stem injury coefficient was significantly correlated with single glucosinolate compounds, particularly aromatic glucosinolate gluconasturtiin and the indolic glucosinolates glucobrassicin, 4-methoxybrassicin and 4-hydroxybrassicin.

The larval parasitism of *C. pallidactylus* and *C. napi* by *Tersilochus obscurator* and *T. fulvipes* (Hym.: Ichneumonidae), respectively, was found to be significantly different between host plant genotypes. In three cultivars of turnip rape, *B. rapa*, the level of parasitism of *C. napi* by *T. fulvipes* was significantly reduced compared to cultivars of winter oilseed rape, *B. napus*. The level of parasitism was also significantly affected by host plant architecture.

In summary, the results present a new method useful in studying essential fundamentals for the development of oilseed rape cultivars with partial resistance to cabbage stem weevil and provide evidence for the impact of host plant genotype on multitrophic interactions between stem-boring insect pests and specialised parasitoids.

Zusammenfassung

Effect of *Brassica* genotype on the infestation by cabbage stem weevil *Ceutorhynchus pallidactylus* (Mrsh.) (Col.: Curculionidae) and the parasitism of stem weevil larvae

Unser Wissen über pflanzeneigene Resistenzquellen gegen Schadinsekten in Winterraps (*Brassica napus* L. var. *oleifera* Metzg.) ist mangelhaft. In dieser Forschungsarbeit wurde erstmals eine Vielzahl von Genotypen der *Brassicaceae* (kommerzielle Sorten von *B. napus* und *B. rapa*, kommerzielle Zuchtlinien, Rapsresynthesen und weitere *Brassica*-Arten) hinsichtlich ihrer Resistenzeigenschaften gegenüber dem Gefleckten Kohltriebrüssler (*Ceutorhynchus pallidactylus*) untersucht. Es wurden zwei neue Labormethoden entwickelt, um die Eignung von *Brassica*-Genotypen als Wirtspflanze unter kontrollierten Bedingungen in Klimakammern schnell zu bewerten, bzw. potentielle Resistenzen gegenüber dem Gefleckten Kohltriebrüssler zu erfassen.

In Zwangsversuchen wurden 106 *Brassica*-Genotypen geprüft, um durch Erfassung der durch den Schädling konsumierten Blattfläche Hinweise auf die Anfälligkeit der Prüfglieder gegenüber dem Gefleckten Kohltriebrüssler zu erlangen. Im Vergleich zur Kontroll-Sorte ‘Express’, die Standardsorte in allen Versuchen fungierte, war die von *C. pallidactylus* konsumierte Blattfläche bei 10 kommerziellen Raps-Sorten, 4 Raps-Resynthesen und 5 weiteren Genotypen um mehr als 40% reduziert.

In Wahlversuchen fanden sich an insgesamt 2 kommerziellen Raps-Sorten, 5 Raps-Resynthesen und 8 Genotypen von *B. oleracea* und *B. fruticulosa* signifikant weniger Eigelege der Weibchen von *C. pallidactylus* als an der Kontroll-Sorte ‘Express’.

Zusätzlich zu den Laboruntersuchungen wurde in dreijährigen Feldversuchen die Anfälligkeit von 42 *Brassica*-Genotypen gegenüber *C. pallidactylus* unter natürlichen Befallsbedingungen untersucht. Dabei zeigten sich signifikante Korrelationen zwischen den Ergebnissen der Laborprüfungen und der Feldversuche, die die Eignung der entwickelten Testmethoden für die Einschätzung der Anfälligkeit von *Brassica*-Genotypen gegenüber *C. pallidactylus* bestätigte.

In einem Halbfreilandversuch fanden sich keine signifikanten Zusammenhänge zwischen dem Gesamtgehalt an Glucosinolaten und der Befallsstärke von

C. pallidactylus in 12 Brassica-Genotypen. Allerdings zeigte sich ein signifikanter Einfluss einzelner Glucosinolate auf den so genannten “stem injury coefficient”: das aromatische Glucosinolat Gluconasturtiin und die indolischen Glucosinolate Glucobrassicin, 4-Methoxybrassicin und 4-Hydroxybrassicin.

Es fanden sich in dieser Studien signifikante Unterschiede in den geprüften Pflanzengenotypen hinsichtlich der Parasitierungsrate von *C. pallidactylus* durch den Parasitoiden *Tersilochus obscurator* (Hym.: Ichneumonidae) und in der Parasitierungsrate von *C. napi* durch den Parasitoiden *T. fulvipes* (Hym.: Ichneumonidae). Im Vergleich zu den geprüften Winterraps-Genotypen, *B. napus,* war die Parasitierungsrate von *C. napi* durch *T. fulvipes* in drei Rübsen-Genotypen, *B. rapa*, signifikant reduziert. Dabei beeinflusste die Architektur der Wirtspflanze die Parasitierungsrate signifikant.

Die Ergebnisse dieser Studie sind für die Pflanzenzüchtung von Bedeutung, um kommerzielle Raps-Zuchtlinen mit (Teil-) Resistenzen gegenüber dem Gefleckten Kohltriebrüssler in naher Zukunft zu etablieren. Ferner konnte ein Einblick in die multi-trophischen Interaktionen zwischen Wirtspflanze, stängelminierendem Pflanzenschädlingen und deren spezifischen Parasitoiden gewonnen werden.

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Finally to my alma mater: vivat, crescat, floreat Georgia Augusta in aeternum.

Extra Gottingam non est vita, si est vita non est ita!

Curriculum vitae

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