

MISE AU POINT DE LA LUTTE CONTRE LE CHARANÇON DES SILIQUES,  
*Ceuthorrhynchus assimilis* (Payk., en France).

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#### RESUME

Le charançon des siliques, *Ceuthorrhynchus assimilis* Payk., et la cécidomye des siliques, *Dasyneura brassicae* Winn., font souvent des dégâts dans les zones de production du colza d'hiver en France.

A partir de 1983, nous avons entrepris des essais et des observations en grandes parcelles pour mieux préciser la stratégie de lutte contre le charançon.

Cette stratégie repose sur les points suivants :

- la période de plus grande sensibilité est celle où se forme le plus grand nombre de siliques;
- au cours de cette période, la mise en évidence du ravageur par piégeage est difficile, et le meilleur indicateur de la présence est l'accumulation de piqûres sur les jeunes siliques;
- dans les conditions françaises, lorsque 5 siliques présentent des piqûres sur la hampe principale, les risques de nuisibilité sont élevés;
- il faut donc intervenir avant ce seuil avec un insecticide à forte action de choc respectant les abeilles; la plupart des pyrethrinoides testées (delthaméthrine à 5 g/ha, cyperméthrine à 25 g/ha, alphaméthrine à 10 g/ha, lambda-cyhalotrin à 5g/ha) satisfont ces conditions.

La persistance d'action de ces produits reste faible (de l'ordre de 4 à 5 jours) dans les conditions de la lutte contre le charançon des siliques.

#### INTRODUCTION

La difficulté pour une bonne maîtrise du charançon des siliques *Ceuthorrhynchus assimilis* Payk. en France tient à plusieurs raisons :

- jusqu'à une date récente, le choix des produits de traitement était limité à des matières actives particulièrement sensibles aux conditions d'application (température, mouillant), avec une faible persistance d'action, comme la phosalone ou le dialiphos (PIERRE et al. 1983).
- l'observation au champ du charançon des siliques est souvent délicate, en raison de l'étalement des vols dans le temps (RISBEC-1952 BONNEMAISON-1957) capable de réinfester des zones traitées avec une forte hétérogénéité spatiale (effets de bordure notamment).

- les colzas manifestent une longue période de sensibilité au charançon adulte, puis aux larves et enfin au ravageur associé qu'est la cécidomyie des siliques *Dasyneura brassicae* Winn., et l'hétérogénéité est très forte d'une plante à l'autre. L'arrivée des pyrèthrinoides de synthèse sur le marché et leur autorisation d'emploi pendant la floraison est un élément nouveau important à prendre en compte pour l'amélioration de la stratégie de lutte contre le ravageur; l'objet principal des expériences entreprises était donc de préciser les conditions d'emploi de ces nouveaux produits pour le contrôle du charançon des siliques.

#### MATERIEL ET METHODES

La méthodologie de l'étude a été mise au point sur 8 parcelles du nord du Bassin Parisien (département de l'Oise) en 1983 et appliquée sur 12 parcelles de la même région en 1984 et sur 12 parcelles en 1985 ainsi que sur 11 parcelles de l'Est de la France (département de Meurthe et Moselle) en 1984.

Un seul traitement (produit ou date d'application) est mis en place par parcelle agricole. Le traitement occupe toute la surface de la parcelle.

Le relevé des nombres de charançons capturés au filet-fauchoir ou le pourcentage de siliques attaquées en différents endroits de la parcelle, montre que au plan qualitatif, l'évolution est identique au sein d'une parcelle (figure 1), voire au sein d'une zone écologique, même si des différences existent au plan quantitatif.

Au sein de la parcelle, un site d'observation, de taille adaptée à la nature des relevés à faire est retenu; par exemple, pour une estimation de la population par filet-fauchoir, le site sera d'environ 500 mètres carrés, et de 20 mètres carrés environ pour la suivi du taux d'attaque des siliques par le ravageur.

Pour les observations sur plantes, on retient une population de plantes types, par exemple celles qui ont 4 ou 5 ramifications. On a pu vérifier la relation établie par LERIN (1982) entre le taux d'attaque sur la hampe principale et le taux d'attaque sur la plante entière (figure 2), et par conséquent, les relevés sont faits sur 10 hampes principales de plantes-types. Ceci permet de minimiser les effets de la variabilité plante à plante.

L'évaluation de la performance des traitements se fait en considérant l'évolution dans le temps d'une ou de plusieurs caractéristiques pertinentes et non pas la valeur absolue de cette ou de ces caractéristiques à une date donnée.

La liste des différents traitements est reportée au tableau 1 pour les parcelles du Nord de la France en 1983-84 et 1984-85.

#### RESULTATS

Une stratégie de lutte efficace et raisonnée repose sur les points suivants : mise en évidence du ravageur, du stade sensible de la plante, de la quantité de dégâts supportables par la culture. Si les règles de décision élaborées amènent alors l'agriculteur à réaliser un traitement, il lui faudra choisir un

produit autorisé pour cet usage, en tenant compte de ses caractéristiques biologiques (efficacité et sélectivité vis-à-vis des auxiliaires en particulier), et de son coût.

#### 1-Mise en évidence du ravageur.

Les études entreprises montrent que l'utilisation du filet-fauchaie reste la méthode de référence pour l'estimation des populations de charançons des siliques : l'attractivité de la cuvette jaune couramment employée en France pour détecter la présence des autres ravageurs du colza est très faible pendant la période de floraison, et le dénombrement visuel des charançons sur une plante présente une fiabilité faible, en raison de sa sensibilité aux conditions climatiques et au coup d'oeil de l'observateur.

On peut enfin se servir de méthodes indirectes pour quantifier la présence de charançons des siliques dans une culture : les piqûres et les pontes de l'insecte sont en effet des marqueurs de son activité. Cette technique nous semble à recommander comme étant la plus simple à mettre en oeuvre avec une bonne fiabilité pendant la floraison.

#### 2-Stade sensible de la plante.

Trois types de dégâts sont imputables directement ou non au charançon des siliques : les piqûres d'alimentation de l'adulte sur boutons et jeunes siliques, les pertes de graines dont s'alimente la larve et les éclatements de siliques dus aux pontes de la cécidomyie des siliques dans les perforations laissées par le charançon.

La nuisibilité entraînée par les seules piqûres d'alimentation sur boutons et sur jeunes siliques est faible, ainsi que l'a montré LERIN (1983) en conditions contrôlées et en absence de cécidomyies. Dans les conditions du Nord et de l'Est de la France, le stade sensible de la plante ne commencera donc pas avant l'apparition des siliques sur la plante.

La fin de la période sensible est difficile à préciser d'après nos résultats. On peut considérer qu'elle s'étend tant qu'il y a présence de jeunes siliques, pratiquement jusqu'à 15 jours après la défloraison du colza. Selon les conditions climatiques, la période de sensibilité durera donc de 5 à 8 semaines.

#### 3-Seuil d'intervention.

Les résultats obtenus en 1985 montrent qu'une intervention décidée lorsque 5 siliques sont piquées en moyenne par le charançon des siliques sur la hampe primaire des plantes types, a permis un contrôle satisfaisant des dégâts; l'intervention faite lorsque dix siliques étaient piquées semble satisfaisante (tableau 2).

#### 4-Choix d'un produit.

Compte-tenu des contraintes liées aux interventions insecticides pendant la période de floraison, il est impératif de n'employer que des produits réputés non dangereux pour les abeilles. Parmi ceux-ci, les pyréthrinoides de synthèse sont plus réguliers que les produits qui les ont précédés sur le marché.

Cependant, dans les conditions de la pratique agricole, c'est surtout leur action de choc qui est importante, leur persistance d'action étant faible à cette époque (tableau 3). Ces derniers résultats ont été obtenus en conditions semi-contrôlées : l'application de l'insecticide se fait au champ, et on teste la mortalité au laboratoire en renouvelant tous les jours les hampe florales et les insectes (PIERRE J.G. et al. 1983).

#### CONCLUSIONS

La stratégie de lutte suivante peut donc être proposée contre le charançon des siliques en France :

- début de la surveillance à la chute des premiers pétales,
  - choisir le type de plantes sur lesquelles seront faits les comptages,
  - compter le nombre de siliques présentant des piqûres de charançon des siliques sur la hampe primaire de 10 plantes-types par zone d'échantillonnage. Le nombre de zones dépendra de la taille de la parcelle.
  - réaliser l'intervention insecticide dès que le seuil moyen de 5 piqûres par plante est atteint, avec une pyréthrianoïde utilisable dans ces conditions,
  - maintenir la surveillance et, éventuellement, envisager un second traitement en cas de réinfestation indiquée par la poursuite de l'accumulation des piqûres sur siliques.
- D'après notre expérience, le seul inconvénient lié à cette méthode est la difficulté de réaliser un échantillonnage correct dans le colza après la floraison.

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Figure 1 :

NOMBRE DE CHARANCONS CAPTURES AU FILET-FAUCHOIR (10 COUPS)  
 POUR CHACUN DES 3 SITES DE LA PARCELLE N°2  
 - OISE 1984 -  
 (les 3 sites sont répartis sur la plus grande diagonale de la  
 parcelle).

- o : 1 - o : 2 - \* : 3

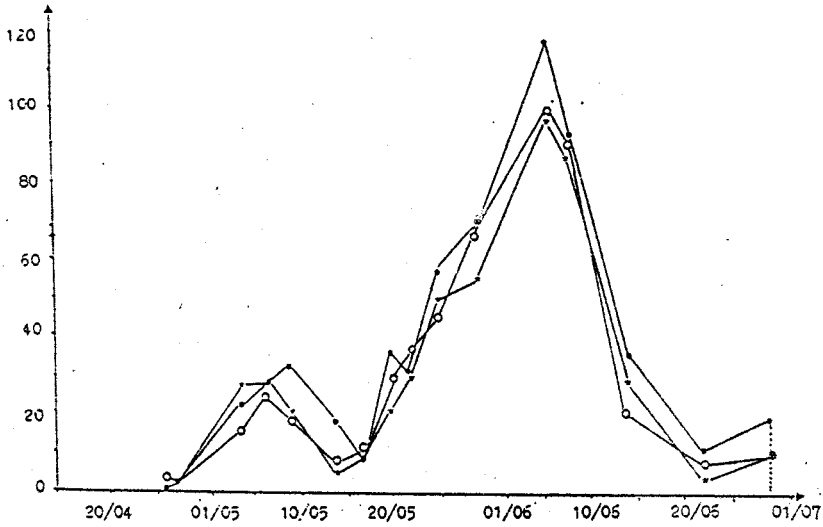


Figure 2 :

ESSAIS CHARANCONS DES SILIQUES  
 - 1983 -

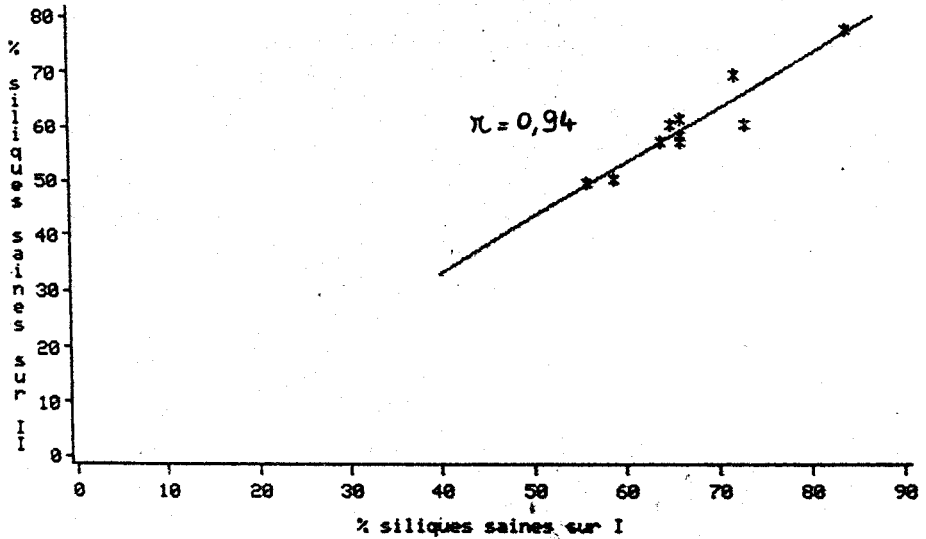


TABLEAU 1 : CARACTERISTIQUES DES PARCELLES

| An    | lieu | parc | variété  | traitement  |       |                |
|-------|------|------|----------|-------------|-------|----------------|
|       |      |      |          | date        | stade | matière active |
| 83/84 | Nord | 2    | Jet Neuf | T E M O I N |       |                |
| "     | "    | 3    | "        | 18/05       | EP    | Phosalone      |
| "     | "    | 4    | "        | 18/05       | EP    | Dialiphos      |
| "     | "    | 5    | "        | 17/05       | EP    | Bromophos      |
| "     | "    | 6    | "        | 16/05       | EP    | Cypermethrine  |
| "     | "    | 7    | "        | 16/05       | EP    | Alphamethrine  |
| "     | "    | 8    | "        | 16/05       | EP    | Deltamethrine  |
| "     | "    | 9    | "        | 08/05       | 1V    | Deltamethrine  |
| "     | "    | 10   | "        | 28/05       | 2V    | Deltamethrine  |
| "     | "    | 11   | "        | 09/05       | 1V    | Deltamethrine  |
| "     | "    | 23   | "        | et 28/05    | 2V    | Deltamethrine  |
| "     | "    | 24   | "        | T E M O I N |       |                |
|       |      |      |          | T E M O I N |       |                |
| 84/85 | Nord | 1    | Jet Neuf | T E M O I N |       |                |
| "     | "    | 2    | "        | T E M O I N |       |                |
| "     | "    | 3    | "        | 20/05       | 1V    | Deltamethrine  |
| "     | "    | 4    | "        | 20/05       | 1V    | Deltamethrine  |
| "     | "    | 5    | "        | et 01/06    | 2V    | Deltamethrine  |
| "     | "    | 6    | "        | 25/05       | 5SP   | Deltamethrine  |
| "     | "    | 7    | Bienvenu | 28/05       | 10SP  | Deltamethrine  |
| "     | "    | 8    | "        | T E M O I N |       |                |
| "     | "    | 9    | "        | T E M O I N |       |                |
| "     | "    | 10   | "        | 16/05       | 1V    | Deltamethrine  |
| "     | "    | 11   | "        | 17/05       | 1V    | Deltamethrine  |
| "     | "    | 12   | "        | et 31/05    | 2V    | Deltamethrine  |
| "     | "    |      | "        | 28/05       | 5SP   | Deltamethrine  |
| "     | "    |      | "        | 28/05       | 10SP  | Deltamethrine  |

|               |                 |
|---------------|-----------------|
| Phosalone     | 1200 g.a.a. /ha |
| Dialiphos     | 540 g.a.a. /ha  |
| Bromophos     | 500 g.a.a. /ha  |
| Cypermethrine | 25 g.a.a. /ha   |
| Alphamethrine | 10 g.a.a. /ha   |
| Deltamethrine | 5 g.a.a. /ha    |

EP : entrée en ponte des femelles de Charançons.

1V : Premier vol du charançon détecté en cuvettes.

2V : Second vol du charançon détecté en cuvettes.

5SP : 5 siliques piquées par le charançon en moyenne sur la hampe primaire des plantes types.

10SP : 10 siliques piquées par le charançon en moyenne sur la hampe primaire des plantes types.

TABLEAU 2 : EVOLUTION DU NOMBRE DE SILIQUES ATTAQUES SUR HAMPES PRINCIPALES - OISE 1985

| Traitement DECIS 0.2 l/ha |          |           | mai |      | juin |     |      | juillet |  |
|---------------------------|----------|-----------|-----|------|------|-----|------|---------|--|
| parc.                     | Variété  | Traité le | 24  | 4    | 13   | 24  | 4    | 15      |  |
| 1                         | J9       | Témoin    | 7.2 | 10.1 | 5.7  | 7.8 | 7.7  | 11.1    |  |
| 2                         | J9       | "         | 4.8 | 7.9  | 8.9  | 8.1 | 12.6 | 13.8    |  |
| 5                         | J9       | 23/05     | 4.2 | 1.0  | 0.5  | 1.2 | 1.4  | 1.3     |  |
| 6                         | J9       | 28/05     | 9.2 | 8.8  | 6.4  | 7.6 | 8.6  | 9.6     |  |
| 7                         | Bienvenu | Témoin    | 4.8 | 6.2  | 2.0  | 4.1 | 9.2  | 8.0     |  |
| 8                         | Bienvenu | Témoin    | 4.3 | 9.3  | 5.9  | 9.1 | 11.6 | 11.1    |  |
| 11                        | Bienvenu | 28/05     | 4.0 | 3.4  | 2.8  | 3.5 | 3.9  | 5.0     |  |
| 12                        | Bienvenu | 28/05     | 8.4 | 6.7  | 7.1  | 6.5 | 4.6  | 6.6     |  |

TABLEAU 3 : EVOLUTION DE LA PERSISTANCE D'ACTION DU DECIS

|                  | Nombre de morts en % |      |      |      |      |
|------------------|----------------------|------|------|------|------|
|                  | J+0                  | J+1  | J+4  | J+7  | J+11 |
| PLANTES TRAITÉES | 100                  | 83.2 | 14.4 | 15.2 | 14.4 |
| TÉMOIN           | 20                   | 11.2 | 2.4  | 8.4  | 4.0  |

APPLICATION OF NEW INSECTICIDES IN THE CONTROL OF  
CEUTORHYNCHUS ASSIMILIS AND DASYNEURA BRASSICAE IN  
WINTER RAPE

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I. Introduction

The major pests of winter rape in Poland, occurring during flowering, are two species: the pod weevil /*Ceutorhynchus assimilis* Payk./ and pod midge /*Dasyneura brassicae* Winn./. The yield losses presently caused by these pests are significant and tend to increase /Czajkowska, Dmoch, 1975; Lewartowski, Piekarczyk, 1987/. The control of these pests should be commonly used everywhere, where damages are measurable.

An efficient protection of winter rape against pests is plant spraying with relevant chemicals /Alford et al., 1979; Pierre et al., 1983; Roa, Garnier, 1983; Skrocki, 1978/. Until now, however, the assortment of these chemicals has not been wide and only some of the insecticides used for rape protection may be applied for the control of the pod weevil and pod midge /Mrówczyński, Ciesielski, 1983; Mrówczyński et al., 1984/. The main barrier in their application is the toxicity to bees and a long period of prevention /Benedek, 1983 a; Benedek, 1983 b; Bacquet et al., 1983; Shires, Murray, 1983/. Hence there was an urgent need in Poland to study not only effective, but also safe to bees new chemicals, which upon their registration may find a wide application in rape protection. For that purpose the Institute of Plant Protection in Poznań started in 1980 a cycle of experiments on the estimate of the effectiveness of a dozen or so new insecticides, which may be suitable for rape protection during its flowering. Characteristics of the studied chemicals are given in Table 1 /Mrówczyński et al., 1984; Stevenson

et al., 1979; Twinn, Lacy, 1979; Worthing, 1987/.

## II. Methods

The field trials on the estimate of the effectiveness of new insecticides were carried out on production rape plantations.

The studied chemicals were applied at different rates. Comparable chemicals were: Thiodan 35 fluid, and Zolone 30 WP. Rape was sprayed at full flowering using pneumatic apparatus, the rate of the working liquid being  $300 \text{ dm}^3/\text{ha}$ . An estimate of the effectiveness of separate chemicals was made during rape ripening. For that purpose 400 pods were randomly collected from each field /in 4 replications with 100 pods each/ and then analysed for the infestation degree by the larvae of the pod weevil and pod midge. The obtained results are given in per cent of injured pods. The results are summarized in Tables 2 and 3.

## III. Discussion of Results

On rape plantations the pod injuries caused exclusively by the pod weevil on the control fields was on the average 22.8%, whereas pod damages caused together by the pod weevil and pod midge averagely amounted to 35.4%. On the fields sprayed with the studied chemicals the percentage of injured pods in separate years was from several to over a dozen times lower.

The performed experiments showed that most of the studied new chemicals were sufficiently effective in controlling pod pests in flowering winter rape. No pronounced differentiation in the effectiveness of the new studied chemicals was found in different years, whereas the comparative compounds, Thiodan 35 fluid and Zolone 30 WP, which have been generally recommended until now for the control of the pod weevil and pod midge in Poland, displayed a markedly weaker action.

## IV. Conclusions

On the basis of the performed experiments on estimating the effectiveness of new insecticides in the control



of pod pests it has been found that:

1. The pod weevil and pod midge occurred on the fields covered by the experiments at a large intensity and caused serious pod damages;
2. the new insecticides effectively controlled the pod weevil and pod midge, reducing pod injuries manyfold.

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Table 1

Characteristics of the insecticides used for the control of *Centorhynchus assimilis* Payk. and *Dasyneura brassicae* Winn.

| Preparations     | Producer      | Active ingredient | Prevention period for bees in hours |
|------------------|---------------|-------------------|-------------------------------------|
| Ambush 25 EC     | ICI           | permethrin        | 2                                   |
| Banco1 50 WP     | Takeda        | bensultap         | 6                                   |
| Cybolt 100 EC    | Cyanamid      | flucythrinate     | 6                                   |
| Cymbush 10 EC    | ICI           | cypermethrin      | 3                                   |
| Cymbush 25 EC    | ICI           | cypermethrin      | 3                                   |
| Cyperkil 25 EC   | Chemie Linz   | cypermethrin      | 3                                   |
| Decis 2.5 EC     | Roussel-Uclaf | deltamethrin      | 1                                   |
| Decis 2.5 flow   | Roussel-Uclaf | deltamethrin      | 1                                   |
| Evisect S        | Sandoz        | thiocyclam        | 3                                   |
| Fastac 10 EC     | Shell         | alphamethrin      | 1                                   |
| Karate 2.5 EC    | ICI           | cyhalotrin        | 1                                   |
| Mavrik 2 E       | Sandoz        | fluvalinate       | 2                                   |
| Permasect 25     | Sandoz        | permethrin        | 2                                   |
| Polytrin 200 EC  | Ciba-Geigy    | cypermethrin      | 3                                   |
| Ripcord 10 EC    | Shell         | cypermethrin      | 3                                   |
| Sherpa 25 EC     | Rhône-Poulenc | cypermethrin      | 3                                   |
| Sumialpha 5 EC   | Sumitomo      | esfenvalerate     | 1                                   |
| Sumicidin 20 EC  | Sumitomo      | fenvalerate       | 4                                   |
| Talstar 10 EC    | FMC           | biphenthrin       | 3                                   |
| Thiodan 35 fluid | Hoechst       | endosulfan        | 5                                   |
| Torak 48 EC      | Schering      | dialifos          | 1                                   |
| Trebon 30 EC     | Nichimen      | ethofenprox       | 3                                   |
| Trebon 10 flow   | Nichimen      | ethofenprox       | 3                                   |
| Zolone 35 EC     | Rhône-Poulenc | phosalone         | 3                                   |
| Zolone 30 WP     | Rhône-Poulenc | phosalone         | 3                                   |
| Zolone 50 flow   | Rhône-Poulenc | phosalone         | 3                                   |

Table 2

The effectiveness of insecticides from the group of organophosphorus compounds, carbamates and others against *Ceutorhynchus assimilis* Payk. and *Dasynoura brassicae* Winn. in the years 1980-1986.

| Preparations     | Doses used<br>in kg or<br>dm <sup>3</sup> per<br>1 ha | Average per cent of injured<br>pods |                           |                 |
|------------------|---|-------------------------------------|---------------------------|-----------------|
|                  |   | pod weevil                          | pod weevil<br>+ pod midge | total<br>injury |
| Danco 50 WP      | 0.75  | 1.3                                 | 1.0                       | 2.3             |
| Danco 50 WP      | 1.0   | 0.5                                 | 0.5                       | 1.0             |
| Evisect S        | 0.5   | 8.5                                 | 7.3                       | 15.8            |
| Evisect S        | 0.7   | 5.9                                 | 3.7                       | 9.6             |
| Thiodan 35 fluid | 1.5   | 16.3                                | 10.7                      | 27.0            |
| Torek 48 EC      | 1.0   | 8.6                                 | 7.0                       | 15.6            |
| Trebon 30 EC     | 0.15  | 1.6                                 | 1.0                       | 2.6             |
| Trebon 30 EC     | 0.3   | 0.7                                 | 0.4                       | 1.1             |
| Trebon 10 flow   | 0.3   | 2.0                                 | 1.7                       | 3.7             |
| Trebon 10 flow   | 0.6   | 0.7                                 | 1.0                       | 1.7             |
| Zolone 35 EC     | 3.0   | 8.1                                 | 6.4                       | 14.5            |
| Zolone 30 WP     | 3.0   | 12.3                                | 7.2                       | 19.5            |
| Zolone 50 flow   | 2.0   | 6.2                                 | 5.9                       | 12.1            |
| Control          | -   | 22.8                                | 12.6                      | 35.4            |

Table 3

The effectiveness of pyrethroids against *Ceutorhynchus assimilis* Payk. and *Dasyneura brassicae* Winn. in the years 1980-1986

| Preparations    | Doses used<br>in kg or<br>dm <sup>3</sup> per<br>1 ha | Average per cent injured |                           |                 |
|-----------------|---|--------------------------|---------------------------|-----------------|
|                 |   | pod weevil               | pod weevil<br>+ pod midge | total<br>injury |
| Ambush 25 EC    | 0.1   | 10.9                     | 5.6                       | 16.5            |
| Cybolt 100 EC   | 0.5   | 0.7                      | 1.0                       | 1.7             |
| Cymbush 10 EC   | 0.25  | 4.4                      | 3.5                       | 7.9             |
| Cymbush 25 EC   | 0.1   | 1.2                      | 0.8                       | 2.0             |
| Cyperkil 25 EC  | 0.1   | 0.8                      | 0.2                       | 1.0             |
| Decis 2.5 EC    | 0.3   | 3.7                      | 2.3                       | 6.0             |
| Decis 2.5 flow  | 0.3   | 1.7                      | 1.9                       | 3.6             |
| Fastac 10 EC    | 0.075   | 2.2                      | 1.6                       | 3.8             |
| Fastac 10 EC    | 0.1   | 1.6                      | 1.0                       | 2.6             |
| Fastac 10 EC    | 0.12  | 0.5                      | 0                         | 0.5             |
| Karate 2.5 EC   | 0.25  | 0.7                      | 0.4                       | 1.1             |
| Karate 2.5 EC   | 0.3   | 0.7                      | 0.3                       | 1.0             |
| Karate 2.5 EC   | 0.4   | 0.5                      | 0                         | 0.5             |
| Mavrik 2 E      | 0.2   | 2.7                      | 2.2                       | 4.9             |
| Mavrik 2 E      | 0.25  | 0.7                      | 1.6                       | 2.3             |
| Mavrik 2 E      | 0.3   | 1.2                      | 0.5                       | 1.7             |
| Permasect 25    | 0.1   | 11.9                     | 6.0                       | 17.9            |
| Polytrin 200 EC | 0.12  | 0.5                      | 1.0                       | 1.5             |
| Ripcord 10 EC   | 0.25  | 3.7                      | 2.2                       | 5.9             |
| Sherpa 25 EC    | 0.1   | 1.1                      | 1.0                       | 2.1             |
| Sumialpha 5 EC  | 0.2   | 0.6                      | 0.8                       | 1.4             |
| Sumialpha 5 EC  | 0.25  | 0                        | 0.5                       | 0.5             |
| Sumicidin 20 EC | 0.3   | 3.6                      | 2.6                       | 6.2             |
| Sumicidin 20 EC | 0.5   | 1.0                      | 0.5                       | 1.5             |
| Talstar 10 EC   | 0.05  | 3.5                      | 3.0                       | 6.5             |
| Talstar 10 EC   | 0.075   | 2.2                      | 2.2                       | 4.4             |
| Talstar 10 EC   | 0.1   | 0.5                      | 1.0                       | 1.5             |
| Control         | -   | 22.8                     | 12.6                      | 35.4            |

DECIS INNOCUITY TOWARDS BEES, CONTROL OF  
CEUTHORRHYNCHUS ASSIMILIS ON RAPSBaumeister R., Roa L.

Roussel Uclaf, France

/Inhalt des Posters/ experimental designs /station  
tests on white mustard, trials  
under tunnels and open field  
tests - treatment by crop  
sprayers and treatment by heli-  
copter/

/Main results:/

- mortality under tunnels
- mortality in 3 open fields  
/treatment bei helicopter/
- honey harvest

## STUDIES ON PYRETHROID RESIDUES IN WINTER RAPE

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## I. Introduction

The Institute of Plant Protection has been conducted studies on the effectiveness of pyrethroids against winter rape pests since 1976. In all the experiments the results obtained after the application of pyrethroids were better than or at least equal to those obtained after the application of organophosphorus chemicals or carbamates /Witkowski et al. 1981; Mrówczyński et al. 1984; Pieczonka, Mrówczyński, 1985; Witkowski et al. 1986/. Table 1 summarizes rates of active ingredients and pre-harvest intervals of pyrethroids registered in Poland for winter rape protection.

In Canada, FRG and Great Britain studies were conducted on cypermethrin residues in rapeseed after the application of the chemical Ripcord. Most of the samples showed no cypermethrin residues above the limit of determination, i.e. 0.01 mg/kg. Only seed samples from Canada 6 weeks after the treatment were found to have cypermethrin residues from 0.01 to 0.12 mg/kg /Anonim 1, 1980/.

In Switzerland, Finland, FRG and France it was found that rape seed during 51-87 days after the application of Decis had deltamethrin residues below 0.05 mg/kg /Anonim 2, 1981/.

Rape seed from Canada after the application of Sumicidiaz were found to have 0.11 mg/kg fenvalerate residues in two samples 42 days after the treatment, whereas the remaining samples showed no fenvalerate residues above 0.01 mg/kg /Anonim 3, 1982/.

In view of the lack of data concerning degradation of pyrethroids in green rape, very long pre-harvest intervals have been established for the actually registered chemicals for winter rape protection, namely: for cypermethrin - 49

days, for fenvalerate - 56 days and deltamethrin - 35 days.

The purpose of this paper was to determine the level of pyrethroid residues in seeds, pods and straw, as well as to determine the dynamics of these residues degradation in green matter.

## II. Methods

Field trials in 1982-1985 were carried out on plots covering 100 m<sup>2</sup> at the Experimental Department - Przybroda belonging to The University of Agriculture in Poznań and at the Experimental Station of the Plant Protection Institute in Winnogóra. In these trials different rates of chemicals were applied. Rape was sprayed once at full flowering against *Ceutorhynchus assymilis* Payk. and *Dasyneura brassicae* Winn. or twice - before plant flowering against *Meligethes assimilis* F. and at full flowering of plants against *C. assymilis* Payk. and *D. brassicae* Winn.

For the analysis samples of green rape /20 plants/ from 0 to 26 days after the treatment and samples of seeds, pods and straw were randomly collected during harvest.

The determination method of alphamethrin, biphenthrin, cypermethrin, deltamethrin and fenvalerate residues in the discussed material was worked out at the Institute of Plant Protection in Poznań. It consists of extraction of samples by organic solvents. Extracts are cleaned-up by solvent partition and on a Florisil column. The final determination is carried out on a gas chromatograph with an electron capture detector. The limit of determination for all the mentioned pyrethroids is 0.01 mg/kg.

## III. Discussion of the Results and Conclusions

In 1982 the dynamics of cypermethrin degradation in green matter /Fig. 1/ as well as cypermethrin residues in straw, pods and seed after the application of Ripcord 10 EC and Ripcord 40 EC for winter rape protection were studied /Table 2/. Cypermethrin residues in rape immediately after the second treatment are from 0.81 to 2.15 mg/kg. These quantities are half-reduced already after 3-4 days, and 12 days



after the second treatment they attain the level below 0.20 mg/kg.

Cypermethrin residues in straw from 0.03 to 0.17 mg/kg are on a similar level as in green matter already 12 days after the second treatment. Seed and pod samples were not found to have cypermethrin residues above the limit of determination of the method /0.01 mg/kg/.

According to the data of FAO/WHO Reports, maximum permissible cypermethrin residues in the seed of oil-plants - 0.2 mg/kg are proposed /Anonim 1, 1980/.

Cypermethrin is a mixture of 8 stereoisomers, of which two the most active ones were isolated as alphamethrin. Alphamethrin is an active ingredient of the preparation Fastac 10 EC /Harris, 1985; Fisher et al. 1983/. In 1984 the dynamics of alphamethrin degradation in green matter and its residues in straw, pods and seeds after the application of Fastac 10 EC were studied /Fig. 2/.

Alphamethrin residues in green matter after treatment are from 0.18 to 0.43 mg/kg depending on the applied dose of the chemical and on the number of treatments. These residues are half-reduced already 3 days after the treatment, but after 12 days they are below 0.1 mg/kg in all the samples. Alphamethrin residues in the green rape samples 25 days the treatment and in straw samples collected during harvest - 68 days after the application of this chemical are the same amounting to 0.02 mg/kg. No alphamethrin residues above the limit of determination of the method were found in seed samples.

In 1983 deltamethrin residues were studied after the application of the insecticides Decis 2.5 EC and Decis 2.5 flow /Table 4/, as well as fenvalerate residues after the application of Sumicidin 20 EC /Table 5/ in green rape, straw, pods and seeds.

The studied seed samples were not found to have deltamethrin and fenvalerate residues above the limit of determination of the method /0.01 mg/kg/.

According to the data of FAO/WHO Reports, 0.1 mg/kg maximum permissible deltamethrin and fenvalerate residues in

the seeds of oil-plants are proposed /Anonim 2, 1981; Anonim 3, 1982/.

In 1985 bifenthrin residues in seeds, straw and pods after the application of Talstar 10 EC were studied /Table 6/. The seeds were not found to have bifenthrin residues above the limits determination of the method /0.01 mg/kg/.

All the discussed studies were carried out on the winter rape cultivar Jet Neuf.

In 1984 chemical treatments with Decis 2.5 EC at the rate of 7.5 g deltamethrin per 1 ha /0.3 l per 1 ha/ and with Fastac 10 EC at the rate of 12 g alphamethrin per 1 ha /0.12 l per 1 ha/ were performed on different winter rape varieties:

- with a high erucic acid content - Górczański and Skrzyszowicki;
- with a low erucic acid content - Beryl, Jupiter and Jet Neuf;
- and erucic acid-free with a low content of thioglikosides - Jantar, Start and the strain BKH-180.

In the studied rapeseed samples no alphamethrin and deltamethrin residues above the limit of determination of the method were found.

Summing that up, it should be inferred that in the studied seed samples of winter rape no pyrethroid residues were found irrespective of the kind of applied chemical, number of treatments and variety of a cultivated plant.

On the basis of the results of the studies concerning the dynamics of degradation of cypermethrin and alphamethrin residues and the residues of bifenthrin and fenvalerate in green matter it may be proposed to reduce the obligatory so far pre-harvest interval for the discussed chemicals to 10-14 days.

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Table 1

Active ingredient rates and pre-harvest intervals of pyrethroids registered in 1987 in Poland for protection of winter rape

| Active ingredient | Preparation   | Dosage<br>/a.i.g/ha/ | Pre-harvest<br>intervals /days/ |
|-------------------|---|----------------------|---------------------------------|
| alphamethrin      | Fastac 10 EC  | 8-12                 | 49                              |
| cypermethrin      | Cymbush 10 EC<br>Cymbush 25 EC<br>Cyperkil 25 EC<br>Ripoord 10 EC<br>Sherpa 25 EC | 25-30                | 49                              |
| deltamethrin      | Decis 2.5 EC<br>Decis 0.5 ULV   | 6.25-8.75            | 35                              |
| fenvalerate       | Sumicidin 20 EC   | 80-100               | 56                              |

Table 2

Cypermethrin residues in winter rape during harvest in 1982

| Preparations  | Rate               |                     | Number of treatments | Cypermethrin residues /mg/kg/ |        |        |
|---------------|--------------------|---------------------|----------------------|-------------------------------|--------|--------|
|               | preparation /l/ha/ | cypermethrin /g/ha/ |                      | straw                         | Pods   | seeds  |
| Ripeord 10 EC | 0.25               | 25                  | 2                    | 0.03                          | < 0.01 | < 0.01 |
| Ripeord 10 EC | 0.40               | 40                  | 2                    | 0.08                          | < 0.01 | < 0.01 |
| Ripeord 40 EC | 0.06               | 24                  | 2                    | 0.17                          | < 0.01 | < 0.01 |
| Ripeord 40 EC | 0.10               | 40                  | 2                    | 0.10                          | < 0.01 | < 0.01 |

Table 3

Cypermethrin residues in winter rape in 1983

| Preparations  | Rate               |                     | Number of treatments | Cypermethrin residues /mg/kg/ |      |        |        |        |
|---------------|--------------------|---------------------|----------------------|-------------------------------|------|--------|--------|--------|
|               | preparation /l/ha/ | cypermethrin /g/ha/ |                      | green matter                  |      | straw  | Pods   | seeds  |
|               |                    |                     |                      | I                             | II   |        |        |        |
| Ripeord 10 EC | 0.25               | 25                  | 1                    | 0.24                          | 0.10 | 0.07   | 0.02   | < 0.01 |
| Ripeord 10 EC | 0.25               | 25                  | 2                    | 0.33                          | 0.06 | 0.02   | < 0.01 | < 0.01 |
| Cymbush 10 EC | 0.25               | 25                  | 1                    | 0.62                          | 0.12 | < 0.01 | < 0.01 | < 0.01 |
| Cymbush 25 EC | 0.10               | 25                  | 1                    | 0.37                          | 0.13 | 0.14   | 0.06   | < 0.01 |
| Cymbush 25 EC | 0.10               | 25                  | 2                    | 0.38                          | 0.07 | 0.01   | < 0.01 | < 0.01 |
| Sherpa 25 EC  | 0.10               | 25                  | 1                    | 0.37                          | 0.06 | 0.06   | 0.02   | < 0.01 |

I - immediately after last treatment

II - 26 days after last treatment

Table 4

Deltamethrin residues in winter rape in 1983

| Preparations   | Rate               |                      | Number of treatments | Deltamethrin residues /mg/kg/ |      |        |        |        |
|----------------|--------------------|----------------------|----------------------|-------------------------------|------|--------|--------|--------|
|                | preparation /l/ha/ | delthamethrin /g/ha/ |                      | green matter                  |      | seeds  |        |        |
|                |                    |                      |                      | I                             | II   |        | straw  | pod    |
| Decis 2.5 EC   | 0.3                | 7.5                  | 1                    | 0.11                          | 0.03 | 0.02   | < 0.01 | < 0.01 |
| Decis 2.5 EC   | 0.3                | 7.5                  | 2                    | 0.22                          | 0.09 | 0.01   | < 0.01 | < 0.01 |
| Decis 2.5 flow | 0.3                | 7.5                  | 1                    | 0.05                          | 0.04 | < 0.01 | < 0.01 | < 0.01 |

I - immediately after last treatment II - 26 days after last treatment

Table 5

Fenvalerate residues in winter rape in 1983

| Preparations    | Rate               |                    | Number of treatments | Fenvalerate residues /mg/kg/ |      |        |        |        |
|-----------------|--------------------|--------------------|----------------------|------------------------------|------|--------|--------|--------|
|                 | preparation /l/ha/ | fenvalerate /g/ha/ |                      | green matter                 |      | seed   |        |        |
|                 |                    |                    |                      | I                            | II   |        | straw  | pod    |
| Sumicidin 20 EC | 0.3                | 60                 | 1                    | 0.57                         | 0.64 | < 0.01 | < 0.01 | < 0.01 |
| Sumicidin 20 EC | 0.3                | 60                 | 2                    | 2.26                         | 0.05 | 0.15   | < 0.01 | < 0.01 |
| Sumicidin 20 EC | 0.5                | 100                | 1                    | 1.78                         | 0.13 | 0.05   | < 0.01 | < 0.01 |

I - immediately after last treatment II - 26 days after last treatment

Table 6

Biphenethrin residues in winter rape in 1985

| Preparations  | Rate               |                     | Number of treatments | Biphenethrin residues /mg/kg/ |       |        |        |
|---------------|--------------------|---------------------|----------------------|-------------------------------|-------|--------|--------|
|               | preparation /l/ha/ | biphenethrin /g/ha/ |                      | straw                         | pod   |        |        |
|               |                    |                     |                      |                               | straw | pod    | seeds  |
| Talstar 10 EC | 0.2                | 20                  | 1                    | 0.31                          | 0.05  | < 0.01 | < 0.01 |
| Talstar       | 0.25               | 25                  | 1                    | 0.88                          | 0.69  | < 0.01 | < 0.01 |

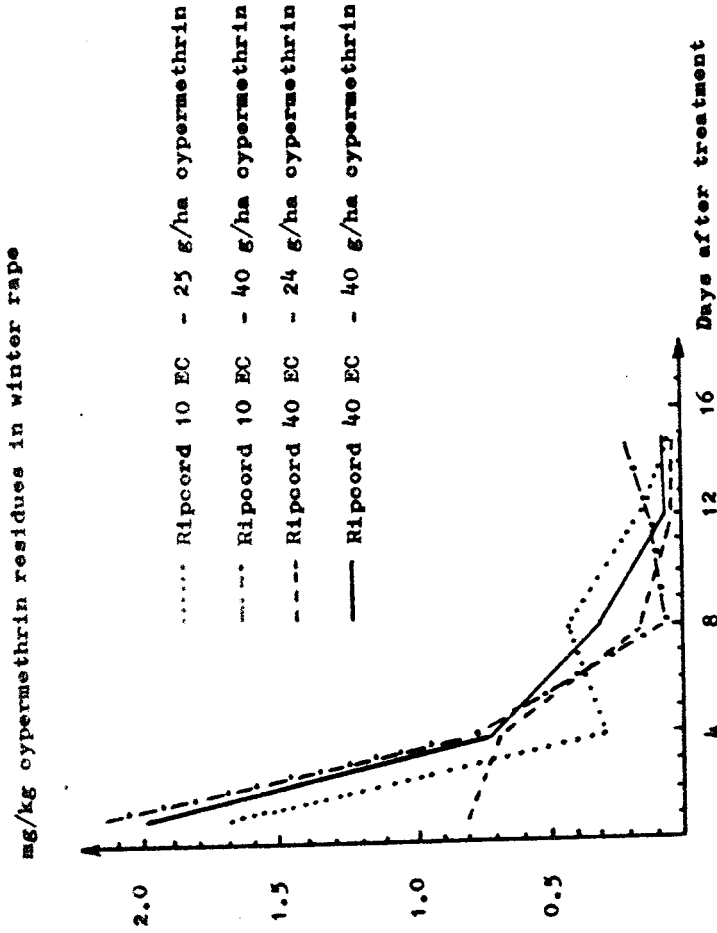


Fig. 1 - Dynamic of cypermethrin degradation in green matter in winter rape in 1982

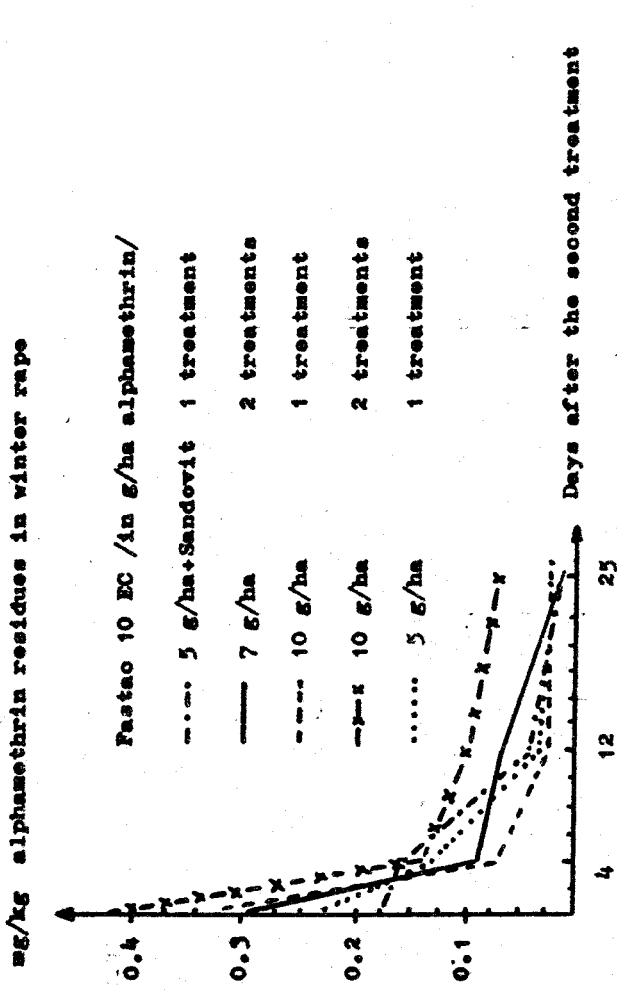


Fig. 2. Dynamic of alphamethrin degradation in green matter in 1984

SOME ASPECTS OF TANK-MIX APPLICATION OF DECIS 2,5 EC  
AND UREA IN WINTER RAPE

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I. INTRODUCTION

Tank-mix application of pesticides and mineral fertilizers is in recent years more and more preferable in agricultural practice for the tendency of investigation of new more economical technologies. In culture of winter rape there is a special need and possibility to apply insecticides against pollen beetles *Meligethes aeneus* F./ and weevils *Ceutorhynchus* sp./ in tank-mix with the second or third dose of nitrogen fertilizers in regard to a coincidence in time of these treatments.

There are also laid some hopes to receive an additional fitosanitary effect by an into-leaf application of some mineral fertilizers /Veverka and Oliberius, 1985 ; Nowosielski, 1986 ; Pałosz, in press /.

Before to recommend in practice this method of application it is necessary to carry out a many-sided experimental work on effectiveness, fitotoxicity, emulsion stability, dynamics of disappearance in plants, toxicity to bees and other useful organisms and other properties of such mixtures. The presented work is concerned some of mentioned problems in regard to 0,1 % emulsion of Decis 2,5EC + 10 % of urea /D+U/ compared to 0,1 % emulsion of Decis 2,5EC/D/.

II. MATERIALS AND METHODS

An examination of surface tension /ST/ of emulsions of D and D+U was made by a stalagmometric method. The values were calculated according to the formula :

$$S = \frac{S_w \cdot N_w \cdot d}{N \cdot d_w} \quad \text{where :}$$



S - relative ST, Sw - ST of distilled water  $\neq 1,0$  /,  
 Nw - number of drops of distilled water, d - density of  
 examined liquid in  $g/cm^3$ , N - number of drops of examina-  
 ted liquid, dw - density of distilled water in  $g/cm^3$   $\neq 1$  /

The emulsion stability /ES/ of D and D+U was examined by an adapted method of biotesting on *Musca domestica* L. /4-days females/. We have tested equal pipette samples of emulsions from the upper, middle and low part of measuring cylinder /100  $cm^3$ /. The samples were being taken off after 3, 24 and 48 hours from the time the emulsion was made up. The testing was made in Petri-dishes with  $1cm^3$  of emulsion on upper and low dish and insects were introduced after the liquid was dried up. The evaluation of ES was carried out on the base of LT-50 values according to graphic method of Lietchfield and Wilcoxon /Sliżyński and Lipa, 1973/.

Dynamics of disappearance of insecticidal activity on plants was evaluated on base of LT-50 values using the same method of biotesting on *Musca domestica* L. Insects were introduced in Petri-dishes with leaves of winter rape previously sprayed with D and D+U on experimental plots. The testing was made in October 1986, 2, 4, 8, 14 and 18 days after treatment. The plants of winter rape /cv Jet Neuf/ were grown on plots in field conditions. For testing were used only freshly picked and not wet leaves.

The field experiment on effectiveness of D and D+U against pollen beetles was carried out in May 1986 on 3-ha crop of winter rape cv. Jet Neuf. For spray was used traditional sprayer Termit with nozzles of high pressure, liquid expenditure -  $200 dm^3$  on ha and width of spray - 10 m. Effectiveness was evaluated for 2, 5 and 10 days after treatment taking into account the number of living beetles on 4 x 25 plants in each combination.

### III. RESULTS AND DISCUSSION

#### 1. Surface tension /ST/

ST is one of the most important physical properties of spray mixtures in plant protection. It is commonly known that liquids with low ST are covering better the surface of sprayed plants due to more increased dispersion.

Relative ST for D and D+U were as follows :

$$S_D = \frac{1,0 \cdot 15,5 \cdot 1,0}{15,9 \cdot 1,0} = 0,9748$$

$$S_{D+U} = \frac{1,0 \cdot 15,5 \cdot 1,025}{17,3 \cdot 1,0} = 0,9179$$

As it is seen from the calculations the mixture D+U had a little lower ST than D but the difference seems to be too small to have any practical significance. It must be underlined that an addition of Sandovit in recommended dose to D or D+U was equally effective in decreasing of ST.

An addition of urea to spray mixture will result in increasing of its specific gravity from 1,0 to 1,025 g/cm<sup>3</sup> and therefore urea may be interpreted as spray charging agent.

### 2. The rapidity of evaporation /RE/ of spray mixture from leaf surface

As it is known the insecticidal activity is much better when an insect pest is contacting with a spray mixture than with a dry film of evaporated spray on leaves /Pałocz, in press/. There are two factors deciding that RE of D+U is less than that of D. At first it results from Raoult's law, that says : the more concentrated is a solution, the more is a decreasing of vapour pressure of dissolvent. At second a phenomenon of higroscopicity has some significance. The critical relative humidity of air /RH/ for urea is 72,5 % /Veverka and Oliberius, 1935/.

We have observed for instance that in glasshouse conditions by a high RH /70-85 %/ the drops of D+U on leaves did not drying up even until 48 hours after treatment. In this conditions it has conducted to a phytotoxicity on plants.

### 3. Emulsion stability /ES/

The results of biotesting of ES are shown on a fig. 1. After 3 and 24 hours the difference in ES between D and D+U wasn't great and a some delay of insecticidal action of D+U confirms the previously stated "disguising" of insecticidal activity in dry deposit of D+U /Pałocz, in press/. In despite of that, LT-50 for low layer of D+U was decreased in comparison to D, what indicates on a process of settlement in emulsion pillar.

## EMULSION STABILITY

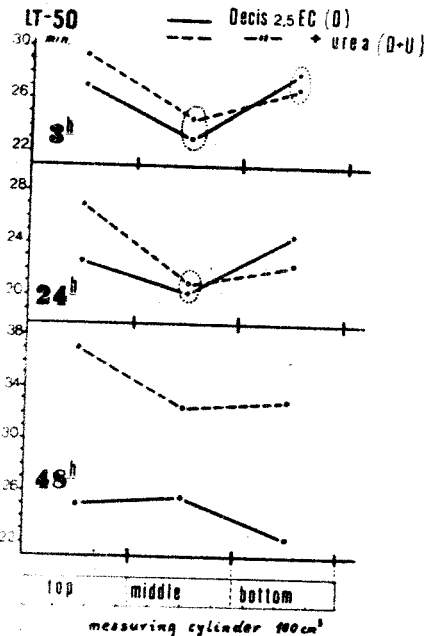


Fig. 1

After 48 hours a process of settlement in emulsion D+U was very clear /the most part of insecticide has probably settled on the bottom of cylinder/ and differences between LT-50 of D and D+U in all layers were statistically significant.

In practice the process of emulsion settlement may be quicker and therefore the immediate using of mixture D+U will be very desirable.

#### 4. Dynamics of disappearance of contact insecticidal activity /CIA/ on plants

Results are shown on fig. 2. Two days after treatment a little quicker CIA has shown D in comparison with D+U. Four days after treatment the difference between D and D+U was statistically non-significant but after 8 days one could state a considerable decrease of CIA. It mainly concerns

D+U but in some degree D too. Supposedly it was caused by a rich rainfall before sample-taking what made us dry out the wet leaves with warm air /possible loss of insecticide/.

In 14 and 18 days after treatment in both variants a progressive decreasing of CIA was happened and the difference between D and D+U was non-significant.

As a conclusion it could be stated that there was no essential difference in dynamics of disappearance of CIA between D and D+U.

## DYNAMICS OF DISAPPEARANCE

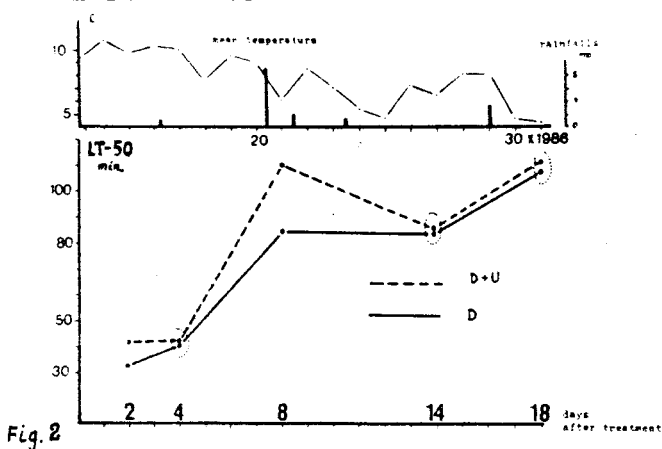


Fig. 2

## FIELD EFFECTIVENESS

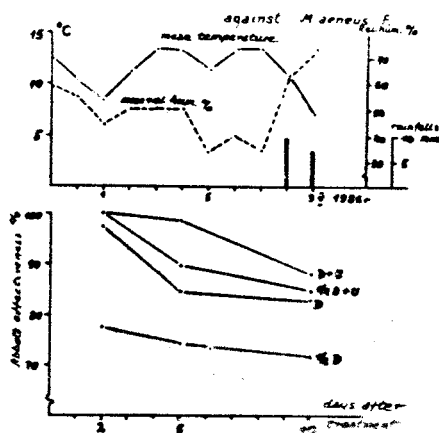


Fig. 3

5. Field effectiveness /FE/ of D and D+U on pollen beetles

Results are shown on fig. 3. The chemical treatment was carried out on 1986-04-29.

Abbott's FE of mixture D+U /even by 50% decreased dose of D/ was higher than of D without urea. It could be supposed that it was caused by a longer contact time of insects with a spray liquid D+U due to its slower degree of evaporation on plants than of D.

The experiment was established by a low infestation of crop by pollen beetles /1,06 - 1,76 beetles on plant in combination without treatment/ and therefore it would be interesting to repeat it in conditions of middle or high infestation.

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## Susceptibility of rape to fungi causing root rot

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Roots of rape and other Cruciferae plants are infected by *Fusarium* sp., *Rhizoctonia solani*, *Sclerotinia sclerotiorum* and other fungi ( Armstrong and Armstrong, 1952; Berkenkamp, Vaartnou, 1972; Frenzel et al, 1984 ).

The aim of the field work ( 1985/86 ) was to estimate the effect of rape seed dressing with Oftanol T on the occurrence of root rot and the kinds of factors causing the rot. In the greenhouse was observed the susceptibility of four rape cultivars to the fungi (*Fusarium Wollenweberii* Raikko, *Rhizoctonia solani* K.) most frequently causing root rot in the field.

### Material and methods

In the field experiment two cultivars (Beryl - „0”, Jantar - „00”), and in the greenhouse four cultivars ( Górczański and Skrzyszowicki - high erucic, Beryl - „0”, Jantar „00”) were used.

The field experiment was carried out in Zlotniki near Poznań. Plots were in a randomized complete block design with four replications. The forecrop was potato. The seeds undressed and dressed with Oftanol T were sown in rows of spaces 45; 22,5 and 15 cm. The amounts of seeds sown were

6, 12 and 18 kg ha, respectively. The roots of 20 plants from each plot were estimated at about one month's intervals (Table 1). Ripcord 10 EC against pests was used on April 10<sup>th</sup>. Isolations of microorganisms from rotting roots were made on potato dextrose agar (PDA) or on potato agar (PA). Ethanol 96 % and sublimate 0,1 % were used for disinfection of roots. The pathogenicity of the isolated microorganisms was checked. Five millimeter discs of PDA overgrown with 14 day old fungi or 2 day old bacteria were placed in steamed soil under each of the five seeds of Beryl as well as of Jantar rape cultivars. At the stage of the first leaves the health state of rape plants was estimated. The species of the rape root pathogens were identified (Cowan, 1974; Kochman, 1973; Parmeter, 1970; Raikko, 1950).

The susceptibility of four rape cultivars to *F. Wollenweberii* and *R. solani* was estimated in the greenhouse. A mixture of corn meal (5 %) and soil (95 %) overgrown with 3 week old fungi was an inoculum. The inoculum in the amount of 2 mg was put close to each of the 20 rape seeds placed in steamed soil. At the stage of the first leaves the health state of the plants was estimated :

- 1 - healthy plants,
  - 2 - slightly
  - 3 - moderately
  - 4 - heavily
- } infected plants.
- 5 - no plants (sprouts destroyed in the soil).

For statistical analysis the numbers were transformed into degrees of Freeman-Tukey. The means were compared by the Duncan test.

## Results and discussion

In the field experiment at temperature and humidity conditions favourable to winter rape, the seed dressing with Oftanol T decreased root rot (Table 1). The numbers of rotting roots were similar in both the rape cultivars. The health state of rape roots was not dependent on quantity of the seeds sown.

Table 1

Occurrence of rape root rot in the successive terms  
(Złotniki, 1985 86)

| Terms of estimation | Percentage of plants with rotting roots, in case of seeds |                  |                   |                   |
|---------------------|---|------------------|-------------------|-------------------|
|                     | dressed   |                  | undressed         |                   |
|                     | Beryl   | Jantar           | Beryl             | Jantar            |
| I - 17.09.          | 0,0   | 0,0              | 0,0               | 0,0               |
| II - 14.10.         | 0,4   | 0,4              | 0,8               | 2,5               |
| III - 19.11.        | 0,0   | 0,0              | 14,1              | 4,1               |
| IV - 16.12.         | 0,0   | 0,4              | 16,2              | 8,3               |
| V - 21.01.          | 0,4   | 2,5              | 15,0              | 14,1              |
| VI - 17.03.         | 2,5   | 2,5              | 14,2              | 15,0              |
| VII - 14.04.        | 0,4   | 1,2              | 18,3              | 14,6              |
| VIII - 19.05.       | 2,1   | 4,6              | 15,0              | 18,3              |
| IX - 23.06.         | 11,7  | 7,9              | 24,1              | 27,5              |
| Mean <sup>x</sup>   | 1,9 <sup>a</sup>  | 2,2 <sup>a</sup> | 13,1 <sup>b</sup> | 11,9 <sup>b</sup> |

x - means followed by the same letter are not significantly different at 5 % level;

The most frequently isolated and pathogenic to rape roots was *Fusarium Wollenweberii* (Table 2). The next frequent was *Rhizoctonia solani*. Rotting degree of roots was higher in the case of *R.solani* (4,5) than in that of *F.Wollenweberii* (3,0).



Table 2

Pathogenic species of fungi and bacteria isolated from rotting rape roots (mean for Beryl and Jantar cultivars, seeds undressed and dressed with Oftanol T)

| Species   | Percentage of the whole number of pathogenic isolates | Mean degree of infection |
|---|---|--------------------------|
| <i>Fusarium Wollenweberii</i> Raijko            | 72,0  | 3,0                      |
| <i>Rhizoctonia solani</i> K.                    | 21,4  | 4,5                      |
| <i>Alternaria brassicicola</i> (Schw.) Wiltisch | 3,6   | 3,9                      |
| <i>Erwinia</i> sp.                              | 3,0   | 1,1                      |

In the greenhouse, all the four rape cultivars were susceptible to both species of fungi. The smallest number of diseased plants and a low degree of root infection by *F.Wollenweberii* and *R.solani* were observed in Beryl cultivar (Table 3).

Table 3

Susceptibility of rape to root rot fungi (Poznań, 1986)

| Rape cultivars | Percentage of infected plants <sup>x</sup> |                  | Mean degree of infection |                 |
|----------------|--|------------------|--------------------------|-----------------|
|                | <i>F.Wollenweberii</i>                     | <i>R.solani</i>  | <i>F.Wollenweberii</i>   | <i>R.solani</i> |
| Górczański     | 100 <sup>b</sup>                           | 95 <sup>cd</sup> | 2,4                      | 3,4             |
| Skrzeszowicki  | 90 <sup>b</sup>                            | 100 <sup>c</sup> | 1,9                      | 2,6             |
| Beryl          | 70 <sup>a</sup>                            | 90 <sup>d</sup>  | 1,9                      | 3,2             |
| Jantar         | 90 <sup>b</sup>                            | 100 <sup>c</sup> | 2,2                      | 3,9             |

x - means followed by the same letter are not significantly different at 5 % level;

In the field, where affected by other microorganisms and

pests, the numbers of plants with rotting roots mean for nine terms of observations were similar in Beryl and Jantar cultivars. In the greenhouse, where steamed soil with inoculum of one of the two fungi was used, Beryl ("O") proved to be less susceptible to *F. Wollenweberii* and *R. solani* than Jantar ("OO"), Górczański and Skrzyszowicki (high erucic).

Different levels of erucic and glucosinolate compounds in plants of rape did not correlate with susceptibility degree of the four cultivars to root rot pathogens.

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CYLINDROSPORIUM CONCENTRICUM (GREV.) : EVOLUTION  
DE LA MALADIE AU CHAMP ET METHODES DE CONTROLE.

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La première grave épidémie due à ce champignon est apparue en France au cours du printemps 1983. Dès la campagne suivante ont été mises en place des observations pour tenter de suivre les vagues de contaminations successives. Les difficultés avec cette maladie sont la grande lenteur de son développement et le très long délai entre la production des nécroses sur les jeunes organes floraux et leur apparition.

Des essais de lutte fongicide ont été réalisés, pour lesquels une méthode d'échantillonnage et de notation a été définie, qui est définie ici. La protection des organes reproducteurs du colza est obtenue au moyen d'un ou deux traitements au printemps à base de benzimidazoles ou prochloraz + benzimidazole.

Des progrès pourraient être faits dans la protection fongicide en se basant sur des résultats épidémiologiques.

- sans texte -

Histology of Primary Infection of Brassica Species  
by Albugo candida Race 7

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INTRODUCTION

Albugo candida (Pers. ex Lev.) Kuntze is an obligate parasite of cruciferous plants. Little is known of the mechanisms responsible for establishing and maintaining such a relationship, or how an incompatible relationship is determined when the fungus confronts host cultivars carrying resistance genes. Histological studies on the sequence of fungal pathogenesis on compatible and incompatible cultivars may reveal critical events in the early infection process. This information is essential for interpreting results of biochemical and physiological research on mechanisms of resistance.

Verma et al. (1975) studied the infection of four Brassica species (B. campestris, susceptible; B. napus, resistant; B. hirta, moderately susceptible and B. juncea, (susceptible) by A. candida. Pidskalny (1984) has described some aspects of the infection process in a compatible interaction between B. campestris cultivars and A. candida race 7. In these studies observations on fungal pathogenesis were initiated 24 h after inoculation. In fact, the initial 24 h is probably the critical period for the establishment of this obligate parasite. It is logical to begin observations at the time of inoculation and observe the earliest host-parasite interactions until a functional relationship between host and parasite either is or is not established. This study describes the sequence and timing of the pathogenesis of zoospores of A. candida race 7 on cotyledons of susceptible and resistant rapeseed lines.

MATERIALS AND METHODS

A. candida race 7 obtained from naturally occurring infections on B. campestris cv. Torch was used to inoculate B. napus cv. Regent, (resistant) and two lines, 2282-9 and GCL (susceptible) and B. campestris cv. Torch, which is highly susceptible to A. candida race 7. Plants were grown in a controlled environment chamber at day/night temperature of 22/17C under 16 h photoperiod. Inoculation was performed approximately 6 days after seeding. A 10  $\mu$ l droplet of zoospore suspension containing  $1 \times 10^7$  zoospores/ml was placed on the centre of the abaxial surface of each half cotyledon. The inoculated seedlings were incubated at 20C and 100% RH, for 24 h with initial 12 h in dark.

Whole cotyledons were sampled at 1, 2, 3, 4, 8, 12, 24, 36, 48, 60, 72, 84 and 96 h after inoculation. They were cleared and stained according to the methods of Bruzese and Hasan (1983). The cleared specimens were mounted in lactophenol and examined using differential interference contrast (DIC) microscopy. The following data were obtained at given sampling times: germ tube length, primary hyphal length, the number of haustoria per infection site and colony area. Only isolated infection sites were examined.

## RESULTS AND DISCUSSION

The sequential process of infection of *A. candida* on compatible rapeseed lines was encystment of zoospores on cotyledon surfaces, cyst germination and germ tube elongation, stomatal penetration, primary hyphal formation and elongation, haustorial development, secondary hyphal formation, mycelial ramification, sporangiophore formation, and sporangial production.

One hour after inoculation, the majority of zoospore cysts accumulated in the stomata on the upper epidermal surface. Zoospore cysts germinated within 2-3 h by producing single germ tubes. Stomatal penetration was soon initiated by the germ tubes (Figure 1). On entering the substomatal chamber, the germ tubes swelled to form a substomatal vesicle and then elongated to form primary hypha which extended to the palisade mesophyll. Primary haustoria usually formed at the tip of the young hyphae and were first detected in the palisade mesophyll cells adjacent to the substomatal chamber 8 h after inoculation (Figure 2). The haustoria were spherical ranging from 2.0 to 4.0  $\mu\text{m}$  in diameter and connected to the hyphae by narrow necks approximately  $1.8 \times 0.4 \mu\text{m}$ . Within 24 h after inoculation, secondary hyphae formed as side branches from the primary hyphae at the sites which previously gave rise to the initial haustoria.

On susceptible lines, primary haustoria formation followed by successful development of the secondary hyphae seems to represent the establishment of a functional relationship between rape and *A. candida*. The fungal growth accelerated dramatically resulting in ramification of hyphae as downward spirals into the spongy mesophyll. By 48 h, hyphal lengths in three susceptible lines investigated averaged 67  $\mu\text{m}$  with production of 1-8 haustoria per infection site (Table 1). Intercellular hyphal growth continued with more haustorial formation in mesophyll cells. By 84 h following inoculation, some of the fungal colonies became so extensive that they merged to form large compact mycelial masses. By 96 h, almost all the intercellular spaces were occupied by mycelia. Even at this stage, the host cells did not seem to be disrupted to any degree. Numerous club-shaped sporangiophores arose from the dense mycelial mat beneath the lower epidermis. White pustules were macroscopically visible within 5-6 days after inoculation.

The average germ tube lengths of 2282-9, GCL, Regent and Torch at 4 h after inoculation were 18.3, 17.4, 18.9 and 18.7  $\mu\text{m}$ , respectively. This implies that the incompatible line possesses no morphological or physiological features which could prevent cyst germination or subsequent penetration through stomata. Differences occurred between compatible and incompatible reactions 12 h after inoculation. The greatest increase in mean primary hyphal lengths occurred between 12-36 h for the compatible lines. On the resistant line fungal growth

did not increase significantly after this time. Hyphal growth ceased between 12 and 48 h after inoculation.

Haustoria were first formed in all lines at 8 h after inoculation. The average and maximum numbers of haustoria formed on each line at each time is shown in Table 2. Usually only one haustorium was formed per primary hypha on the resistant line. A resistant response, involving cell necrosis of the host, first appeared 12 h after inoculation (Figure 3). The number of infection sites showing cell necrosis then increased rapidly and amounted to 95% within 48 h. At this stage, it was no longer possible to make an accurate measurement of incompatible reaction since the fungal structure was obscured by collapsed host cells. Only the cells penetrated by haustoria became necrotic; adjacent uninfected cells and mesophyll cells below the dead cells remained healthy.

Fungal growth in the compatible lines was rapid following production of functional haustoria. Hyphae ramified intercellularly forming numerous secondary haustoria in the host mesophyll cells. As many as 44 haustoria per infection site were observed in Torch at 84 h after inoculation. More haustoria were present at 96 h or later, but the coalescence of fungal thalli made it impossible to obtain the accurate numbers of haustoria per infection site.

This study reveals that differences in reaction to A. candida race 7 between resistant and susceptible rapeseed lines only become pronounced after primary haustorial formation in the host palisade mesophyll cells. This suggests that successful formation of the primary haustoria is essential for the establishment and maintenance of a compatible relationship between A. candida and its host. The fact that the sequence and timing of pre- and post-penetration events were similar on resistant and susceptible lines implies that specific recognition is prerequisite to an incompatible reaction.

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Table 1. Effect of rapeseed lines and hours after inoculation on the length of primary hyphae.

| Hours after Inoculation | Mean length of primary hyphae (um) <sup>①</sup> |        |        |        |
|-------------------------|---|--------|--------|--------|
|                         | Regent  | GCL    | 2282-9 | Torch  |
| 8                       | 22.7 a*   | 21.6 a | 24.2 a | 24.4 a |
| 12                      | 23.6 a  | 25.1 a | 26.6 a | 28.5 a |
| 24                      | 24.3 a  | 39.4 b | 42.5 b | 45.1 b |
| 36                      | 24.8 a  | 60.0 c | 61.6 c | 63.5 c |
| 48                      | 25.0 a  | 66.4 c | 66.9 c | 68.2 c |

<sup>①</sup> Average of four replications and five sampling units except that values of Regent at 36 and 48 hours after inoculation were average of 18 and 1 observations, respectively due to necrosis of host cells.

\* Values in each column and each row followed by different letters differ significantly according to Duncan's Multiple Range Test, P=0.05.

Table 2. Mean and maximum numbers of haustoria per infection site in cotyledons of susceptible and resistant rapeseed lines.

| Hours | Line  |      |        |      |      |      |        |      |       |
|-------|-------|------|--------|------|------|------|--------|------|-------|
|       | Torch |      | 2282-9 |      | GCL  |      | Regent |      |       |
|       | Mean  | Max. | Mean   | Max. | Mean | Max. | Mean   | Max. | Nec.* |
| 8     | 0.7   | 1    | 0.7    | 1    | 0.5  | 1    | 0.6    | 1    | 0 %   |
| 12    | 1.0   | 1    | 0.8    | 1    | 0.7  | 1    | 0.7    | 1    | 26 %  |
| 24    | 1.1   | 2    | 0.9    | 2    | 0.9  | 1    | 0.3    | 1    | 53 %  |
| 36    | 1.6   | 3    | 1.5    | 3    | 1.3  | 3    | 0.2    | 1    | 80 %  |
| 48    | 3.1   | 8    | 1.7    | 3    | 1.5  | 3    | 0.2    | 2    | 95 %  |
| 60    | 6.3   | 16   | 4.8    | 9    | 4.9  | 10   | --     | --   | --    |
| 72    | 6.0   | 18   | 5.8    | 12   | 5.6  | 12   | --     | --   | --    |
| 84    | 22.8  | 44   | 19.9   | 38   | 13.5 | 31   | --     | --   | --    |

<sup>①</sup> Observations made on 5 infection sites on each of 4 cotyledons for all 4 rapeseed lines.

\* Percentage of infected host cells which were necrotic.

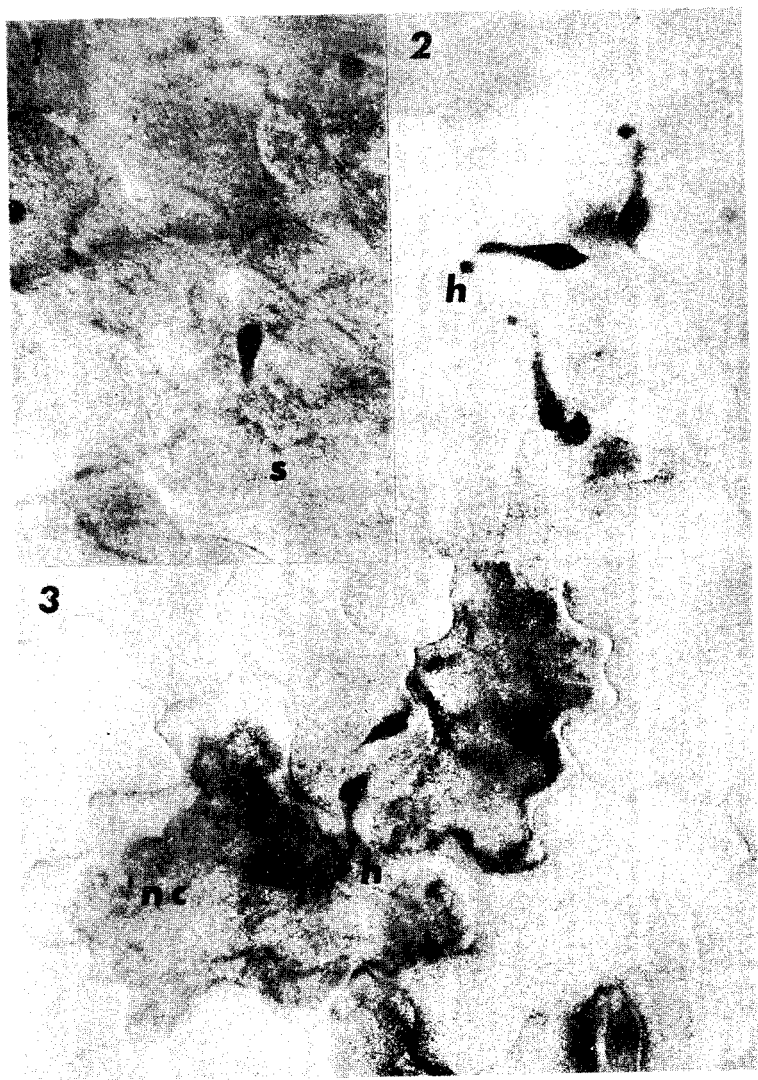


Figure 1. Encysted zoospore with germ tube and penetration of germ tube into host stomate (3h after inoculation).

Figure 2. Primary haustoria formed in mesophyll cells from tips of germ tubes (8h after inoculation).

Figure 3. Host cell necrosis on cv. Regent following primary haustorial formation (12h after inoculation).



Confirmation of a Digenic Model of Inheritance of Resistance  
to Albugo candida Race 7 in Brassica napus

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INTRODUCTION

White rust, caused by Albugo candida (Pers. ex Lev.) Ktze., is a major hazard to the production of turnip rape (Brassica campestris L.) and of brown mustard (B. juncea L.) in western Canada and other regions of the world. Average yield losses of turnip rape in Alberta and Saskatchewan due to white rust were reported to be between 1.2 and 9.0% (Berkenkamp, 1971; Petrie, 1973). In Manitoba, yield reduction ranging from 30 to 60% occurred in heavily infected fields (Bernier, 1972).

In western Canada, commercial varieties of summer rape (B. napus) are highly resistant to A. candida. Moreover, resistance in B. napus has remained effective over 40 years of continuous cultivation. In central and eastern China, however, some cultivars are quite susceptible. In a field survey of the Shanghai region in 1973, more than 70% of the plants had systemically infected inflorescences (stagheads) causing yield losses between 20 and 30%.

A model of inheritance of resistance to white rust in B. napus has been proposed by Fan et al. (1983). In this model resistance is conditioned by three independent dominant genes designated Ac7-1 ( $R_1$ ), Ac7-2 ( $R_2$ ), and Ac7-3. The first two genes are homozygous in cultivar Regent. As a single dominant allele at any one of the three loci is sufficient to confer resistance, the heterozygosity or heterogeneity of the third locus tends to be masked.

In the present study,  $F_1BC_1$  plants from 2282-9 (susceptible) x Regent (resistant) x 2282-9 were developed and tested to confirm the digenic model with dominant resistance conferred by Ac7-1 and Ac7-2.

MATERIALS AND METHODS

The experiment was conducted in the growth cabinet and greenhouse during 1985-1986. Seedlings from the backcross (2282-9 x Regent) x 2282-9 were tested for resistance to A. candida race 7 by inoculating cotyledons with zoospores. Resistant  $F_1BC_1$  plants were selected and grown to flower. While one inflorescence of each selected plant was self-pollinated, another was backcrossed to the susceptible line 2282-9.

The genotype of each resistant  $F_1BC_1$  plant was determined by inoculating the cotyledons of  $F_2BC_1$  and  $F_2BC_2$  plants with race 7. Resistant plants were selected from the  $F_2BC_1$  progenies which had

segregated in the ratio of 3 resistant to 1 susceptible. They were self-pollinated and backcrossed to 2282-9. Plants of genotype  $R_1R_1r_2r_2$  and  $r_1r_1R_2R_2$  could be obtained when  $F_3BC_1$  and  $F_2BC_2$  populations were tested for white rust resistance. Non-segregating resistant progenies were considered to be derived from the  $F_2BC_1$  parent of homozygous dominance, and segregating progenies were assumed to be derived from the  $F_2BC_1$  parent of heterozygous dominance.

To determine the genotype of each  $F_3BC_1$  accession, one established accession was assumed to be  $R_1R_1r_2r_2$  and used as a tester to cross with the others. Each accession was divided into two subunits. As they were derived from same  $F_3BC_1$  plant, paired subunits were similar genetically and expected to give identical results when crossed with the tester. Also, when paired subunits were sib-mated and then self-pollinated, subsequent progenies should not segregate unless mutation had occurred. The objective of employing two subunits for each  $F_3BC_1$  accession was to check the validity of experimental results.

$F_1$  progenies from the crosses (Accession 1, 2, ... 8 x tester) were self-pollinated and backcrossed to 2282-9 once more. The genotype of each accession was then determined according to the segregation ratios of the  $F_2$  progenies and verified by the corresponding backcross data. The digenic model could be confirmed if some of the progenies segregated into 15:1 ( $F_2$ ) ratio and 3:1 ( $BC_1$ ) ratio. The Chi-square test was used for analysis of the data from the segregating progenies.

#### RESULTS AND DISCUSSION

Progeny from the backcross (2282-9 x Regent) x 2282-9 segregated with the ratio of 3 resistant to 1 susceptible. The genotypes of the  $F_2BC_1$  plants were considered as  $R_1r_1R_2r_2$ ,  $R_1r_1r_2r_2$ ,  $r_1r_1R_2r_2$  and  $r_1r_1r_2r_2$ . When the resistant  $F_2BC_1$  plants were self-pollinated and backcrossed to the susceptible line 2282-9, some resulting progenies segregated into 15:1 and 3:1 ratios respectively, while others segregated into 3:1 and 1:1 ratios. This can be explained by assuming that the former were derived from the  $F_2BC_1$  parent of genotype  $R_1r_1R_2r_2$ , whereas the latter were derived from the  $F_2BC_1$  parent of genotype  $R_1r_1r_2r_2$  or  $r_1r_1R_2r_2$ . Resistant plants were selected from  $F_2BC_1$  progenies (Accession numbers 1 to 9) which segregated for white rust resistance in the ratio of 3 resistant to 1 susceptible and advanced to the  $F_3BC_1$ . The observed segregations and Chi-square tests for these nine accessions ( $F_2BC_1$ ) are shown in Table 1. The data from the corresponding backcross progenies ( $F_2BC_1$ ) are given in Table 2. Approximately, one third of the resistant plants in each selected  $F_2BC_1$  progeny were homozygous dominant at either of the two loci ( $R_1R_1r_2r_2$  or  $r_1r_1R_2R_2$ ) involved in white rust resistance. Accessions of these two genotypes were obtained by inoculating  $F_3BC_1$  and  $F_2BC_2$  plants.

One  $F_3BC_1$  was assumed to be  $R_1R_1r_2r_2$  and used as parent for test crosses with the other accessions. The resulting  $F_1$  progenies from the crosses (Accession 1, 2, 3, ... 8 x Tester) were all resistant to white rust. So were the progenies from the sib-mating between paired subunits ( $F_3BC_1$ ).

The  $F_2$  progenies from the crosses of the tester with Accession 1, 2, 3 and 4 segregated for white rust resistance in the ratio of 15 resistant to 1 susceptible. The results are a good fit ( $P > 0.05$ ) to the ratio expected for a segregation of two independent dominant genes

(Table 3). These data indicate that resistance in the four accessions was conferred by a pair of homozygous dominant alleles at the second locus ( $r_1 r_1 R_2 R_2$ ). We have, therefore, conclusive evidence that  $R_1$  and  $R_2$  are two non-allelic resistance genes at two discrete loci.

Progenies from the backcross (Accession 1, 2, 3 and 4 x Tester) x 2282-9 segregated with the ratio of 3 resistant to 1 susceptible ( $P > 0.05$ ) (Table 4), thus confirming the non-allelism of gene  $R_1$  and  $R_2$ .

The  $F_2$  progenies from the crosses of the tester with Accession 5<sup>2</sup>, 6, 7 and 8<sup>2</sup> were all resistant (Table 9), indicating that resistance in these four accessions was conditioned by a pair of homozygous dominant alleles at the first locus ( $R_1 R_1 r_2 r_2$ ). In other words, these accessions had the same genotype as the tester. This was confirmed by the data from the backcross (Accession 5, 6, 7 and 8 x Tester) x 2282-9 (Table 5).

From these data, the digenic model with dominant resistance conferred by  $R_1$  and  $R_2$  has been confirmed. Presence of a dominant allele at either of the two loci will confer resistance to a plant, whereas homozygous recessives at both loci will result in a susceptible phenotypical expression.

In Canada, resistance in B. napus cultivars to A. candida appears to be so durable that it has remained effective over 40 years of exposure to the isolates of A. candida which can attack B. campestris and B. juncea. This can be ascribed to the number of resistance genes carried by B. napus cultivars and/or the low capacity of the pathogen to adapt to the resistance genes in B. napus. Even so, rapeseed breeders should be cautious not to introduce susceptibility from Oriental cultivars or through interspecific crosses between B. napus and B. campestris.

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Table 1. Observed segregations and Chi-square tests for  $F_2BC_1$  data from 2282-9 x Regent involving resistance (R) and susceptibility (S) to A. candida race 7.

| Accession              | Reaction |    | Ratio | $\chi^2$ | P       |
|------------------------|----------|----|-------|----------|---------|
|                        | R        | S  |       |          |         |
| 1                      | 20       | 6  | 3:1   | 0.051    | .75-.90 |
| 2                      | 44       | 11 | 3:1   | 0.733    | .25-.50 |
| 3                      | 52       | 9  | 3:1   | 3.415    | .05-.10 |
| 4                      | 65       | 15 | 3:1   | 1.667    | .10-.25 |
| 5                      | 30       | 9  | 3:1   | 0.077    | .75-.90 |
| 6                      | 41       | 12 | 3:1   | 0.157    | .50-.75 |
| 7                      | 16       | 4  | 3:1   | 0.267    | .50-.75 |
| 8                      | 25       | 5  | 3:1   | 1.111    | .25-.50 |
| 9                      | 69       | 20 | 3:1   | 0.303    | .50-.75 |
| Total                  | 362      | 91 | 3:1   | 7.781    |         |
| Deviation $\chi^2$     |          |    |       | 5.829    | .01-.03 |
| Heterogeneity $\chi^2$ |          |    |       | 1.952    | .97-.99 |

<sup>a</sup>Each  $F_2BC_1$  population was derived from a single identified  $F_1BC_1$  plant.

Table 2. Observed segregations and Chi-square tests for  $F_1BC_2$  data from [(2282-9 x Regent) x 2282-9] x 2282-9 involving resistance (R) and susceptibility (S) to A. candida race 7.

| Accession <sup>a</sup> | Reaction |     | Ratio | $\chi^2$ | P       |
|------------------------|----------|-----|-------|----------|---------|
|                        | R        | S   |       |          |         |
| 1                      | 19       | 19  | 1:1   | 0.000    | >.99    |
| 2                      | 15       | 16  | 1:1   | 0.032    | .75-.90 |
| 3                      | 19       | 15  | 1:1   | 0.735    | .25-.50 |
| 4                      | 22       | 22  | 1:1   | 0.000    | >.99    |
| 5                      | 19       | 18  | 1:1   | 0.027    | .75-.90 |
| 6                      | 26       | 20  | 1:1   | 0.783    | .25-.50 |
| 7                      | 17       | 25  | 1:1   | 1.524    | .10-.25 |
| 8                      | 16       | 11  | 1:1   | 0.926    | .25-.50 |
| 9                      | 17       | 17  | 1:1   | 0.000    | >.99    |
| Total                  | 170      | 163 | 1:1   | 4.315    |         |
| Deviation $\chi^2$     |          |     |       | 0.147    | .50-.70 |
| Heterogeneity $\chi^2$ |          |     |       | 4.168    | .75-.90 |

<sup>a</sup>The accession numbers correspond to those in  $F_2BC_1$ , indicating that both populations were derived from the same maternal parent.

Table 3. Observed segregations and Chi-square tests for  $F_2$  data from crosses  $F_3BC_1$  x Tester involving resistance (R) and susceptibility (S)<sup>1</sup> to *A. candida* race 7.

| Accession <sup>a</sup> | Reaction |    | Ratio | $\chi^2$ | P       |
|------------------------|----------|----|-------|----------|---------|
|                        | R        | S  |       |          |         |
| 1 (A)                  | 94       | 5  | 15:1  | 0.244    | .50-.75 |
| 1 (B)                  | 140      | 10 | 15:1  | 0.044    | .75-.90 |
| 2 (A)                  | 276      | 19 | 15:1  | 0.018    | .75-.90 |
| 2 (B)                  | 272      | 16 | 15:1  | 0.237    | .50-.75 |
| 3 (A)                  | 208      | 10 | 15:1  | 1.031    | .25-.50 |
| 3 (B)                  | 210      | 11 | 15:1  | 0.610    | .25-.50 |
| 4 (A)                  | 83       | 5  | 15:1  | 0.048    | .75-.90 |
| 4 (B)                  | 142      | 8  | 15:1  | 0.217    | .50-.75 |
| Total                  | 1425     | 84 | 15:1  | 2.449    |         |
| Derivation $\chi^2$    |          |    |       | 1.202    | .25-.50 |
| Heterogeneity $\chi^2$ |          |    |       | 1.247    | .95-.99 |

<sup>a</sup>The accession numbers correspond to those in  $F_2$ , indicating that both populations were derived from the same maternal parent.

Table 4. Observed segregations and Chi-square tests for backcross data from ( $F_3BC_1$  x Tester) x 2282-9 involving resistance (R) and susceptibility (S) to *A. candida* race 7.

| Accession <sup>a</sup> | Reaction |    | Ratio | $\chi^2$ | P       |
|------------------------|----------|----|-------|----------|---------|
|                        | R        | S  |       |          |         |
| 1 (A)                  | 26       | 4  | 3:1   | 2.178    | .10-.25 |
| 1 (B)                  | 25       | 7  | 3:1   | 0.044    | .75-.90 |
| 2 (A)                  | 40       | 10 | 3:1   | 0.667    | .25-.50 |
| 2 (B)                  | 31       | 8  | 3:1   | 0.419    | .50-.75 |
| 3 (A)                  | 43       | 11 | 3:1   | 0.617    | .25-.50 |
| 3 (B)                  | 28       | 6  | 3:1   | 0.980    | .25-.50 |
| 4 (A)                  | 47       | 14 | 3:1   | 0.137    | .50-.75 |
| 4 (B)                  | 64       | 19 | 3:1   | 0.197    | .50-.75 |
| Total                  | 304      | 79 | 3:1   | 5.239    |         |
| Deviation $\chi^2$     |          |    |       | 3.907    | .03-.05 |
| Heterogeneity $\chi^2$ |          |    |       | 1.332    | .95-.99 |

<sup>a</sup>The accession numbers correspond to those in  $F_2$ , indicating that both populations were derived from the same maternal parent.

Table 5. Reaction of  $F_2$  and  $F_1BC_1$  from the crosses  $F_3BC_1 \times$   
Tester to A. candida race 7

| Accession | Reaction |   |           |   |
|-----------|----------|---|-----------|---|
|           | $F_2$    |   | $F_1BC_1$ |   |
|           | R        | S | R         | S |
| 5 (A)     | 140      | 0 | 30        | 0 |
| 5 (B)     | 138      | 0 | 55        | 0 |
| 6 (A)     | 145      | 0 | 34        | 0 |
| 6 (B)     | 128      | 0 | 35        | 0 |
| 7 (A)     | 191      | 0 | 45        | 0 |
| 7 (B)     | 152      | 0 | 39        | 0 |
| 8 (A)     | 149      | 0 | 35        | 0 |
| 8 (B)     | 141      | 0 | 35        | 0 |

ESTIMATION METHODS AND RESISTANCE OF WINTER RAPE CULTIVARS  
TO THE DRY ROT AND STEM CANCKER /Phoma lingam;  
Leptosphaeria maculans/ IN A GREENHOUSE TEST AND IN FIELD  
TRIALS RESEMBLING NATURAL INFECTION CONDITIONS

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#### INTRODUCTION

These studies were conducted as a continuation of re-  
search programme of fungal diseases occurring in Polish  
winter rape cultivation conditions in the aspect of their  
economic problem and plant resistance breeding /Frencel  
1983/. The aim of the present part of the studies was to  
investigate the cultivar differences in the "field" resis-  
tance to Phoma lingam/Leptosphaeria maculans, as well as  
to work out a greenhouse test suitable for early selection  
of plant breeding materials. It was also intended to veri-  
fy and correlate the two different resistance estimation  
methods, as we stress that the reliability of the experi-  
mental infection under artificial inoculation at the seed-  
ling and/or the young plant stage used in routine resis-  
tance breeding tests should be well documented.

#### MATERIALS AND METHODS

##### I. Field trials.

**P l a n t s** . The studies were carried out on 16 winter  
oilseed rape cultivars: Beryl, Brink, Doral, Emil,  
Górczański, Herkules, Janpol, Jet Neuf, Jupiter, Librador,  
Ligora, Marinus, Primor, Quinta, Skrzyszowicki, Start.

In the experiments the method of random blocks was used with  $2m^2$  plots, each in four replications. The sowing date as a rule was the end of the second decade of August /1982, 1983, 1984/ and the sowing rate was 6 kg/ha.

The field trials were carried out in the north-western Poland, one of the intensive winter rape cultivation regions, with a high potential of Phoma - disease incidence under natural condition. Moreover, in autumn, at the leaf-rosette stage of plants, a preliminary infection was provoked by dispersing post-harvest rape straw from the infected plantation, approximately at about 1 fragment/  $15-20cm^2$  of the experimental field.

Estimation of cultivar susceptibility and resistance /infection degree/. Each time throughout the vegetation period three to four general observations and one main - immediately prior to harvesting - were made. Disease lesions on the stems were classified according to a grade scale 0 - 6 /healthy plant - the highest infection/.

## II. Greenhouse experiments.

Plants. The following 8 winter rape cultivars were used in greenhouse investigations: Doral, Jet Neuf, Jupiter, Górczański, Primor, Skrzyszowski, Start, Quinta.

Inoculum. The inoculum consisted of three week fungus cultures, maintained on the solid sandy-oat medium /52g sand + 45g gravel + 37,5g rough-grinded oat meal + 37,5ml water/. Three isolates /Ph<sub>0</sub>, Ph<sub>1</sub>, Ph<sub>2</sub>/ of *Phoma lingam* /Tode ex Fr./ Desm., stad. gener. *Leptosphaeria maculans* /Desm./ Ces. et de Not., differing in pathogenicity were used as pathogens. Single-spore pathogen cultures were originated from *Leptosphaeria* /Ph<sub>0</sub> and Ph<sub>2</sub>/ or from *Phoma* /Ph<sub>1</sub>/. Immediately before using, the outgrown mycellium - culture medium was disintegrated.

Inoculation method. The inoculum was introduced into the soil in pots /50cm<sup>3</sup> per pot/, after which rape seeds were sown into the infected soil.

Three different concentrations of the inoculum were



compared: 1/ the standard culture = nondiluted, 2/ sandy-diluted 1 : 1, 3/ sandy-diluted 1 : 2.

15 seeds per pot were inoculated. Each experiment was performed in three parallel replications and in three individual series during the spring time /March - April/ in a greenhouse, where the temperature ranged from about 22°C /day/ to 16°C /night/, under the satisfied humidity.

Three weeks after seed emergence and plant growth they were estimated for their response to experimental infection. Plants were taken out of the soil, washed and the infection degree was estimated on the 0 - 3 grade scale /healthy plant to the highest infection/. The plants were weighed and their dry mass was also taken as a response parameter against the control /healthy noninoculated plants/.

## RESULTS AND DISCUSSION

### I. Field trials.

During the same vegetative season particular rape cultivars differed in the Phoma infection degree. Simultaneously, the analysis of variance documented the existence of statistically significant intercultivar differences in the vegetation season of 1982/83 and 1983/84. In 1984/85 the statistical significance for intervarietal differences was not proved, nevertheless the same hierarchy of the infection degree between the cultivars was maintained /tab. 1/. Highly significant correlation coefficients of the cultivar infection were found in different years of the trials /tab. 2/.

### II. Greenhouse experiments.

Because of a short form of this report, a detailed documentation of the results will be published elsewhere. In general, on the basis of the statistical analyses it may be inferred that dry mass of inoculated plants / in % of the control/ better differentiated individual cultivars than the mean infection degree as response parameter, according to the grade scale of disease symptoms. The ana-

lysis of variance also showed a significant influence of different isolates of *Phoma lingam* interacting with different cultivars. Also, the severity of disease symptoms on the investigated cultivars appeared to be highly correlated with the inoculum concentration, as indicated by highly significant correlation coefficients for the dry mass as a response parameter.

On the basis of the correlation coefficients between the infestation degree of cultivars under "natural" infection /in field trials/ and under the experimental infection /artificial inoculation in greenhouse test/ it may be suggested, that the both estimation methods of disease response are principally comparable /table 2/.

#### CONCLUSIONS

The results of differences of cultivar responses to *Phoma lingam* under the field "natural" high potential infection conditions were found to be statistically significant. In general, the Polish conditions of rape cultivation appeared to be comparable to those of West European countries regarding susceptibility and resistance response of rape cultivars. Like in other countries, Jet Neuf and Doral, for instance, are the most resistant, whereas Primor, Quinta, Marinus, Start are the most susceptible /Krüger 1983/. The most resistant and the most susceptible cultivars were more stable in their response to infection during each year of experiment. Cultivars of "medium" response deviated more or less in their susceptibility/resistance responses.

In the years of the performed experiments no visible symptoms of early infection /autumn/ and disease phase of leaf spot were found. Infection on plants became manifest usually from spring onward throughout the vegetative season. In observations during early spring, immediately after overwintering and onset of vegetation, only sporadic local infection patches with a large intensity of disease symptoms, suggesting the effects of autumn infection of plants, were detected.

There were some statistically significant coefficients between the results of glasshouse experiments and field trials for the infection degree as the reaction parameter. It suggests, that the resistance tests, at least at the young plant level, if they are not at the seedling stage, are reliable and may be used in plant breeding selection, instead of the resistance tests conducted on grown plants. Further studies intending to standardization of resistance breeding tests are under way.

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Table 1: Infection degree /in %<sup>x</sup>/ of winter rape cultivars under natural high potential infection of *Phoma lingam*/*Leptosphaeria maculans* in field trials. Results of the experiments in 1982/83, 1983/84 and 1984/85

| Cultivar      | vegetative season |         |         |
|---------------|-------------------|---------|---------|
|               | 1982/83           | 1983/84 | 1984/85 |
| Doral         | 6,67              | 10,50   | 9,83    |
| Jet Neuf      | 7,50              | 7,17    | 12,67   |
| Beryl         | 7,50              | 9,83    | 14,50   |
| Górczański    | 14,17             | 14,83   | 13,30   |
| Primor        | 29,17             | 31,33   | 17,33   |
| Brink         | 15,00             | 17,17   | -       |
| Quinta        | 21,67             | -       | 17,67   |
| Janpol        | 22,50             | -       | -       |
| Start         | 44,17             | -       | -       |
| Jupiter       | -                 | 12,83   | 10,00   |
| Skrzeszowicki | -                 | 15,50   | 15,33   |
| Herkules      | -                 | 13,17   | -       |
| Marinus       | -                 | 30,50   | -       |
| Librador      | -                 | 16,67   | -       |
| Ligora        | -                 | 17,50   | -       |
| Emil          | -                 | -       | 16,67   |

x/

$$\text{Infection degree in \%} = \frac{\sum /n \cdot v/ \cdot 100}{V \cdot N}$$

where:

- n = number of plants in the particular infection degree /0-6/  
 N = the whole number of plants studied  
 v = the particular infection degree  
 V = the highest infection degree

Table 2: Correlation coefficients between the infection degree /*Phoma lingam*; *Leptosphaeria maculans*/ of winter rape cultivars. Comparisons of results in "natural" field infection and artificial inoculation in glasshouse experiments

| Parameter       | Field trials |          |        | Glasshouse tests - pathogen isolates |                 |                 |
|-----------------|--------------|----------|--------|--------------------------------------|-----------------|-----------------|
|                 | 82/83        | 83/84    | 84/85  | Ph <sub>1</sub>                      | Ph <sub>0</sub> | Ph <sub>2</sub> |
| 82/83           | -            | 0,988*** | 0,867  | 0,819*                               | 0,722           | 0,650           |
| 83/84           | -            | -        | 0,761* | 0,863**                              | 0,509           | 0,691           |
| 84/85           | -            | -        | -      | 0,894**                              | 0,887**         | 0,861**         |
| Ph <sub>1</sub> | -            | -        | -      | -                                    | 0,659           | 0,847**         |
| Ph <sub>0</sub> | -            | -        | -      | -                                    | -               | 0,778*          |

\*\*\* P = 0,1 %

\*\* P = 1 %

\* P = 5 %

A FIELD STUDY OF RAPESEED (BRASSICA NAPUS)  
RESISTANCE TO SCLEROTINIA SCLEROTIORUM

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INTRODUCTION.

*Sclerotinia sclerotiorum* is an important disease agent for oilseed rape grown in France. Increasingly intensive exploitation of this crop, and the introduction into the rotation of others also susceptible such as sunflower, faba beans, peas, soya beans and beans, may favour increase in sclerotes abundance in soil, thus risk of epidemics and gravity of attack. A certain number of fungicides can be used to appreciably reduce parasite incidence (CETIOM, 1987). However, the sporadicity in disease outbreak and difficulties in establishment of an efficient predicting system, result in a certain amount of useless applications. Furthermore, increase, in the practise of applying treatments at the onset of flowering may favour appearance of strains resistant to fungicides. Commercialisation of oilseed rape cultivars resistant to the fungus may usefully offset these problems, and provide a means of combatting the disease. The existence of genotypes having an appreciable potential for resistance to *S. sclerotiorum* has been demonstrated in a variety of species [e.g. *Phaseolus vulgaris* var. Ex. Rico (TU and BEVERSDORF, 1982) and *Helianthus annuus* (TOURVIELLE and VEAR, 1984). The aim of the present study was to study genetic variability in oilseed rape resistance to *S. sclerotiorum* with different inoculation conditions.

MATERIALS AND METHODS.

Natural and artificial inoculation techniques were used to detect oilseed genotypes resistant to *S. sclerotiorum*.

1. Natural inoculation in field crops : Different oilseed genotypes were planted in a complete block design in plots (four blocks, variable treatment number) which had previously carried bean crops, sensitive to and attacked by *S. sclerotiorum* . Experimental plots were located in the department of Finistere, where climate is oceanic, and springs usually moist and mild. Most data is presented as percentage plants showing symptoms of fungus attack.

2. Artificial inoculation in field crops : Several methods were compared. Only those which resulted in high infection levels and were practically easy to apply were retained.

a) Inoculation with ascospores under aluminium paper.

Sclerotes were obtained from infected oilseed plants, or from sunflower heads or sliced carrot. They were placed in pots with slightly humid perlite, at 2 cm depth, and at 10°C . Two months later, sclerotes

were extracted, stipes removed, and they could then be kept for up to two years at 4°C. They were placed on wet cotton in 5 cm diameter covered glass dishes. Apothecia produced the first ascospores at 15 days, in daylight, and at 20°C, and these were recovered and stored following (STEADMAN, 1974). An ascospore suspension in sterile water was prepared by scraping on 2 µ micropore filter, then ultrason treatment for 7 seconds.

Inoculation flower (BRUN and al., 1981) involved insertion of a 5 mm diameter filter paper pastel laid on a flower petal to ensure infection at the base of a leaf. Pastels were wetted with 30 µl of a  $5 \cdot 10^5$  spore/ml suspension, prior to insertion. They were covered with a slightly moistened pad of cotton wool, and the whole envelopped in aluminium paper.

b) Inoculation with mycelium in matchstick fragments.

Short fragments of specially furnished crude matchsticks (5 cm) were sterilised twice at 120°C at a 24 hr interval, in a 2% malt solution. They were then placed in 14 cm diameter petri dishes, after slight drying on filter paper. Dishes contained 4-day cultures of *S. sclerotiorum* on a 2% malt / 2% gelose water medium, and cultures then held at 20°C for eight days. Match fragments were then inserted in oilseed rape stems in a cut at 30 cm height (fig. 1).

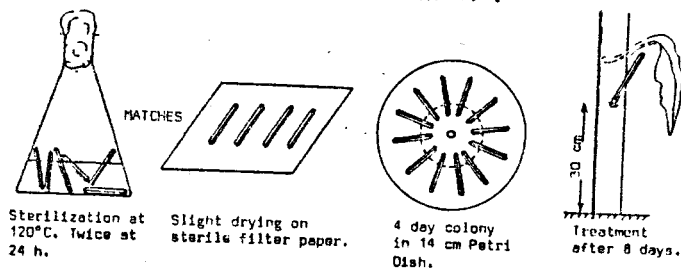


Fig. 1. INOCULATION USING "MATCHES" IMPREGNATED WITH *S. SCLEROTIORUM* MYCELIUM.

c) Experimental apparatus and scoring.

A single inoculation was made per plant, and 30 plants inoculated per block (four replicates per block). At three weeks percentage plants with overt lesions and length of apparent mould were scored.

3. Artificial contamination in green houses.

Overwintered plants (rosettes) were taken from field plots. Collection was staggered to obtain regrowth at different times. Gelose discs were extracted from the edges of experimental cultures of *S. sclerotiorum*. These were placed on subject stems (one per stem) at 30 cm height and covered with a strip of parafilm.

Twelve plants were inoculated per growth stage, with three replicates per growth stage. Scores concerned percent infected plants, and lengths of apparent stem necrosis.

RESULTS.

1. Natural infections.

Plant resistance mechanisms are best understood under natural conditions without artificial intervention. However, the limits of natural infection as a means of studying oilseed resistance to *S. sclerotiorum* are now apparent after eight years of study. With high infection levels,

varietal ranking is not consistent from year to year (fig. 2). The reason for this may be the high intervarietal variability in degree of early flowering for oilseed cultivars. Because of this, all genotypes are not necessarily or invariably subjected to the same number of contaminatory phases (conjunction of ascospores, petals, favorable climatic conditions). Moreover, despite being in an area that is generally humid, infection levels have been too low in some years (attributable to particularly dry spells) to allow varietal ranking.

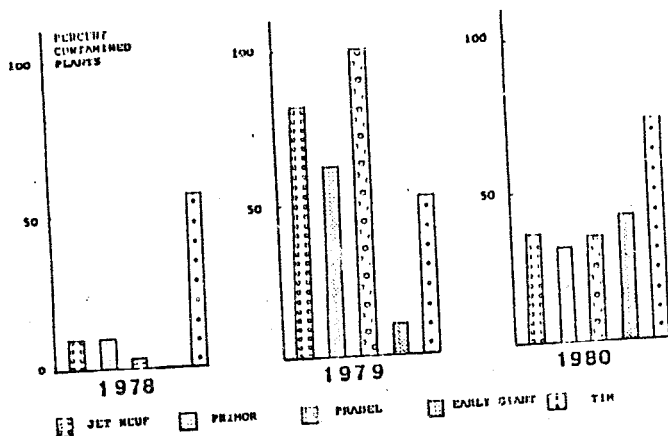


Fig. 2. VARIETAL RANKING OF FIVE GENOTYPES UNDER NATURAL INFECTION IN THE FIELD.

## 2. Artificial infection.

The two artificial inoculation techniques confirm low variability in resistance to *S. sclerotiorum* of oilseed rape varieties currently grown in France (or being introduced to the french catalogue CTPS) (Table 1). This contrasts with major differences between genotypes of asiatic origin (Table 2).

Good performance of Norin 9 has been confirmed over several years of experimental study, both in terms of percent infection and length of apparent necrosis. However, for some genotypes there may be a correlation between resistance performance and flowering precocity. This possibility was tested by simultaneous inoculation of Norin 9, Jet Neuf, and Bienvenu in green house trials. Results (Table 3) indicate that growth phase of Norin 9 was not a factor affecting fungus development in stems.

## DISCUSSION.

Results of the present study indicate the need for artificial inoculation techniques in study of oilseed rape cultivar variability in resistance to *S. sclerotiorum*, and search for resistance factors. The two artificial inoculation techniques suggest a possible difference in resistance potential of asiatic oilseed rape strains in comparison to those currently grown in France. Inoculation using matchstick fragments has proved more rapid in application and production of research results, than that using ascospores, and may be more useful in selection studies. Further work will be required to determine the nature of Norin 9 resistance, and possibilities for its transfer to winter varieties.

| INNOCULATION WITH ASCOSPORE SUSPENSION |                                  |          |   |
|--|----------------------------------|----------|---|
| Genotypes                              | % plants attacked<br>(arc sin %) | Stages • | Newman and Keuls<br>Test for<br>homogeneity |
| 1 NORIN 9                              | 16,09                            | G3       | A   |
| 2 KID                                  | 47,91                            | G2       | B   |
| 10 PR 18                               | 52,72                            | G1       | B C   |
| 12 DORAL                               | 56,44                            | F2 (G1)  | B C   |
| 5 JET NEUF                             | 58,09                            | G1       | B C   |
| 8 MIKADO                               | 58,68                            | G2       | B C   |
| 9 EGK 1002                             | 62,79                            | F2 (G1)  | B C   |
| 4 DARMOR                               | 64,02                            | F2       | B C   |
| 3 BIENVENU                             | 68,74                            | G2       | B C   |
| 11 PERLE                               | 72,06                            | F2       | B C   |
| 7 BELINDA                              | 74,17                            | F2 (G1)  | B C   |
| 6 KORINA                               | 76,49                            | G1 (G2)  | C   |
| CVM %                                  | 18,9                             |          |   |

| INNOCULATION WITH MYCELIUM ON MATCHES |                                  |          |   |
|---------------------------------------|----------------------------------|----------|---|
| Genotypes                             | % plants attacked<br>(arc sin %) | Stages • | Newman and Keuls<br>Test for<br>homogeneity |
| 1 NORIN 9                             | 10,59                            | G3       | A   |
| 8 MIKADO                              | 52,20                            | G2       | B   |
| 2 KID                                 | 54,21                            | G2       | B   |
| 3 BIENVENU                            | 59,29                            | G2       | B   |
| 12 DORAL                              | 62,82                            | F2 (G1)  | B   |
| 9 EGK 1002                            | 63,54                            | F2 (G1)  | B   |
| 4 DARMOR                              | 65,89                            | F2       | B   |
| 10 PR 18                              | 66,18                            | G1       | B   |
| 11 PERLE                              | 68,14                            | F2       | B   |
| 5 JET NEUF                            | 71,51                            | G1       | B   |
| 7 BELINDA                             | 71,52                            | F2 (G1)  | B   |
| 6 KORINA                              | 80,70                            | G1 (G2)  | B   |
| CVM %                                 | 21,2                             |          |   |

• CETIOM, 1987

TABLE 1 : BEHAVIOUR OF CULTIVATED VARIETIES OR THOSE BEING INTRODUCED TO THE FRENCH CATALOGUE.



| ASCOSPORE INNOCULATION UNDER ALUMINIUM |                                  |          |   |
|--|----------------------------------|----------|---|
| Genotypes                              | % plants attacked<br>(arc sin %) | Stages * | Newman and Keuls<br>Test for<br>homogeneity |
| 5 GENKAI                               | 29,99                            | G2       | A   |
| 4 MIYUKI                               | 30,32                            | G2       | A   |
| 6 ISUZU                                | 33,90                            | G2       | A B   |
| 1 NORIN 9                              | 34,92                            | G2       | A B   |
| 7 KOGANE                               | 38,75                            | G2       | A B   |
| 3 NORIN 16                             | 39,67                            | G3       | A B   |
| 9 TOWADA                               | 60,90                            | F2       | A B C                                       |
| 10 TAISETSU                            | 68,63                            | F1       | B C   |
| 8 HOKKAIDO                             | 79,15                            | G1       | C   |
| 2 KID                                  | 80,80                            | F2       | C   |
| CVM %                                  | 32,8                             |          |   |

| INNOCULATION WITH MYCELIUM ON MATCHES |                                  |          |   |
|---------------------------------------|----------------------------------|----------|---|
| Genotypes                             | % plants attacked<br>(arc sin %) | Stages * | Newman and Keuls<br>Test for<br>homogeneity |
| 1 NORIN 9                             | 2,66                             | G2       | A   |
| 3 NORIN 16                            | 8,36                             | G2       | A E   |
| 5 GENKAI                              | 9,94                             | G2       | A B   |
| 6 ISUZU                               | 13,04                            | G2       | A B   |
| 7 KOGANE                              | 18,08                            | G2       | A B C                                       |
| 4 MIYUKI                              | 19,69                            | G2       | A B C                                       |
| 8 HOKKAIDO                            | 21,69                            | G1       | B C D                                       |
| 2 KID                                 | 32,25                            | F2       | C D E                                       |
| 9 TOWADA                              | 35,30                            | F2       | D E   |
| 10 TAISETSU                           | 41,09                            | F1       | E   |
| CVM %                                 | 39,2                             |          |   |

\* CETIOM, 1987

TABLE 2 : BEHAVIOUR OF ASIATIC GENOTYPES UNDER ARTIFICIAL INFECTION.

| Genotypes | Growth Stages | Mean necrosis length (cm) | Newman and Keuls test for homogeneity | CMV % |
|-----------|---------------|---------------------------|---------------------------------------|-------|
| NORIN 9   | E             | 3,8                       | A                                     | 17,4  |
| NORIN 9   | G3            | 4,7                       | A                                     |       |
| NORIN 9   | G1            | 5,3                       | A                                     |       |
| JET NEUF  | G3            | 7,6                       | B                                     |       |
| JET NEUF  | G1            | 8,0                       | B                                     |       |
| BIENVENU  | G3            | 8,2                       | B                                     |       |
| BIENVENU  | G1            | 8,3                       | B                                     |       |

TABLE 3 : INFLUENCE OF GROWTH STAGE ON MEAN NECROSES LENGTH ON OILSEED STEMS IN GREENHOUSE.

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RESISTANCE OF SOME CRUCIFERS TO ALTERNARIA  
BRASSICAE (Berk) Sacc.

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INTRODUCTION

Black spot due to Alternaria brassicae infection is frequent on oilseed rape, and under certain conditions can result in significant yield loss. Resistance of several crucifer to a french strain of A. brassicae is described and discussed in this paper. Tests involved study of infection of cotyledonary stages in controlled climatic conditions, and of infection of pods.

MATERIAL and METHODS

Infection under field conditions is variable and dependent on climate, in particular : high temperatures of the order of 22°C, and alternance of wet and dry periods favour proliferation of the parasite. The present study of source of, and variability in resistance to the parasite was therefore experimental.

1. TEST on COTYLEDONARY STAGES

The test was adapted from that describes for Phoma (WILLIAMS, 1985). Seeds were subjected to 48 hrs pregermination at 20°C on moistened filter paper. Ten seeds per genotype, were then planted, two per 8cm diameter pot, in a mixture of 1/3 fresh soil, 1/3 sand and 1/3 peat. Plants were grown and tested in a constant growing chamber at 18°C. A. brassicae spores taken from a sixteen day culture on V8 medium were suspended at 5. 10<sup>6</sup> spores/ml and 10<sup>7</sup> spores/ml in sterile water. Two calibrated 10µl droplets were placed on each cotyledon eight days after establishment. Evaporation was prevented by holding plants for 17hrs in darkness in small covered plastic boxes. The droplets were then remove with filter paper, and box covers removed. Conditions in the room were then 18°C, 80% relative humidity, and 14hrs light. Plants were scored 48 and 96hrs later as follows.

0. No apparent infection.
1. 1 to 2 spots
2. 2 to 7 distinct spots.
3. 8 to 15 distinct spots.
4. Extended spotting, but whole surface of droplet location not completely covered.
5. Whole droplet location completely covered.

Only the highest score for each plant retained for analysis.

## 2. TEST on CUT MAINS

Mainstems from stage G4 plants (first ten pods showing rounding) were collected in the fields, five per genotype, and placed in jars with water. They were then sprayed with a  $1. 10^4$  spore/ml suspension until stems dripped water. Mainstems were then kept in constant temperature at 20°C with relative humidity between 85% (day) and 100% (night). Samples were scored fifteen days later, for 30 randomly chosen pods per mainstems. Scoring was as follows:

0. No apparent infection
1. 1 to 3 visible spots.
3. 4 to 10 visible spots.
5. Up to 20 visible spots or no more than 1/4 pod affected.
7. High spot density on a single valve or no more than 1/2 pod affected.
9. High spot density on two valves or 1/2 pod affected.

## RESULTS.

### 1. TEST at COTYLEDONARY STAGE.

Brassica campestris from various area all showed greater sensitivity than oilseed rape varieties, Bruto, and Jet Neuf, except for Arkus. However, some variability in resistance appeared between turnips (Table 1). All radish varieties (Raphanus sativus) tested were similar seed in sensitivity (Table 2). Brown mustard (Brassica juncea) was more sensitive (Table 2). Significantly higher levels of resistance were found for white mustard genotype (Sinapsis alba) as compared with oilseed (Table 3). Lesions were small, and limited in extent.

### 2. TESTS on MAINSTEMS PODS

Some variability in oilseed resistance was found (Table 4). Jet Neuf had best resistance to inoculation of pods. White mustards had lower attack levels compared to oilseed, for all genotype. Black mustards (Junius spp) had intermediate response, while radish was at least sensitive as oilseed.

| Species               | Genotype | Mean scores<br>1 to 5 | Test for homo-<br>geneity (1) |
|-----------------------|----------|-----------------------|-------------------------------|
| <i>Brassica napus</i> | BRUTOR   | 0.58                  | A                             |
| <i>B. napus</i>       | JET NEUF | 0.83                  | A B                           |
| <i>B. campestris</i>  | ARKUS    | 1.08                  | A B                           |
| <i>B. c.</i>          | CHICON   | 1.83                  | B C                           |
| <i>B. c.</i>          | PREKO    | 1.94                  | B C                           |
| <i>B. c.</i>          | DAISY    | 2.50                  | C D                           |
| <i>B. c.</i>          | IARI     | 2.75                  | C D                           |
| <i>B. c.</i>          | TORIA 6  | 2.83                  | C D                           |
| <i>B. c.</i>          | ZW311    | 3.17                  | C D E                         |
| <i>B. c.</i>          | APPIN    | 3.33                  | D E F                         |
| <i>B. c.</i>          | TORIA 13 | 3.42                  | D E F                         |
| <i>B. c.</i>          | DYS 1    | 3.58                  | D E F                         |
| <i>B. c.</i>          | T25 YSP  | 3.58                  | D E F                         |
| <i>B. c.</i>          | YSP      | 3.58                  | D E F                         |
| <i>B. c.</i>          | D21      | 4.25                  | E F                           |
| <i>B. c.</i>          | R5000    | 4.33                  | E F                           |
| <i>B. c.</i>          | TEXI     | 4.58                  | E F                           |
| <i>B. c.</i>          | T17      | 4.58                  | E F                           |
| <i>B. c.</i>          | T7       | 4.58                  | E F                           |
| <i>B. c.</i>          | T26      | 4.67                  | F                             |

TABLE 1 : Mean infection scores for turnip genotype at cotyledonary stage.

| Species                | Genotype  | Mean scores<br>1 to 9 | Test for homo-<br>geneity (1) |
|------------------------|-----------|-----------------------|-------------------------------|
| <i>Sinapis alba</i>    | EMERGO    | 0.96                  | A                             |
| <i>Brassica napus</i>  | JET NEUF  | 4.71                  | B                             |
| <i>B. n.</i>           | BRUTOR    | 5.54                  | B C                           |
| <i>Raphanus stivus</i> | RESAL     | 5.61                  | B C                           |
| <i>R. s.</i>           | PEGLETTA  | 5.68                  | B C                           |
| <i>R. s.</i>           | NEMEX     | 5.75                  | B C                           |
| <i>R. s.</i>           | REMUS     | 6.29                  | B C                           |
| <i>R. s.</i>           | CLOVIS    | 6.35                  | B C                           |
| <i>R. s.</i>           | SILETTINA | 6.45                  | B C                           |
| <i>R. s.</i>           | IRIS      | 6.54                  | B C                           |
| <i>R. s.</i>           | SILETTA   | 6.75                  | B C                           |
| <i>Brassica juncea</i> | PICRA     | 8.78                  | C                             |

TABLE 2 : Infection indices of radish tested at cotyledonary stage

(1) : Newman & Keuls

| Species                | Genotype | Mean scores<br>0 to 5 | Test for homo-<br>geneity (1) |
|------------------------|----------|-----------------------|-------------------------------|
| <i>Sinapis alba</i>    | 3178     | 0.70                  | A.                            |
| S. a.                  | 1676     | 0.83                  | A.                            |
| S. a.                  | 977      | 0.93                  | A.                            |
| S. a.                  | 1477     | 0.98                  | A B                           |
| S. a.                  | EMERGO   | 1.15                  | A B                           |
| S. a.                  | 1375     | 1.20                  | A B                           |
| S. a.                  | CARLA    | 1.28                  | A B                           |
| S. a.                  | 277      | 1.48                  | A B                           |
| S. a.                  | SIGNAL   | 1.55                  | A B                           |
| S. a.                  | CARINE   | 1.57                  | A B                           |
| S. a.                  | SERVA    | 1.75                  | A B C                         |
| S. a.                  | DIALBA   | 1.78                  | A B C                         |
| S. a.                  | 6718     | 1.85                  | A B C                         |
| S. a.                  | PERRINE  | 2.41                  | B C D                         |
| S. a.                  | MAXI     | 2.90                  | C D E                         |
| <i>Brassica napus</i>  | JET NEUF | 3.10                  | D E                           |
| B. n.                  | BRUTOR   | 3.75                  | E F                           |
| <i>Brassica juncea</i> | PICRA    | 4.50                  | F                             |

TABLE 3 : Mean infection scores for white mustard genotype at cotyledonary stage.

(1) : Newman & Keuls

| Genotype Species | Attack index<br>0 to 9 | Test for homogeneity (1) |
|------------------|------------------------|--------------------------|
| JET NEUF         | 3.08                   | A                        |
| LEMKES           | 3.97                   | A B                      |
| BIENVENU         | 4.16                   | A B                      |
| MATADOR          | 4.76                   | A B                      |
| AZTEC            | 5.00                   | A B                      |
| VICTOR           | 5.31                   | A B C                    |
| RAPORA           | 5.48                   | A B C                    |
| DAMOR            | 5.65                   | A B C                    |
| JUPITER          | 6.22                   | B C                      |
| SYNRA            | 6.26                   | B C                      |
| BELINDA          | 6.71                   | B C                      |
| KORINA           | 7.84                   | C                        |

TABLE 4 : Pod attack index (0-9) for alseed rape genotype.

| Genotype Species | Attack index<br>0 to 9 | Test for homogeneity (1) |
|------------------|------------------------|--------------------------|
| 2375 S.a.        | 0.12                   | A                        |
| 6718 S.a.        | 0.18                   | A                        |
| MAXI S.a.        | 0.18                   | A                        |
| SERVAA S.a.      | 0.27                   | A                        |
| 1676 S.a.        | 0.32                   | A B                      |
| CARINE S.a.      | 0.35                   | A B                      |
| 3277 S.a.        | 0.37                   | A B                      |
| 1477 S.a.        | 0.38                   | A B                      |
| 3178 S.a.        | 0.40                   | A B                      |
| 1375 S.a.        | 0.43                   | A B                      |
| SIGNAL S.a.      | 0.72                   | A B                      |
| 277 S.a.         | 0.73                   | A B                      |
| 977 S.a.         | 0.88                   | A B                      |
| CARLA S.a.       | 0.93                   | A B                      |
| EMERGO S.a.      | 1.17                   | A B                      |
| DIALBA S.a.      | 1.17                   | A B                      |
| 475 S.a.         | 1.23                   | A B                      |
| PERRINE S.a.     | 1.49                   | A B                      |
| JUNIUS B.ni      | 2.67                   | B                        |
| CRESOR 1 B.n.    | 4.90                   | C                        |
| CRESOR 2 B.n.    | 5.37                   | C                        |
| BRUTOR 1 B.n.    | 5.47                   | C                        |
| BRUTOR 2 B.n.    | 5.50                   | C D                      |
| REMUS R.s        | 5.67                   | C D                      |
| PEGLETTA R.s     | 5.80                   | C D                      |
| IRIS R.s         | 6.70                   | C D                      |
| RESAL R.s        | 6.73                   | D                        |
| CLOVIS R.s       | 7.53                   | D                        |

TABLE 5 : Pod attack indice for crucifer species after artificial inoculation of mainstem.

(1) : Newman & Keuls

## DISCUSSION

Test result reveal a source of resistance in white mustard and high sensitivity in turnip rape which confirms results obtained by HUSAIN and THAKUR, 1963. Result of tests at cotyledonary stage correlate with those for pod tests, and demonstrate the interest of early testing. Transfer of white mustard resistance to oilseed rape is currently being attempted, using protoplasmic fusion hybrids (PRIMARD and al., 1987).

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University of Wisconsin, Madison - USA



## VERTICILLIUM WILT ON RAPE SEED

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### INTRODUCTION

*Verticillium dahliae* is the most serious disease on winter rape in Sweden. Especially in the southwest part where rape has been grown since the last war. The disease, which is a crop rotation problem, are now showing up in different parts of Europe. This indicate that the fungus could be of significant importance outside Sweden too.

### SYMPTOMS

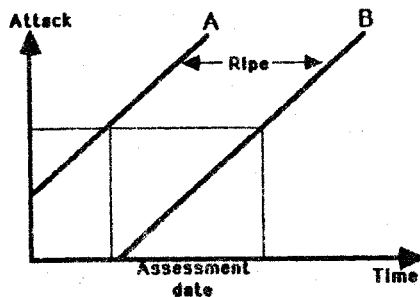
The first symptoms are light green color of the stem. Often half of the stem is attacked.

Some days later the color turns to brown.

Later on the stem will be whitish with huge amounts of black micro-sclerotia.

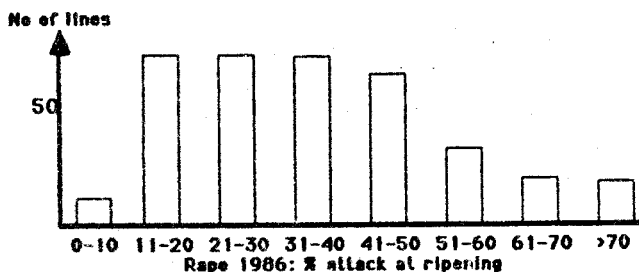
Stem will broke.

### RIPENING AND DISEASE ATTACK



Variety A and B are equally susceptible, but B is later in ripening. The late variety would then look more resistant if the assessment is done on the same day. To be comparable, the assessment should be performed in equal stage of ripening. At Svalöf all breeding material are assessed at the same stage of maturity, at ripening stage  $\pm 1$  day.

### RESISTANCE



In more resistant lines the symptoms are developed later and the plants will be healthy or just slightly attacked at harvest time.

No variety with complete resistance on heavily infected soils has yet been found. But there is a wide variation within the breeding material when compared at ripening.

SUSCEPTIBILITY OF SELECTED CULTIVARS AND LINES OF WINTER  
RAPESEED TO DOWNY MILDEW

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SUMMARY. During the years 1983-85 there were studied the susceptibility of various cultivars and lines of winter rapeseed to downy mildew under field and greenhouse conditions. None of these cultivars and lines appear to have any degree of resistance to downy mildew. However they do exhibit significant difference in their degree of susceptibility. Under field conditions, the most infected were Górczański, Kocenas, Linakosta, whereas the least infected were BOH 183, BOH 284, Jupiter. Under greenhouse conditions, the most susceptible were Górczański, Lingot, Jupiter, while the least susceptible were BOH 183, BOH 283, Jet Neuf.

INTRODUCTION

An increase in area of winter rape cultivation and the change from high to low erucic cultivars and glucosinolates have made it necessary to pay closer attention to the diseases attacking the plant. The most frequent and commonest rape disease in Poland is downy mildew (*Peronospora parasitica* Pers. ex. Fr.). In the world literature, we can find fairly many papers dealing with the effect of the incidence of this pathogen on brassicas, however are relatively few papers discussing its on rape. Besides, some authors have been of the opinion that downy mildew is the disease which has a non-significant effect on yield, while others stress the necessity of making more precise research (Downy and Dolton 1961, Rawlinson and Muthyalu 1979, Brokenshire and Prasanna 1984, Hornig 1985).

In Poland, the pathogen occurs commonly but there has been elaborated no detailed paper dealing with its presence on rape. According to the author's earlier observations, agrotechnical methods have a small effect on its incidence and severity. The fungicides containing metalaxyl reduce, relatively easily, the development of the pathogen but these fungicides hardly reduce the presence of other pathogens such as *Botrytis*, *Alternaria*, *Sclerotinia*, *Phoma* which can occur with a high severity as well.

Thus, the author has decided to make an analysis of the infection of various cultivars and lines of rape to state whether there are considerable differences among them and whether this fact might be helpful and useful for breeding new cultivars of a higher resistance to *Peronospora parasitica*.

#### MATERIAL AND METHODS

Over the period 1983-86, there were made observations of the infection of various cultivars and lines of winter rape infected in a natural way under field conditions and under greenhouse conditions with artificial infection. The field observations were made at the Regional Research Station for Plants Evaluation Chrzastowo, near Bydgoszcz, in four replications on plots of 3 x 10m. On each plot there were estimated 50 plants with an analysis of all mature leaves. The percent of infected leaves and infection degree were studied. The infection degree was determined using the scale 0-5 where 0 denoted healthy, 5 - the highest degree /Sadowski 1987/.

Greenhouse experiments were performed at the Technical Agricultural University in Bydgoszcz. In spring, there were sown surface sterilized rape seeds in the pot. The experiment was performed in four replications, each pot contained 10 plants. The cotyledones and leaves were rubbed slightly to remove some of the wax from their surface then inoculated by atomising with a suspension containing about  $10^5$  spores/ml of *P. parasitica*. The suspension was prepared according to the method described by Greenhalgh and Dickinson /1975/. During the infection the seedlings

had cotyledons and two leaves.

After spraying the seedlings were kept in plastic chambers, in darkness for 16 h, at a temperature ca 16°C and humidity ca 100%. Then they were kept in the greenhouse for 7 days at 16-21°C. After that period the seedlings were again under the conditions of very high humidity for 16 h and than estimated. The infection rate of cotyledons and percent of infected leaves was estimated. For determining the infection rate of cotyledons the five-degree scale was used (Natti et al. /1987/).

#### RESULTS AND DISCUSSION

Although there were noticed a great number of infected cultivars and lines of rape, the list of results included only those which were observed for, at least, two years.

Table 1

The infection of cultivars and lines under examination over the period 1983-84 and 1984-85

| 1983-84       |      |      | 1984-85       |      |      |
|---------------|------|------|---------------|------|------|
| Cultivar-line | % L  | DI   | Cultivar-line | % L  | DI   |
| Jupiter       | 21   | 0.3  | Jupiter       | 22   | 0.6  |
| BKH           | 26   | 0.5  | BOH 293       | 20   | 0.8  |
| MAH 181       | 26   | 0.5  | BOH 183       | 23   | 0.8  |
| Lirabu        | 28   | 0.6  | MAH 181       | 26   | 1.0  |
| Skrzeszowicki | 30   | 0.6  | Jet Neuf      | 37   | 1.0  |
| PUR 182       | 30   | 0.6  | Coral         | 28   | 1.2  |
| Jet Neuf      | 34   | 0.6  | Korins        | 30   | 1.2  |
| Herkules      | 34   | 0.6  | POB 182       | 32   | 1.2  |
| Librador      | 35   | 0.7  | Skrzeszowicki | 33   | 1.2  |
| POB 182       | 35   | 0.7  | Librador      | 37   | 1.2  |
| MAH 281       | 43   | 0.7  | Belinda       | 29   | 1.3  |
| Quinta        | 36   | 0.8  | Beryl         | 29   | 1.3  |
| WV 343        | 47   | 0.8  | Liglander     | 37   | 1.5  |
| Perle         | 47   | 0.8  | Górczański    | 40   | 1.7  |
| Górczański    | 49   | 1.0  | Mariusz       | 41   | 1.7  |
| LSD /P=0.05/  | 19.2 | 0.28 |               | 16.5 | 0.32 |

% L - % of leaves infected; DI - disease index, see text

From among 15 cultivars and lines which were studied in the years 1983-84 the cultivar Jupiter was infected the least, whereas Górczański - the most. In the years 1984-85 the least infected was again Jupiter and lines BOH 283 and BOH 183, the most - Marinus and Górczański /Table 1/.

In the years 1985-86 the cultivars Marinus and Lirakotta were infected the most, whereas Jupiter, Jet Neuf and lines BOH 283, BOH 183 - the least /Table 2/.

Table 2

The infection of the cultivars and lines under examination over the period 1985-86

| Cultivar-line  | % L | DI  | Cultivar-line | % L  | DI   |
|--|-----|-----|---------------|------|------|
| Jupiter  | 20  | 0.6 | Tandem        | 23   | 0.9  |
| Jet Neuf   | 25  | 0.7 | Liropa        | 24   | 0.9  |
| BOH 283  | 20  | 0.7 | Korina        | 27   | 0.9  |
| BOH 183  | 21  | 0.7 | Lindora       | 22   | 1.0  |
| BOH 484  | 19  | 0.9 | Ridana        | 24   | 1.0  |
| Gundula  | 21  | 0.9 | Beryl         | 27   | 1.0  |
| Doral  | 21  | 0.9 | WW 956        | 27   | 1.1  |
| Belinda  | 22  | 0.9 | Marinus       | 25   | 1.2  |
| Vamara   | 22  | 0.9 | Lirakotta     | 29   | 1.2  |
| LSD /P=0.05/   |     |     |               | 12.5 | 0.19 |
| % L - % of leaves infected; DI - disease index, see text |     |     |               |      |      |

In greenhouse experiments the least infected were BOH 183, Jet Neuf, BOH 283, Start and PUR 182, the most - Górczański, Lingot, Jupiter, Quinta, Janpol, Marinus, Perle, Liglandor, Skrzyszowicki. There was not confirmed a low susceptibility of the cultivar Marinus under the field conditions and natural infection. Under greenhouse conditions Marinus belonged to the group of plants infected the most /Table 3/.

On the basis of the results obtained we may state that under field conditions, differentiation of the infection of the lines and cultivars was not considerable. Significant differences were obtained under greenhouse conditions.

with artificial infection.

Table 3

The infection of the cultivars and lines under greenhouse conditions, May 1985

| Cultivar-line | % L | CI  | Cultivar-line | % L | CI  |
|---------------|-----|-----|---------------|-----|-----|
| BOH 183       | 0   | 1.5 | Ligora        | 11  | 3.5 |
| Jet Neuf      | 0   | 1.6 | MAH 181       | 3   | 3.9 |
| BOH 283       | 2   | 1.9 | Korina        | 0   | 4.0 |
| Start         | 2   | 2.1 | BKH 180       | 0   | 4.0 |
| PUR 182       | 0   | 2.2 | Skrzeszowicki | 9   | 4.1 |
| Belinda       | 0   | 3.1 | Liglandor     | 0   | 4.2 |
| WW 843        | 0   | 3.1 | Perle         | 15  | 4.2 |
| Lirabu        | 0   | 3.3 | Marinus       | 3   | 4.3 |
| Herkules      | 0   | 3.3 | Janpol        | 20  | 4.5 |
| Doral         | 5   | 3.3 | Quinta        | 7   | 4.6 |
| POB 182       | 5   | 3.3 | Jupiter       | 18  | 4.7 |
| Librador      | 16  | 3.4 | Lingot        | 33  | 4.8 |
| Tomek         | 0   | 3.5 | Górczański    | 35  | 4.8 |
| LSD /P=0.05/  |     |     |               | 1.1 |     |

% L - % of leaves infected; CI - index of cotyledons infection, see text

Distinct differences in the infection of 14 cultivars under field conditions during five years were obtained by Dixon /1975/ and he suggests that in the rape cultivars of differentiated susceptibility there are sources of resistance to *Peronospora parasitica*. A differentiation of infection was also the case in Rawlinson and Muthyalu's research /1979/. Natti /1958/ and Natti et al. /1967/ report a considerable differentiation of broccoli cultivars susceptibility. However D'Ercole /1972/ during his observation did not notice any clear differences in infection of 30 cultivars of cauliflower under field conditions.

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ESTIMATION OF INFECTION DEGREE OF WINTER RAPE CULTIVARS  
BY *Sclerotinia sclerotiorum* UNDER CONDITIONS OF  
EXPERIMENTAL INFECTION IN FIELD TRIALS

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INTRODUCTION

Several years ago we initiated a complex programme of experiments on fungal diseases in oilseed winter rape. That economic problem has not been properly recognized in Poland so far /Frencel 1983; Frencel et al. 1985/.

This paper is a continuation of the programme in the previously established hierarchy of the importance of problems. The purpose of our studies was to estimate differences in the susceptibility and resistance of winter rape cultivars to *Sclerotinia* stem rot /*Sclerotinia sclerotiorum* Lib. de Bary/ under field conditions of Poland. It was intended through these experiments to work out reliable tests for resistance breeding.

MATERIALS AND METHODS

**P l a n t m a t e r i a l s .** The experiments were carried out on twelve winter rape cultivars: Beryl, Brink, Doral, Górozański, Jet Neuf, Jupiter, Korina, Primor, Rubin, Skrzyszowicki, Tamara, Tandem. Seeds were sown on August, 30-31 on 2x5m experimental plots, at the rate of 6kg/ha.

**Inoculum preparation.** *Sclerotinia sclerotiorum* was cultured for 3 weeks at 25°C on a stable medium consisting of: sand-gravel /260g/: wheat rough /100g/: water /112ml/.



Prior to the inoculation the medium overgrown by mycelium with sclerotia was crumbled.

**I n o c u l a t i o n .** During the spring season 15 plants from each row on the plots were inoculated in 3 replications /= 45 inoculated plants from each cultivar/, dispersing 100ml of granulated inoculum around the stem base of the plants. The term of inoculation fell on flowering, usually in the third decade of May. The lower part of inoculated plants were shaded with aluminium foil stripes and occasionally watered to keep an appropriate humidity for the fungus stabilization on the plants. There were also 3 replications of 15 control /with no inoculation/ plants included.

**S u s c e p t i b i l i t y a n d r e s i s t a n c e e s t i m a t i o n .**

Disease symptoms on the stems were evaluated before harvesting in the scale from 0 to 5 /0=normal plants, 1 = spots: periphery of the stems is infected in 1 to 5%, 2 = up to 25%, 3 = up to 50%, 4 = up to 75%, 5 = up to 100%/.

The seed yield of inoculated and control replications was weighed for each cultivar.

#### RESULTS AND DISCUSSION

The effectiveness of the inoculation method under assigned experimental conditions was high. The initiation, stabilization and efficiency of infection were achieved. At the same time disease symptoms determined as the mean infection degree and reduction of the seed yield displayed differences in the resistance of winter rape cultivars /table 1 and 3/. However, it seems that a detailed interpretation of results in each particular case is impossible, because differences between the cultivars were not sufficiently recurrent. Correlation in the infection degree between the cultivars in the successive years was not achieved /table 2/.

There were also differences in the infection degree

between the years. It is presumed that the weather conditions were the most responsible for infection effectiveness of the field trials, since the inoculation method was the same every year and the localization of the experiments does not implicate significant differences of soil environmental conditions.

According to the analysis of variance the infection degree between winter rape cultivars was statistically significant in 1984 and 1986. In 1985 no statistical differences were observed between cultivars, presumably because of a generally higher level of infection owing to the weather conditions more suitable for the fungus colonization.

A reduction in the seed yield was an evident effect of inoculation for each cultivar /table 3/. That parameter, like the infection degree, was different depending on the cultivar and the year of experiment. However, statistically significant differences were found only in 1985, when the general infection level was higher.

The intercultural differences between the successive years of experiment were more correlated for the seed yield /table 4/ than for the infection degree /table 2/ as the Sclerotinia response parameter.

The cultivar Jet Neuf, which now has become popular in Poland, appeared more susceptible to Sclerotinia infection than some other cultivars investigated. This is similar to German results /Kruger 1983/, but on the contrary to French authors /Renard and Brun 1982/.

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Table 1: Infection degree /in %/ $\bar{x}$  of winter rape cultivars experimentally inoculated with *Sclerotinia sclerotiorum* under field conditions. A comparison of three vegetative seasons

| Cultivar      | vegetative season |         |         |
|---------------|-------------------|---------|---------|
|               | 1983/84           | 1984/85 | 1985/86 |
| Górczański    | 30,08             | 70,33   | 28,00   |
| Jet Neuf      | 55,83             | 85,27   | 35,07   |
| Doral         | 33,75             | 83,60   | 27,93   |
| Jupiter       | 28,00             | 76,67   | 51,13   |
| Skrzeszowicki | 45,42             | 54,53   | 25,40   |
| Brink         | 44,17             | 63,40   | -       |
| Beryl         | 34,58             | 68,47   | -       |
| Primor        | 60,42             | -       | -       |
| Korina        | -                 | -       | 31,20   |
| Tamara        | -                 | -       | 33,40   |
| Rubin         | -                 | -       | 32,13   |
| Tandem        | -                 | -       | 33,60   |

Mean /in angular Bliss degree/

32,64                      63,39                      36,38

LSD for  $\alpha = 0,05$  - 9,644

LSD for  $\alpha = 0,01$  - 14,609

$$\bar{x} / \text{Infection degree in \%} = \frac{\sum /n \cdot v/ \cdot 100}{V \cdot N}$$

where:

n = number of plants in particular infection degree /0-5/

N = the whole number of plants studied

v = the particular infection degree

V = the highest infection degree

Table 2: Correlation coefficients of the infection degree of three vegetative seasons

|         | 1983/84 | 1984/85 | 1985/86 |
|---------|---------|---------|---------|
| 1983/84 | -       | 0,017   | 0,328   |
| 1984/85 | -       | -       | 0,305   |

Table 3: Seed yield of winter rape cultivars experimentally inoculated with *Sclerotinia sclerotiorum* under field conditions /in % of the control/

| Cultivar      | vegetative season |         |         |
|---------------|-------------------|---------|---------|
|               | 1983/84           | 1984/85 | 1985/86 |
| Górczański    | 82,90             | 62,93   | 67,83   |
| Jet Neuf      | 63,83             | 51,92   | 61,86   |
| Doral         | 72,81             | 46,59   | 52,01   |
| Jupiter       | 79,16             | 66,33   | 65,72   |
| Skrzeszowicki | 79,94             | 79,69   | 85,63   |
| Brink         | 62,55             | 83,01   | -       |
| Beryl         | 80,35             | 66,55   | -       |
| Primor        | 55,65             | -       | -       |
| Korina        | -                 | -       | 86,95   |
| Tamara        | -                 | -       | 63,08   |

Mean /in angular Bliss degree/

60,99                      49,54                      52,13

LSD for  $\alpha = 0,05$  - 5,078

LSD for  $\alpha = 0,01$  - 7,692

Control = 100 % seed yield from plants without inoculation

Table 4: Correlation coefficients of seed yield of the three vegetation seasons

|         | 1983/84 | 1984/85 | 1985/86             |
|---------|---------|---------|---------------------|
| 1983/84 | -       | 0,131   | 0,623               |
| 1984/85 | -       | -       | 0,960 <sup>xx</sup> |

<sup>xx</sup> P = 1 %

LEAF SURFACE CONSTITUENTS OF BRASSICA SPECIES IN RELATION  
TO ALTERNARIA LEAF BLIGHT /ALTERNARIA BRASSICAE /BERK/  
SACC. AND A.BRASSICICOLA /SCHW/ WILTS.7

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Abstract

The leaf surface constituents viz. wax, total phenols, soluble nitrogen, total soluble sugars and reducing sugars of Brassica species in relation to Alternaria leaf blight /Alternaria brassicae and A.Brassicicola/ were determined. The species Brassica campestris CV. BSH-1 and YSPb-24, Brassica juncea CV. RH-30 were susceptible while Brassica napus CV. Tower and HNS-3, Brassica carinata CV. HC-2 and Brassica alba local cultivar were resistant to the disease. The leaf samples for analysis were collected at 30, 50, 70 and 90 days after sowing. Wax was obtained by washing the surface of the leaf with chloroform. Total phenols, soluble nitrogen, total soluble sugars and reducing sugars were obtained by washing the leaf with 75 per cent alcohol. Wax and soluble nitrogen level increased continuously except at the last stage under study in all the species. Total phenols after the initial increase were observed to drop at later stages. Total soluble and reducing sugars, however, increased continuously with the age of a plant. The concentrations of the wax and phenolic were markedly higher in resistant species as compared to susceptible ones at all the stages of plant growth. Total soluble sugars, reducing sugars and soluble nitrogen levels were, however, lower in resistant species. The presence of higher amount of wax and phenolics on the leaf surface of disease resistant species projects the possibility in claiming resistance to Alternaria leaf blight in Brassica.

### Introduction

Wax present on the leaf surface plays an important role in the protection of plants against environmental conditions /Kolattukudy, 1970; Bengtson et.al., 1978/ pests /Allebone et.al., 1971/ and diseases /Blakeman and Sztejnberg, 1973; Jeffree, 1974/. There are also reports on the quantitative differences in wax content of disease resistant and susceptible genotypes of Brassica campestris /Sharma, 1934/ and Arachis hypogaea /Gupta et.al., 1985/. The total amounts of phenolics, soluble sugars and nitrogen of the plant tissue have also been related to disease resistance in different crops. /Farkas and Kirlyay, 1962; Brahamchari and Kolte, 1983; Gupta et.al., 1984/, but the reports regarding the presence of these constituents on the leaf surface are lacking. Present investigation was, therefore, undertaken to study the quantitative differences in wax, total phenols, total soluble sugars, reducing sugars and soluble nitrogen on the leaf surface of brassica species, susceptible and tolerant to *Alternaria* leaf blight during plant development.

### Materials and methods

Five brassica species, namely Brassica campestris CV. BSH-1 and YSPb-24, Brassica juncea CV. RH-30 susceptible while Brassica napus CV. Tower and HNS-3, Brassica carinata CV. HC-2 and Brassica alba local cultivar resistant to *Alternaria* leaf blight /Alternaria brassicae and A. brassicicola/ were grown in the Experimental Farm of Haryana Agricultural University, Hisar, India. The leaves of the plant of each cultivar were collected at 30, 50, 70 and 90 days after sowing /DAS/. The wax from the leaf surface was obtained by washing the leaves with chloroform and was estimated according to the method of Ebercon et.al., /1970/. Total phenols, total soluble sugars, reducing sugars and soluble nitrogen from leaf surface were obtained by washing the leaves with 75 per cent alcohol. From the washing, total phenols were determined by the method of Swain and Hills /1959/, total soluble and reducing sugars as per

method described by Hulme and Narain /1931/ and soluble nitrogen by conventional micro-Kjeldahl's method. The concentration of each of the constituent was expressed as  $\mu\text{g}/\text{cm}^2$  surface of fresh leaf.

### Results and discussion

The wax content was maximum /Table 1/ or the constituents estimated on the surface of leaf during plant development. It was observed to increase rapidly from 30 DAS to 50 DAS and then at a slower rate up to 70 DAS, thereafter it declined marginally irrespective of the cultivars. However, the content was appreciably higher in disease resistant species as compared to susceptible ones at all the stages of plant growth. These results indicate that the presence of higher amounts of wax on the leaf of disease resistant species may prevent the penetration and establishment of the pathogen. Blakeman and Szejnberg /1973/ reported about 40-70 per cent reduction in germination of Botrytis cineria conidia when grown on varied concentrations of wax removed from surface of the leaves of beet root. Higher amount of leaf surface wax in resistant cultivars of Brassica campestris was also reported by Sharma /1984/.

The leaf of total phenols after an initial increase declined till the last date of sampling in all the species. The phenolic compounds play an important role in the defence of the plant against diseases /Farkas and Kirly, 1962/. Their concentration also enhances in the plant tissue which is attacked by the pathogen /Nyerges et.al., 1975/ and their higher amount was found in the leaves of disease resistant genotypes of Brassica juncea /Gupta et. al., 1984/. In the present study, also total phenols on the leaf surface were markedly higher in disease resistant species than the susceptible ones during plant growth. This might be helpful in protecting the plant from primary infection by the pathogen.

The concentration of total as well as reducing sugars on the leaf surface was observed to increase consistently



with the age of the plant in all the species. Contrary to wax and phenols, both types of sugars were considerably higher in susceptible species than the resistant ones. Since, diseases have been classified into high and low sugar diseases /Horsfall and Dimond, 1957/, therefore the high amount of sugars in susceptible species indicate that *Alternaria* leaf blight pathogen might require high amount of sugars for its pathogenesis. As regards soluble nitrogen it was found to increase from 30 DAS to 70 DAS after which it dropped a little in all the species. In comparison to susceptible species, soluble nitrogen was less in resistant species at all the stages of plant growth. Low amount of nitrogen in a plant decreases the severity of the disease /Naidu et.al., 1979/. Similarly, low level of soluble nitrogen on leaf surface of resistant species of brassica might restrict the incidence of *Alternaria* leaf blight disease.

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Table 1. Changes in biochemical constituents on the surface of the leaf of brassica species in relation to *Alternaria* leaf blight. ( $\mu\text{g}/\text{cm}^2$  fresh leaf).

| Days after sowing(DAS) | Brassica species            |       |                  |                 |                    |                |       |
|------------------------|-----------------------------|-------|------------------|-----------------|--------------------|----------------|-------|
|                        | Susceptible                 |       |                  | Resistant       |                    |                |       |
|                        | <i>B. campestris</i>        |       | <i>B. juncea</i> | <i>B. napus</i> | <i>B. carinata</i> | <i>B. alba</i> |       |
| BSH-1                  | YSPB-24                     | RH-30 | Lower HNS-3      | HO-2            | Local cultivar     |                |       |
|                        | <u>Wax</u>                  |       |                  |                 |                    |                |       |
| 30                     | 8.10                        | 7.91  | 8.35             | 12.25           | 16.63              | 12.35          | 10.16 |
| 50                     | 16.86                       | 17.01 | 16.80            | 26.04           | 31.10              | 25.06          | 22.07 |
| 70                     | 21.12                       | 19.95 | 21.66            | 31.93           | 34.33              | 31.10          | 27.66 |
| 90                     | 17.08                       | 17.68 | 18.36            | 27.91           | 29.75              | 27.63          | 24.95 |
|                        | <u>Phenols</u>              |       |                  |                 |                    |                |       |
| 30                     | 2.08                        | 1.71  | 1.56             | 2.39            | 2.48               | 2.48           | 2.81  |
| 50                     | 2.85                        | 2.30  | 2.03             | 3.25            | 3.36               | 3.18           | 3.96  |
| 70                     | 2.45                        | 1.98  | 1.78             | 2.88            | 2.85               | 2.78           | 3.25  |
| 90                     | 1.93                        | 1.43  | 1.25             | 2.40            | 2.38               | 2.51           | 2.61  |
|                        | <u>Soluble Nitrogen</u>     |       |                  |                 |                    |                |       |
| 30                     | 5.41                        | 5.55  | 6.13             | 4.83            | 3.81               | 4.66           | 5.16  |
| 50                     | 10.61                       | 10.51 | 11.38            | 8.56            | 8.11               | 8.75           | 9.12  |
| 70                     | 12.10                       | 12.53 | 12.33            | 9.68            | 9.55               | 10.21          | 10.33 |
| 90                     | 9.26                        | 9.71  | 10.26            | 7.81            | 8.40               | 8.78           | 8.73  |
|                        | <u>Reducing sugars</u>      |       |                  |                 |                    |                |       |
| 30                     | 6.16                        | 5.53  | 7.51             | 4.18            | 2.11               | 4.75           | 4.95  |
| 50                     | 9.78                        | 9.26  | 8.05             | 7.45            | 6.11               | 7.61           | 8.18  |
| 70                     | 10.95                       | 10.45 | 13.51            | 8.81            | 7.51               | 8.95           | 9.38  |
| 90                     | 11.81                       | 11.58 | 14.26            | 9.26            | 8.45               | 9.51           | 10.06 |
|                        | <u>Total Soluble sugars</u> |       |                  |                 |                    |                |       |
| 30                     | 7.61                        | 7.08  | 8.90             | 5.26            | 4.48               | 6.35           | 6.06  |
| 50                     | 11.38                       | 11.18 | 15.93            | 8.01            | 7.83               | 8.93           | 9.16  |
| 70                     | 12.88                       | 12.41 | 16.95            | 9.05            | 9.01               | 9.81           | 10.75 |
| 90                     | 13.43                       | 13.25 | 17.11            | 9.85            | 9.60               | 10.31          | 11.21 |

PATHOGENS OF THE SEEDLING BLIGHT  
OF CANOLA IN ALBERTA

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#### INTRODUCTION

Seedling blight of canola (*Brassica campestris* and *B. napus*), which is a pre- and post-emergence damping-off disease, has been a serious problem in Alberta for the past many years. While precise loss estimates through this disease have not been made, the disease can destroy up to 100 per cent of a canola crop or it could partially thin it out. The losses may result from mortality of the seedlings and through reduced vigor of the surviving plants. Many farmers over-seed to compensate for the effects of seedling blight.

While several reports on this disease have been published from Saskatchewan, little information is available from Alberta (Acharya *et al.*, 1984; Kaminski and Verma, 1985; Yitbarek *et al.*, 1987). This paper reports on the isolation and identification of the causal agents of the seedling blight of canola in Alberta. A few brief reports on this work have been published (Calman and Tewari, 1984, 1987; Calman *et al.*, 1986; Furuya and Tewari, 1985).

#### MATERIALS AND METHODS

During 1984, canola seedlings were collected from a total of 73 fields located in northern, central and southern Alberta. In each field, collections were made from 20 equidistant locations at 1 meter intervals along an inverted V-shaped transect. To reduce boundary effects, the initial and the final sampling sites were no closer than 10 meters from the field boundary. From each field, 40-50 canola seedlings showing the blight symptoms were collected. The seedlings collected belonged to the growth stage 1 (Harper and Berkenkamp, 1975) and up to the formation of the first set of true leaves. Using standard methods, the pathogens were isolated on broad-spectrum and selective media for isolation of fungi.

Pathogenicity experiments were conducted with many isolates of the presumptive pathogens. The methodology (including inoculum production, determination of inoculum density, etc.) was standardized using B. campestris cv. Candle. Finally, the pathogenicity experiments were conducted in growth cabinets using B. campestris cvs. Candle and Tobin and B. napus cvs. Altex and Westar.

For anastomosis group (AG) typing of Rhizoctonia solani isolates, about 3 mm<sup>2</sup> of the colony was cut from the margin of an actively growing colony and placed on 2% water agar plate opposite a similar inoculation of a tester AG strain. Most isolates were purified by single hyphal isolation before testing. The tester AG strains (AG1, AG2-1, AG2-2, AG3, AG4 and AG5) were kindly supplied by Dr. N.A. Anderson, Dept. of Plant Pathology, University of Minnesota. Each test was run in duplicate and rated for fusion and killing reactions (Anderson, 1982).

#### RESULTS AND DISCUSSION

A number of fields surveyed showed patchy distribution of canola seedlings (Fig. 1). The disease symptoms included small to up to a few cms long brownish to blackish lesions and constrictions on the hypocotyl (Fig. 2), tapering of the hypocotyl and evidence of lesion formation and rotting of the roots.

Of the 2,299 cultures isolated from the field collected diseased seedlings, 777 (33.8%), 404 (17.6%) and 134 (5.8%) belonged to the genera Rhizoctonia, Fusarium and Pythium, respectively (Table 1). The remaining cultures (984 or 43.1% of the total) have been grouped as 'Others' in Table 1. These belong to many diverse groups of fungi. Some of these are deemed to be contaminants during isolation while others have been shown to be mycoparasites of the three pathogen genera (Tewari and Furuya, unpublished data).

Pathogenicity testing with most of the presumptive pathogens has satisfied Koch's postulates. However, the degrees of virulence varied considerably among the isolates. Highly virulent strains were present in all the three genera tested. The pathogenic species, so far identified, include Rhizoctonia solani, Fusarium avenaceum, E. acuminatum, Pythium paroecandrum, P. sylvaticum, and P. sp. Group G. Of the isolates of R. solani tested for AG affiliations, 183 (95.8%) belonged to AG2-1, four (2%) belonged to AG4 and affinities of five (2.6%) isolates are still inconclusive (Table 2).

Table 1. Number of Various Fungi Isolated from Diseased Canola Seedlings Collected from Alberta during 1984

| Census Division | Number of Fields Sampled | Total Number of Seedlings Plated | Total Number of Cultures Isolated | Numbers of Cultures of Fungi Isolated |                 |                |            |  |
|-----------------|--------------------------|----------------------------------|-----------------------------------|---------------------------------------|-----------------|----------------|------------|--|
|                 |                          |                                  |                                   | <u>Rhizoctonia</u>                    | <u>Fusarium</u> | <u>Pythium</u> | Others     |  |
| 2               | 7                        | 276                              | 135                               | 35                                    | 14              | 6              | 80         |  |
| 5               | 9                        | 320                              | 313                               | 142                                   | 4               | 17             | 110        |  |
| 6               | 2                        | 46                               | 13                                | 5                                     | 2               | 0              | 6          |  |
| 8               | 2                        | 76                               | 56                                | 20                                    | 2               | 1              | 33         |  |
| 10              | 12                       | 342                              | 218                               | 88                                    | 26              | 22             | 82         |  |
| 11              | 18                       | 681                              | 425                               | 166                                   | 42              | 30             | 187        |  |
| 13              | 1                        | 28                               | 11                                | 7                                     | 1               | 2              | 1          |  |
| 15              | 22                       | 880                              | 1128                              | 314                                   | 273             | 56             | 485        |  |
| <b>Total</b>    | <b>73</b>                | <b>2649</b>                      | <b>2299</b>                       | <b>777</b>                            | <b>404</b>      | <b>134</b>     | <b>984</b> |  |

Table 2. Anastomosis group typing of *Rhizoctonia solani* isolates from canola seedlings in Alberta.

| No. of fields from which <i>R. solani</i> was isolated/no. of fields sampled | No. of <i>R. solani</i> isolates obtained | AG affiliations of the isolates |       |      |              |
|--|---|---------------------------------|-------|------|--------------|
|  |   | No. of isolates tested          | AG2-1 | AG-4 | Inconclusive |
| 22/22 <sup>a</sup>   | 272                                       | 72                              | 69    | 1    | 2            |
| 26/31 <sup>b</sup>   | 270                                       | 64                              | 60    | 3    | 1            |
| 15/18 <sup>c</sup>   | 167                                       | 56                              | 54    | 0    | 2            |
| Total 63/71  | 709                                       | 192                             | 183   | 4    | 5            |

Fields located in northern (a), central (b) and southern (c) Alberta.

The results presented here give information on the pathogens that become associated with canola seedlings after they have emerged from the ground, as seedlings mostly in the post-emergence phase were sampled from the field during this study. There is evidence that *Pythium* spp. may be preferentially associated with the pre-emergence phase of the disease (Calman and Tewari, 1987).

This study has indicated that the seedling blight of canola in Alberta is caused by multiple pathogens and that control measures, to be adequate, must address to all these pathogens belonging to diverse groups of fungi. The preponderance of AG2-1 isolates of *R. solani* in Alberta may explain the high incidence of the seedling blight of canola in this province.

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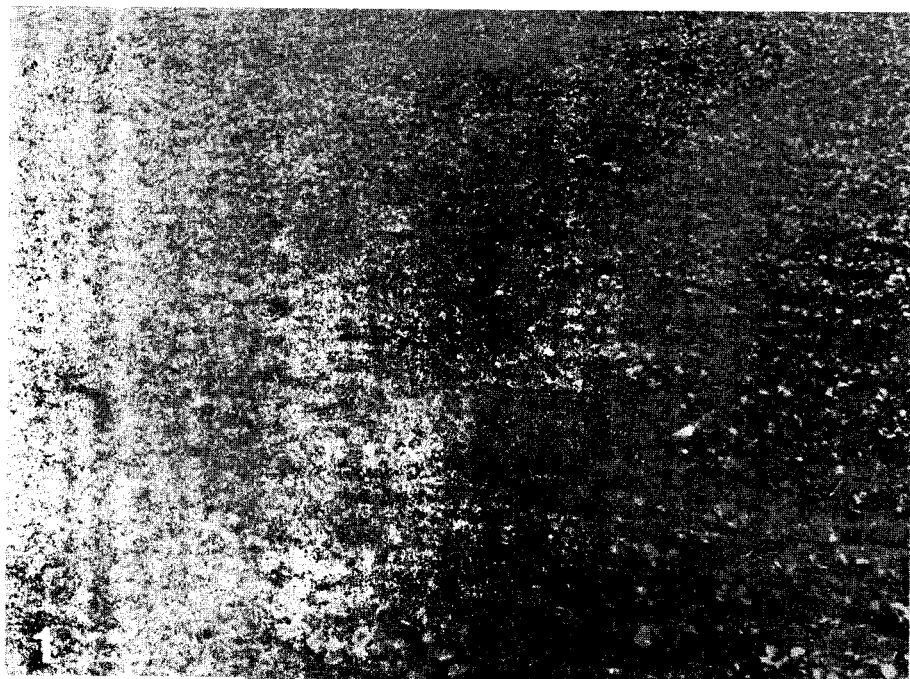
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## LEGEND

- Fig. 1. A canola field near Vegreville, Alberta showing patchy distribution of plants associated with the seedling blight disease.
- Fig. 2. Seedlings of canola showing lesions and constrictions on the hypocotyls due to the seedling blight disease.



The testing of rape for clubroot  
( *Plasmodiophora brassicae* L. )

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Isolation, obtaining and preservation of pathogen

Inoculum of durable spores is obtained from galls of plants infected, and the obtainment just before plant disintegration is the best. Galls can be stored at the temperature  $-13^{\circ}\text{C}$ , even in the course of several years without any loss of their viability. The viability is negatively influenced by thawing and then refreezing the material. Such an inoculum is also possible to obtain when sowing sensitive plants (Chinese cabbage cv. 'Granaat' is the most suitable) into the soil severely infested.

Inoculum preparation

An appropriate inoculum is prepared by homogenizing galls being fresh or frozen in distilled water. The homogenate obtained is filtered through a close plastic screen. Suspension is then adjusted with haemocytometer to the concentration required (to  $10^6$  spores/ml the most often). The suspension prepared in this way can be used even during two months provided that it is maintained at the temperature up to  $+5^{\circ}\text{C}$ .

Preparation of material

Thirty plants at least from every genotype are necessary to be evaluated. Only uniform seed being of good germinating capacity and untreated must be used for testing.

Inoculation

Growing substrate is prepared by mixing soil, peat and expanded perlite to be in ratio 2:4:1. Afterwards the substrate is sterilized with superheated steam for 30 min. Growing substrate obtained is filled into multipots the surface of which is smoothed. Then the substrate in individual pots is pressed in

order that five germinative seeds can be sown. By means of a doser 0,5 ml of inoculum is added to each of seeds. After inoculation seeds are covered up with perlite.

#### Incubation

Gall development is dependent on satisfactory moisture content in growing substrate as well as on its temperature (optimum temperature is 20 - 25°C). In winter period there is a need of supplementary illumination. When emerged the number of plantlets per pot is reduced to 3. After 10 days the temperature is possible to decrease to 18°C.

#### Evaluation

Evaluation is carried out within 6-7 weeks after inoculation. Individual plantlets are carefully pulled out, the substrate is removed from them, and then are rinsed in water so that galls arisen can be seen well.

Evaluation is made using the scale ranging from 0 to 3

- 0 - without infestation
- 1 - small galls on secondary roots
- 2 - medium-sized galls
- 3 - all or majority of roots modified into galls

Using values obtained the percentage of infestation is calculated, and even disease index (D. I.) is possible to determine. D. I. in sensitive genotypes is usually 80 - 100, and that in highly resistant genotypes is less than 20. Distinctness and/or agreement of disease severity between individual genotypes is expressed on the basis of statistical evaluation.

PRODUCTION OF A HOST-SPECIFIC  
PHYTOTOXIN BY ALTERNARIA BRASSICAE

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INTRODUCTION

Alternaria brassicae causes the blackspot disease of rapeseed (Brassica campestris and B. napus) which is economically important in Canada and many other parts of the world. Despite the importance of this disease, little information on the mode of pathogenesis is available. Several species of Alternaria Nees ex Fr. are reported to produce toxins. There are at least six host-parasite combinations, involving species of Alternaria, where the toxins produced are host-specific (Nishimura and Kohmoto, 1983).

Toxic effect of the culture filtrate of A. brassicae on B. campestris var. yellow sarson seedlings was reported by Husain and Thakur (1966). Later, two groups of non-specific toxins (in semi-purified preparations) produced by the fungus and capable of reproducing the disease symptoms on leaves were reported (Degenhardt, 1978). This paper reports on the isolation and identification of a host-specific toxin (AB-toxin) and several other interesting metabolites produced by A. brassicae in culture.

MATERIALS AND METHODS

Two methods were developed to isolate the AB-toxin produced by A. brassicae in VB juice culture broth supplemented with glucose. The first method included extraction with ethyl acetate, dissolution of the crude extract in water-methanol, successive partitioning with hexane, ethyl acetate and n-butanol, dry flash column chromatography (Pena-Rodriguez, 1986), and thin layer chromatography. The second method included gel filtration using Sephadex G-50 and G-25 columns, adsorption on activated charcoal, successive desorption with ethyl acetate and water saturated

n-butanol followed by silica gel column and Sep-Pak C<sub>18</sub> cartridge chromatography, respectively, and high pressure liquid chromatography (HPLC).

All fractions at each step were assayed for biological activity on B. napus cv. Altex leaves. Various chemical (ninhydrin reaction and derivatization) and physical techniques (infra-red spectroscopy, <sup>1</sup>H and <sup>13</sup>C nuclear magnetic resonance (NMR) spectroscopy, high resolution mass spectroscopy and fast atom bombardment mass spectroscopy) were used to identify the AB-toxin and some other metabolites.

Host-specificity of the AB-toxin was studied by applying the toxin both on the hosts and non-hosts of A. brassicae. The hosts included B. nigra, B. campestris var. yellow sarson, var. toria and cv. Candle, B. juncea cv. Lethbridge 22A, B. napus cv. Altex, B. hirta cv. Gisilba and B. rapa and the non-hosts included barley, corn, cowpea, cucumber, flax, oats, rye, tomato and wheat.

#### RESULTS AND DISCUSSION

Using the first method of AB-toxin isolation, extraction of the concentrated aqueous broth with ethyl acetate provided a complex mixture of compounds, including steroidal glycosides and glycerides, which proved difficult to separate. However, when a solution of the metabolites in aqueous methanol was extracted successively with hexane, ethyl acetate, and n-butanol, it was found that the active components were concentrated in the ethyl acetate extract. This "medium polarity" fraction was separated into its components by dry flash chromatography followed by thin layer chromatography.

Using the second method of toxin isolation, toxic activity was observed both in ethyl acetate and n-butanol extracts of activated charcoal on which the active component was previously adsorbed. Silica gel column and Sep-Pak C<sub>18</sub> cartridge chromatography of ethyl acetate and n-butanol fractions, respectively, and HPLC of both the fractions led to the isolation of AB-toxin in pure state.

Application of AB-toxin on rapeseed leaf resulted in the development of necrosis and chlorosis, characteristic of the blackspot disease (Figs. 1, 2).

The most active component (AB-toxin), mp 225 - 227 degrees,  $[\alpha]_D^{237}$  degrees, was shown by high resolution mass spectrometry to possess the molecular formula  $C_{30}H_{51}N_5O_7$ . Analysis of the mass spectrum and the  $^1H$  NMR and  $^{13}C$  NMR spectra of the compound and its methanolysis product indicated that it was destruxin B (Fig. 3a), a cyclodepsipeptide previously isolated from *Metarhizium anisopliae*, a fungus which is pathogenic to silkworms (Tamura *et al.*, 1964). Comparison with an authentic sample confirmed the identity. Two other cyclodepsipeptides, the known desmethyldestruxin B (Fig. 3b) and the previously unknown homodestruxin B (Fig. 3c), were also isolated and identified. The location of the extra methylene group in homodestruxin B was determined by mass spectrometry.

During the course of separation of the biologically active cyclodepsipeptides depicted as a, b, and c in Figure 3, we also isolated and determined the structures of three new sesquiterpenes, albrassitriol (Fig. 3d), isoalbrassitriol (Fig. 3e), and deoxyuvidin (Fig. 3f). A biogenetically interesting new compound which we have called brassicadiol was also obtained and shown to possess the structure shown in Figure 3g. The details of the isolation and structure elucidation of the metabolites have been described (Ayer *et al.*, 1987a, b; Bains and Tewari, 1987).

The order of sensitivity of Brassicas to AB-toxin was similar to their order of susceptibility to *A. brassicae*. In a decreasing order of sensitivity/susceptibility it was *B. nigra*, *B. campestris* var. *yellow sarson* and var. *toria* > *B. juncea* cv. Lethbridge 22A > *B. campestris* cv. Candle, *B. napus* cv. Altex and *B. hirta* cv. Gisilba > *B. rapa*. On the most susceptible host, the minimum concentration of AB-toxin required to cause symptoms was between 15 and 30  $\mu g ml^{-1}$ . On the non-hosts, even toxin concentration of 300  $\mu g ml^{-1}$  did not cause any symptoms. These results indicated that the toxin isolated is host-specific (Bains and Tewari, 1987).

This is the first report on production of a host-specific toxin by a pathogen of rapeseed. The AB-toxin can be used for host resistance selection using conventional and tissue culture methods.

#### ACKNOWLEDGEMENT

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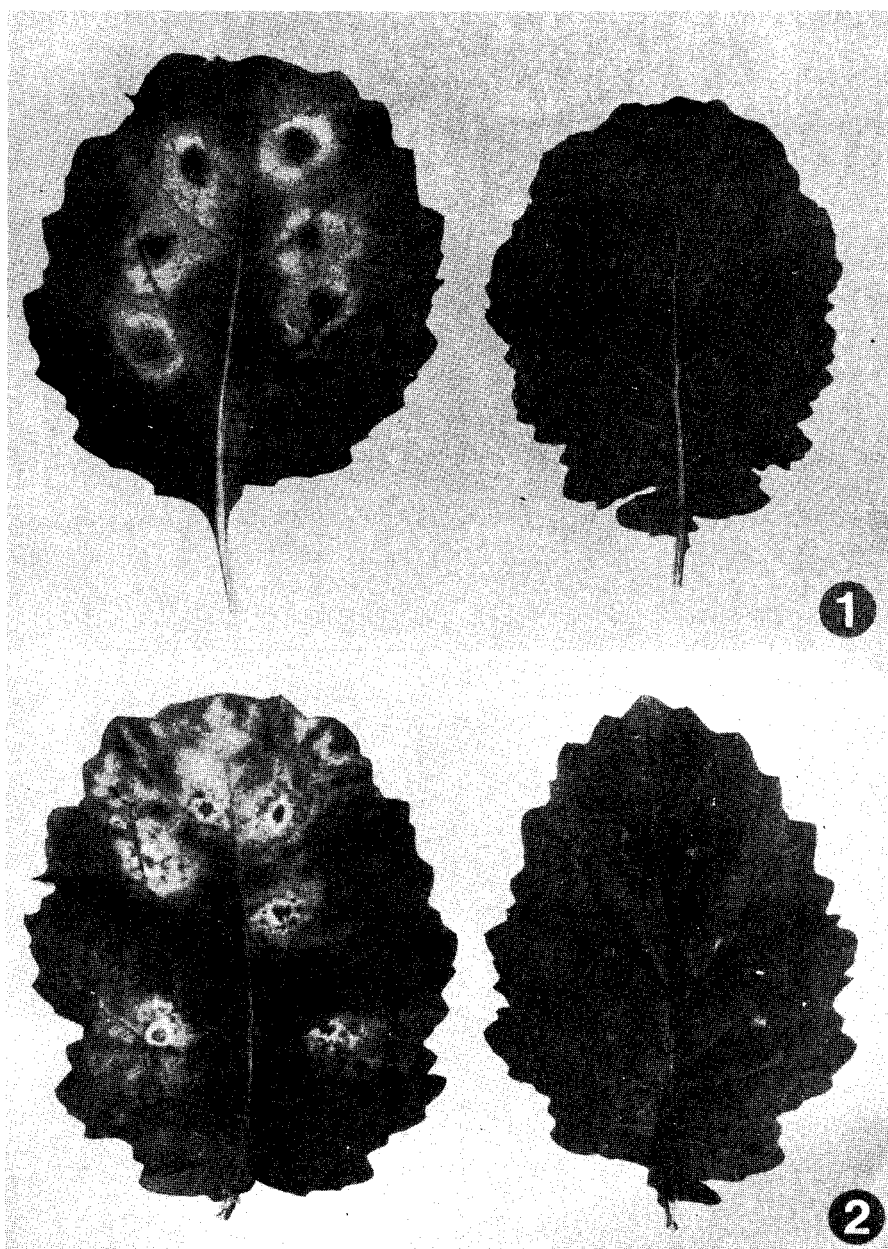
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## LEGEND

- Fig. 1. Symptoms caused by A. brassicae on B. napus cv. Altex (left leaf). Right leaf-control.
- Fig. 2. Symptoms caused by AB-toxin on B. napus cv. Altex. (left leaf). Right leaf-control.
- Fig. 3. Structural formulae of A. brassicae metabolites. a. Destruxin B (AB-toxin). b. Desmethyldestruxin B. c. Homodestruxin B. d. Albrassitriol. e. Isoalbrassitriol. f. Deoxyuvudin. g. Brassicadiol.





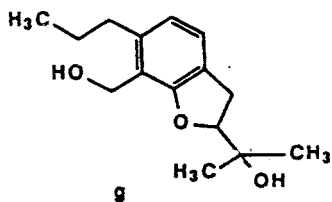
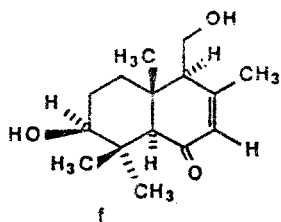
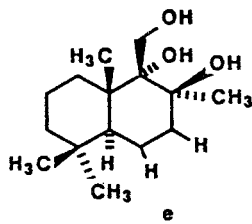
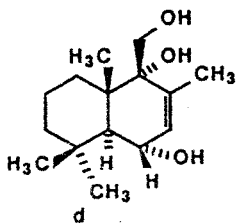
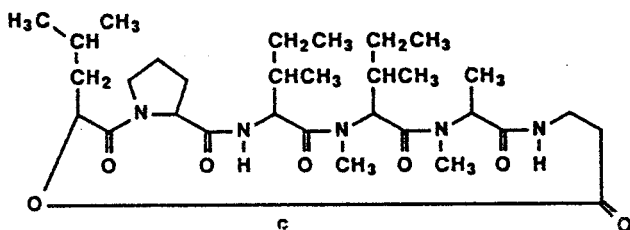
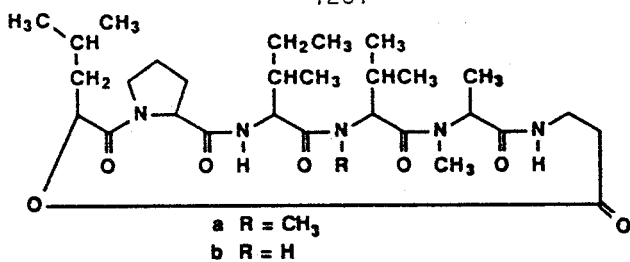


Fig. 3

THE PERFECT STAGE OF PYRENOPEZIZA BRASSICAE ON OILSEED RAPE AND ITS  
AGRICULTURAL IMPLICATIONS

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Introduction

Light leaf spot of brassicas, caused by *Pyrenopeziza brassicae* Sutton and Rawlinson (Rawlinson, Sutton & Muthyalu, 1978), has been common for a decade on oilseed rape (*Brassica napus* spp. *oleifera*) crops in the United Kingdom and is now regarded by some as the most important disease of rape in the United Kingdom. The disease occurs on oilseed rape elsewhere in Europe, and recently there have been severe attacks in France. Spread of the disease was thought to occur over only short distances by rainsplash of conidia (Rawlinson, Sutton & Muthyalu, 1978). The perfect stage of *P. brassicae* has previously been reported only on brassica crops in Ireland (Staunton & Kavanagh, 1966) and New Zealand (Cheah, Hartill & Corbin, 1980). We report the natural occurrence of the perfect stage of *P. brassicae* on an oilseed rape crop in the United Kingdom and discuss the possible implications for disease spread. A full account of the experimental procedures has been given by McCartney, Lacey & Rawlinson (1986) and Lacey, Rawlinson & McCartney (1987).

Identification of the teleomorph of *P. brassicae*

During the spring of 1986 leaf debris was collected from the soil surface beneath an infected oilseed rape crop, cv. Jet Neuf. Samples

were placed in a large wind tunnel and spores, mainly in groups of four, were trapped 3 m downwind. Spores were also found when samples were suspended over a microscope slide in a small enclosed chamber. The debris was examined microscopically and small apothecia (80  $\mu\text{m}$  in diameter) with asci containing spores were found on the leaf laminae. Although the apothecia were much smaller than those described from culture of the type (Rawlinson *et al.*, 1978), and lacked the dark outer excipulum, the asci and ascospores were similar to those of other records (Table 1).

Table 1. Measurements of apothecia, asci and ascospores of *Pyrenopeziza brassicae* found on oilseed rape compared with the type culture.

| Source  | Apothecia diameter (mm) | Asci length x width ( $\mu\text{m}$ ) | Ascospores length x width ( $\mu\text{m}$ )                        |
|---|-------------------------|---------------------------------------|--|
| Type (culture) (Rawlinson <i>et al.</i> , 1978) | 1                       | 80-100 x 7-9.5                        | 12.5-18.5 x 2.5-3  |
| Oilseed rape this study                         | small 0.03-0.1          | 33-63 x 4.2-8.4                       | 7.5-15.1 x 1.8-2.8*  |
|   | large 0.13-0.58         | 44-89 x 5.2-8.9                       | 7.9-13.1 x 1.8-2.8*  |
|   |                         |                                       | 9.4-18.8 x 1.8-3.7 <sup>a</sup><br>9.4-18.8 x 1.8-3.2 <sup>b</sup> |

\* Measurements taken mostly from spores remaining within the ascus.

<sup>a</sup> Released in laboratory. <sup>b</sup> Caught in Burkard trap.

As the season progressed more debris was examined and larger apothecia (up to 0.58 mm diameter) were found on veins and petioles (Fig. 1). The apothecia were black, cup-shaped structures with a pale margin. Asci within the hymenium measured 33-89  $\mu\text{m}$  x 4.2-8.9  $\mu\text{m}$  and contained eight ascospores. Paraphyses were hyaline, septate, as long as the asci and 2  $\mu\text{m}$  in diameter. The ascospores were hyaline, cylindrical, straight or slightly curved with rounded ends and were

sometimes septate. The structures from oilseed rape differed little in size from those recorded on other brassica crops.

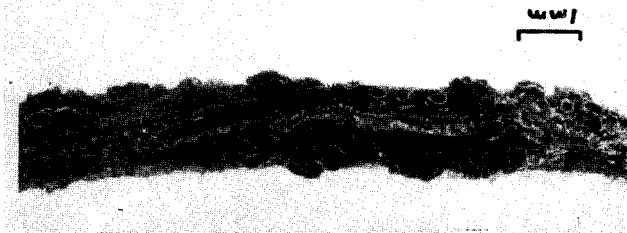


Fig. 1: Apothecia of *P. brassicae* on a decaying petiole of oilseed rape cv. Jet Neuf.

Apothecia with immature asci developed on cultures of *P. brassicae* grown from mass isolates of conidia taken from natural acervuli on rape leaves. Cultures typical of the *P. brassicae* anamorph were also produced by allowing ascospores from apothecia on a petiole to discharge on to agar in an inverted petri dish. After 56 days in an incubator at 15°C these cultures produced apothecia with asci containing differentiated ascospores. Conidial suspensions from these cultures were sprayed onto rape plants (cv. Jet Neuf) and produced typical light leaf spot lesions thus confirming the identity of the teleomorph.

#### Dispersal of ascospores

*P. brassica* spores were caught using a seven day recording volumetric spore trap (Burkard Manufacturing Co. Ltd., Rickmansworth, U.K.), placed in the centre of an infected crop. The trapped spores were frequently in groups of four similar to those caught in the wind tunnel.

Comparison of daily rainfall with daily average spore concentration, (Fig. 2) showed that periods of spore liberation and dispersal were closely associated with periods of rain. However, the

largest spore concentrations did not necessarily coincide with days with the highest rainfall. Indeed, spores were present in the air on days when no rainfall was recorded. A more detailed examination of spore catches showed that spores were airborne several hours after rain. Maximum spore concentrations were often found within 12 h of rain and in the early morning, suggesting that the crop must be wet, either from rain or dew, to initiate spore liberation.

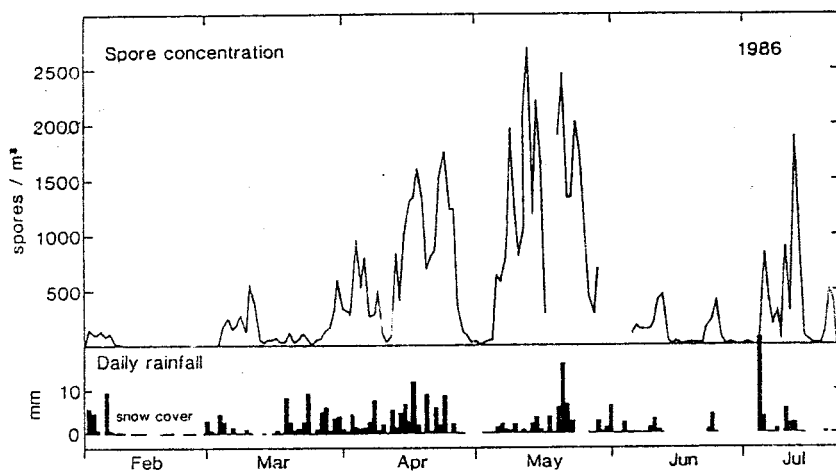


Fig. 2: Average number of spores  $\text{m}^{-3}$  sampled over 24 h (0900-0900 GMT) in the Burkard trap and total rainfall for the same period from 1 February to 20 July 1986.

Ascospore concentrations at five heights above and four distances downwind of the infected crop were measured using rotorod traps on several occasions during the summers of 1985 and 1986. Spore concentrations decreased with downwind distance from the crop. The pattern of decrease was approximately exponential, concentration near the ground decreasing by half in a distance of between 7 and 10 m from

the field edge, typical of dry windborne spores. Spore concentration also decreased with height above the crop, showing that the spores originated within the crop. The concentration at between 1 and 1.5 m above the crop was about half that at the top of the crop.

A gradient transfer model of spore diffusion was used to investigate the implications of the observed ascospore concentrations for spore dispersal away from the crop (McCartney, Lacey & Rawlinson, 1986). Ascospore concentrations up to 100 m downwind of the field were calculated. The calculations suggested that the spore plume would have dispersed so that at least 50% of the spores carried beyond the edge of the field would have still been airborne 100 m downwind of the field edge. Then spore transport over distances of several km would have been possible.

#### Agricultural implications

Our discovery of the teleomorph of *P. brassicae* and of the possibility of long distance spore dispersal, may help explain the increase in light leaf spot seen in vegetable brassica crops grown near oilseed rape (Gladders & McPherson, 1985). If the teleomorph becomes common on other cultivars of oilseed rape and on vegetable brassicas, there will be important consequences for agriculture and horticulture arising from the greater potential for genetic variation both in pathogenicity and in sensitivity to commonly-used fungicides (Iltott *et al.*, 1986). The possible consequences should now be investigated further in view of the importance of oilseed rape in Western Europe and the high cash value of vegetable brassicas. Wider searches for the teleomorph in crops in other locations must precede a full evaluation of the agricultural and horticultural significance of our finding.

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THE NEW MODELS OF TRAPS FOR MONITORING SOME OF  
THE OIL SEED RAPE PESTS

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The yellow water traps designed by Moericke are commonly used for forecasting some pests on the plantations of oil seed rape. Different models of that kind of traps are in use in Europe. They are accepted by local plant protection service. One of the most important weakness of the yellow traps is the fact that proper estimation of the insects number needs a lot of work.

Two kinds of new traps have been designed and tested in 1986. The first model was the ball-shaped, the second one pyramid-shaped. The colour of the traps is sulphur yellow. Solvurode aerosol /Société de Produits chimiques Sovilo France/ have been used to cover the surface of the trap with glue.

The results of the catch of the cabbage seedpod weevil /*Ceutorhynchus assimilis* Payk./ on the above mentioned sticky-yellow traps were compared with yellow-water traps.

The number of the weevils on sticky-yellow traps was 5-7 times higher than in the yellow-water traps. The time of the work was 3 times shorter when new models were used.

The new traps seem to be very promising for forecasting cabbage seedpod weevil and other *Ceutorhynchus* spp.

ALTERNARIA BRASSICAE ( Berk.) Sacc.: ETUDE DE PRODUITS  
FONGICIDES ET METHODES D'ECHANTILLONNAGE

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### I / INTRODUCTION

L'étude en serre de la lutte fongicide contre ALTERNARIA BRASSICAE réalisée au CETIOM en 1980, 1981 et 1982 a conduit à des résultats encourageants (6ème Congrès International sur le colza - Paris - 1983)/1/PERES 1983/. Cette recherche a été poursuivie en 1983 et 1984 dans un triple but:

- rendre plus performante la technique de contamination artificielle
- améliorer les méthodes d'appréciation de l'action des produits
- sélectionner de nouveaux produits efficaces contre le champignon en lutte préventive et curative.

### II / METHODE DE CONTAMINATION ARTIFICIELLE

Rappelons que la contamination artificielle du végétal est réalisée par la pulvérisation d'une suspension contaminante de spores d'ALTERNARIA BRASSICAE (30000 spores/ml) produites in vitro selon la méthode décrite par BILLOTTE /2/1963/.

Cette méthode est rendue plus performante par l'utilisation, dans la serre, d'un système diffuseur de brouillard permettant de contrôler avec précision les conditions de température (19-22°C) et d'hygrométrie (100%) favorables à la réussite de la contamination .

### III / METHODES D'APPRECIATION DE L'ACTION DES PRODUITS

Elles reposent sur deux critères d'observation:

- le niveau d'attaque visible (nombre et taille des taches d'ALTERNARIA)
- le pouvoir de sporulation des taches d'ALTERNARIA (à mettre en évidence au laboratoire).

#### A/Niveau d'attaque visible

Deux méthodologies différentes d'observation du niveau d'attaque en serre sont adaptées à la lutte curative d'une part et à la lutte préventive d'autre part.

a/En lutte curative

En lutte curative, l'observation s'appuie sur l'étude de l'évolution, après traitement, d'une population de 30 taches d'ALTERNARIA (définie et repérée sur une feuille ou une silique de chaque plante juste avant les traitements). Toutes les plantes de l'essai sont ainsi observées 10 et 20 jours après les traitements (tableau 3)

b/En lutte préventive

En lutte préventive on compare à un moment précis les niveaux d'attaque sur feuilles ou siliques entre les traitements. Les risques de fluctuations nécessitent de fixer les caractéristiques qualitatives et dimensionnelles de l'échantillon à observer:

- nombre de plantes et d'organes à observer
- localisation de l'organe à observer
- surface de l'organe à observer

Les résultats de cette étude méthodologique qui s'appuie sur une analyse statistique fine conduisent aux conclusions suivantes:

- nombre de plantes et d'organes à observer

\* dans le cas de l'étude sur feuilles le nombre de plantes à observer par traitement est de 3 ou 4, au dessus de 4 la précision ne s'améliore pas

\* dans le cas de l'étude sur siliques le nombre minimum de plantes à observer est de 5; au dessus de 5 la précision ne s'améliore que peu. Le nombre minimum de siliques à observer par plante est de 6; au delà de 6 siliques la précision ne s'améliore qu'assez peu (graphique 1).

- localisation de l'organe à observer

\* dans le cas de l'étude sur feuilles il apparaît que la 3ème et la 4ème feuilles en partant de la base (niveau moyen) sont les plus fiables

\* dans le cas de l'étude sur siliques le maximum de précision est obtenu en observant les siliques des trois ramifications supérieures.

- surface de l'organe à observer (pour CV < 12)

\* sur feuilles (graphique 2), la surface totale à observer sur l'ensemble de l'échantillon d'un même traitement sera de 20 à 30 cm<sup>2</sup> pour les feuilles des niveaux 3 et 4 retenus (soit 5 à 7,5 cm<sup>2</sup>/feuille quand l'observation est répartie sur 4 feuilles ou répétitions)

\* sur siliques des trois premières ramifications il faudra observer 20 à 30 secteurs de 1cm sur l'ensemble de l'échantillon d'un même traitement (soit 4 à 6 secteurs de 1cm / silique quand l'observation est répartie sur 5 siliques ou répétitions).

Tous ces résultats, qui fixent le nombre et la localisation des organes à observer ainsi que le nombre de plantes à observer, n'ont valeur de règle que sur des essais conduits dans les mêmes conditions (essais de serre avec contamination artificielle à un stade bien précis).

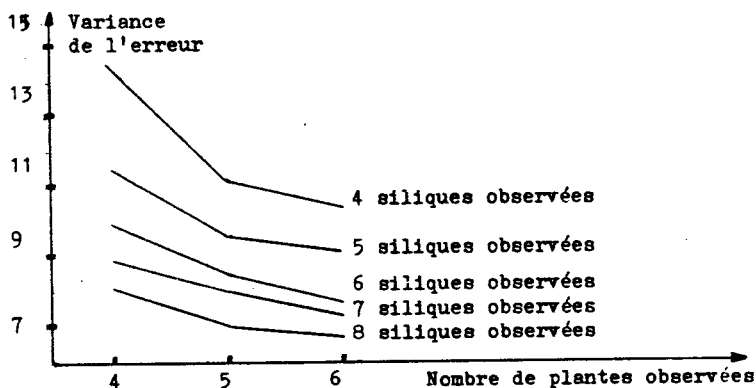
B/Pouvoir de sporulation des taches d'ALTERNARIA

On prélève sur 5 feuilles ou siliques malades un fragment d'organe porteur de taches d'ALTERNARIA que l'on place en chambre humide pendant 4 à 5 jours et on note la présence de spores émises par les taches (observations microscopiques) en les quantifiant suivant l'échelle suivante:

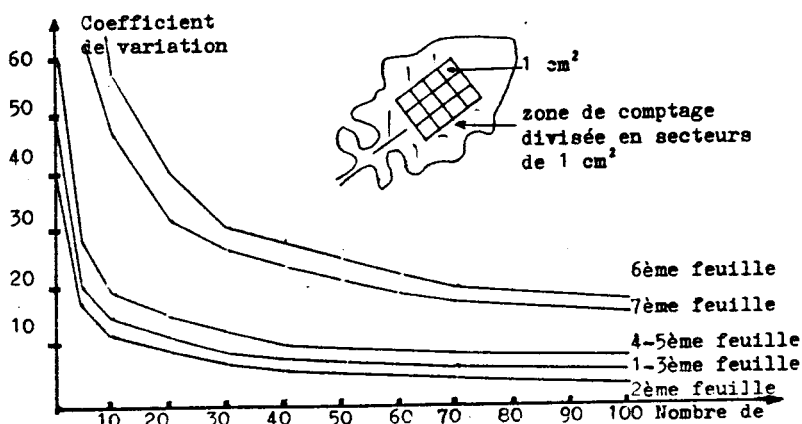
- note 0 : absence de spores  
 ,, 1 : spores en quantité faible  
 ,, 2 : ,, ,, ,, moyenne  
 ,, 3 : ,, ,, ,, forte  
 ,, 4 : ,, ,, ,, très forte (+ feutrage)

Ainsi 4 contrôles sont effectués la veille et le lendemain des traitements puis 10 et 20 jours plus tard (graphique 3).

Ce type de contrôle permet notamment d'observer si les produits empêchent ou limitent, par leur action antisporeuse, les contaminations secondaires développées le plus souvent à partir des feuilles de la base.



Graphique 1: évolution de la variance de l'erreur en fonction du nombre de plantes et de siliques observées



Graphique 2 : évolution du coefficient de variation en fonction du nombre de secteurs observés sur les feuilles

#### IV / RESULTATS D'ESSAIS DE LUTTE PREVENTIVE ET CURATIVE

Ces essais sont conduits suivant la méthodologie précédemment décrite.

##### A/ Essais de lutte préventive

Les produits sont appliqués 1 jour avant la contamination artificielle.

###### Premier essai - 1983 - (tableau 1)

Tous les produits expérimentés dans cet essai donnent des résultats d'efficacité et de rémanence pendant trois semaines hautement significatifs, qu'ils soient appliqués au stade "feuilles" (D2) ou au stade "siliques" (G4+5 jours). Ces efficacités ne sont pas significativement différentes entre produits, néanmoins des produits tels que IPRODIONE à 1kg de p. f./ha ou l'association CARBENDAZIME + THIRAME + FOLPEL à 4 l de p.f./ha assurent une protection beaucoup plus complète que le soufre micronisé à 14 l de p.f./ha.

###### Deuxième essai - 1984 - (tableau 2)

Cet essai, réalisé au stade "siliques" (G4+10 jours), permet de confirmer les meilleurs produits de l'essai précédent et d'expérimenter de nouvelles matières actives commercialisées ou non. Les résultats sont excellents puisque, sous l'effet des produits, la formation des taches d'ALTERNARIA est pratiquement nulle.

Les efficacités des produits observés sur ces deux essais sont d'autant plus satisfaisantes que les niveaux d'attaque révélés par les témoins non traités sont élevés (technique de contamination artificielle très performante).

##### B/ Essai de lutte curative - 1983 -

Les produits étudiés sont appliqués au stade D2 sur feuilles dès apparition des premières taches d'ALTERNARIA, 3 jours après la contamination artificielle.

Les résultats exposés dans le tableau 3 et le graphique 3 permettent de porter une appréciation sur l'activité fongicide de chaque produit.

L'IPRODIONE à 1 kg de p.f./ha reste le meilleur produit en assurant un très bon contrôle de la population de taches et une très bonne action antisporelante.

L'activité significative du FENARIMOL à 2,5 l de p.f./ha et du NUARIMOL à 0,8 l/ha sur la population de taches ne se manifeste qu'au bout de 20 jours mais leur activité antisporelante est du même niveau que celle de l'IPRODIONE.

L'association CARBENDAZIME + THIRAME + FOLPEL à 4 l de p.f./ha assure un très bon contrôle de la population de taches, par contre elle n'exerce pas d'action antisporelante.

A l'inverse, le FG 163 à 2,75 kg de p.f./ha n'agit pas sur la population de taches mais par contre réduit significativement leur pouvoir de sporulation pendant 10 jours.

Le soufre micronisé à 14 l de p.f./ha est par contre totalement inefficace dans cet essai.

## V / CONCLUSION

L'ensemble de ces travaux conduit à des résultats positifs tant sur le plan de la méthodologie que de l'efficacité des produits.

La technique de contamination artificielle est rendue plus performante et deux méthodes d'appréciation de l'action des produits adaptées respectivement à la lutte curative et préventive sont mises au point et analysées par une étude statistique fine permettant de fixer les caractéristiques idéales de l'échantillon à observer.

Tous les produits expérimentés en lutte préventive s'avèrent statistiquement efficaces aussi bien en applications précoces (D2) qu'en applications plus tardives (G4 dépassé).

En lutte curative, l'efficacité des produits se manifeste différemment suivant les cas:

- IPRODIONE, FENARIMOL et NUARIMOL contrôlent à la fois les populations de taches et leur pouvoir de sporulation (l'IPRODIONE restant le meilleur produit)

- d'autres produits contrôlent, ou la population de taches (CARBENDAZIME + THIRAME + FOLPEL), ou le pouvoir de sporulation (FG 163).

\*\*\*\*\*

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| MATIERE ACTIVE (PA) | QUANTITE EN M.A. DU P.F. | DOSES/HA DE PRODUIT FORMULE (PF) | NOMBRE DE TACHES D'ALTERNARIA |              |                           |              |
|---------------------|--------------------------|----------------------------------|-------------------------------|--------------|---------------------------|--------------|
|                     |                          |                                  | 10 JOURS APRES TRAITEMENT     |              | 20 JOURS APRES TRAITEMENT |              |
|                     |                          |                                  | SUR FEUILLES                  | SUR SILIQUES | SUR FEUILLES              | SUR SILIQUES |
| IPRODIONE           | 50 %                     | 1 kg                             | 1,3 **                        | 3,2 **       | 1 **                      | 3,5 **       |
| FENARIMOL           | 40 g/l                   | 2,5 l                            | 22 **                         | 26 **        | 21,8 **                   | 29,2 **      |
| CARBENDAZIME        | 30                       | 4 l                              | 1,7 **                        | 5,5 **       | 2,3 **                    | 5,8 **       |
| + THIRAME + FOLPEL  | +230+430g/l              |                                  |                               |              |                           |              |
| SOUFRE MICRONISE    | 80 %                     | 14 l                             | 59,7 **                       | 39,7 **      | 60,2 **                   | 41,7 **      |
| FG 163              | 58% + 21%                | 2,75 kg                          | 15,8 **                       | 4,2 **       | 13,2 **                   | 4,5 **       |
| TEMOIN NON TRAITÉ   |                          |                                  | 213,2                         | 145,5        | 259,2                     | 154,5        |
| F DES TRAITEMENTS   |                          |                                  | HAUTEMENT SIGNIFICATIF        |              |                           |              |

TABLEAU 1 : LUTTE PREVENTIVE - Premier essai - 1983 :  
 ETUDE COMPARÉE DU NOMBRE DE TACHES D'ALTERNARIA DENOMBREES SUR 4 FEUILLES  
 ET SUR 8 SILIQUES PAR PLANTE

\*\* : efficacité hautement significative

| MATIERE ACTIVE M.A.      | QUANTITE EN M.A. DU P.F. | DOSES/HA DE PRODUIT FORMULE (P.F) | NOMBRE DE TACHES D'ALTERNARIA |                            |
|--------------------------|--------------------------|-----------------------------------|-------------------------------|----------------------------|
|                          |                          |                                   | 10 JOURS APRES TRAITEMENTS    | 20 JOURS APRES TRAITEMENTS |
| IPRODIONE                | 50 %                     | 1 kg                              | 0,02                          | 0,02                       |
| PROCHLORAZ               | 450 g/l                  | 1,5 l                             | 0,12                          | 0,10                       |
| FG 163                   | 58 % + 21 %              | 2,5 kg                            | 0                             | 0                          |
| CARBENDAZIME             | 30                       | 3,5 l                             | 0,04                          | 0,10                       |
| +THIRAME+FOLPEL          | +230+430g/l              |                                   |                               |                            |
| MANCOZEBE                | 453 g/l                  | 7 l                               | 0,02                          | 0,02                       |
| PROCYMIDONE              | 500 g/l                  | 1 l                               | 0,04                          | 0,06                       |
| CHLOROTHALONIL           | 500 g/l                  | 3 l                               | 0                             | 0                          |
| FOLPEL+CAPTAFOL +CAPTANE | 44 % + 8 % +8 %          | 3 kg                              | 0,04                          | 0,04                       |
| TEMOIN NON TRAITÉ        |                          |                                   | 16,4                          | 17,3                       |

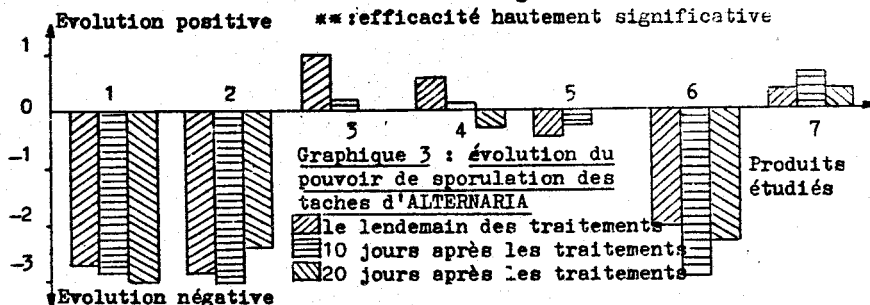
TABLEAU 2 : LUTTE PREVENTIVE - Deuxième essai - 1984 :  
 ETUDE COMPAREE DU NOMBRE MOYEN DE TACHES D'ALTERNARIA DENOMBREES PAR SILIQUE (secteur de comptage de 4 cm)

| CODE | MATIERE ACTIVE (M.A.) | QUANTITE EN M.A. DU P.F. | DOSES/HA DE PRODUIT FORMULE (P.F) | % D'AUGMENTATION DU NOMBRE DE TACHES |                            |
|------|-----------------------|--------------------------|-----------------------------------|--------------------------------------|----------------------------|
|      |                       |                          |                                   | 10 JOURS APRES TRAITEMENTS           | 20 JOURS APRES TRAITEMENTS |
| 1    | IPRODIONE             | 50 %                     | 1 kg                              | -3,3 **                              | 1,7 **                     |
| 2    | FENARIMOL             | 40 g/l                   | 2,5 l                             | 11,7                                 | 12,8 *                     |
| 3    | CARBENDAZIME          | 30                       | 4 l                               | 1,7 **                               | 6,1 **                     |
| 4    | +THIRAME+FOLPEL       | +230+430g/l              |                                   |                                      |                            |
| 5    | SOUFRE MICRONISE      | 80 %                     | 14 l                              | 33,3                                 | 33,3                       |
| 6    | FG 163                | 58 % + 21 %              | 2,75 kg                           | 15,6                                 | 26,7                       |
| 7    | MUARIMOL              | 120 g/l                  | 0,8 l                             | 20,6                                 | 18,3 *                     |
| 7    | TEMOIN NON TRAITÉ     |                          |                                   | 23,3                                 | 32,2                       |
|      | F DES TRAITEMENTS     |                          |                                   | HAUTEMENT                            | SIGNIFICATIF               |

TABLEAU 3 : LUTTE CURATIVE - 1983 : EVOLUTION COMPAREE DU NOMBRE DE TACHES D'ALTERNARIA SUR FEUILLES (exprimée en %)

\* : efficacité significative

\*\* : efficacité hautement significative



CONTRIBUTION A L'ETUDE DE LA LUTTE CONTRE L'INOCULUM  
DE SCLEROTINIA SCLEROTIORUM DANS LES CULTURES DE  
COLZA.

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RESUME

Les premiers résultats positifs dans la recherche de la lutte par voie chimique destinée à limiter la carpogénèse des sclérotés de SCLEROTINIA SCLEROTIORUM présents dans le sol ont incité à poursuivre les travaux.

Plusieurs thèmes sont abordés, l'objet de cette publication étant de préciser les conséquences à plus long terme de l'application de ces produits sur la physiologie des sclérotés.

Les résultats confirment la prééminence du mélange CIANAMIDE (200kg) + PERLUREE (100 kg) soit tel que, soit en association avec le CRYPTONOL SPECIAL E (20 kg), la dose de PERLUREE étant alors réduite à 50 kg/ha.

La conservation des sclérotés dans le sol dure plus longtemps lorsque les sclérotés ont été soumis à l'action de produits efficaces, l'inhibition des apothécies même totale au moment du stade sensible de la culture, restant provisoire.

INTRODUCTION

L'étude systématique de l'efficacité des produits chimiques dans le but de bloquer par traitement du sol la formation des apothécies, a fait l'objet au CETIOM, depuis plusieurs années, d'un programme important: PIERRE 1983, 1984, 1985). Mais l'aspect de l'influence du traitement des sclérotés sur leur viabilité n'avait pas été jusqu'à présent envisagée dans nos essais.

Dans ce travail, la viabilité de la population des sclérotés est considérée dans ses diverses composantes: conservation dans le sol, aptitude à la fructification et intensité de la carpogénèse.

METHODE EXPERIMENTALE:

Le dispositif expérimental, directement tiré des conclusions de (1) et (2) est le suivant: les sclérotés, issus de plantes de tournesol attaquées, répartis en trois



TABLEAU n° 1 : EVOLUTION DE LA VIABILITE DES SCLÉROTES DE SCLEROTINIA SCLEROTIUM EN FONCTION DU TEMPS.

| N° de l'essai<br>Durée de l'essai  | EVOLUTION DE LA POPULATION |                            | APTITUDES A LA CARPOGENESE |               | INTENSITE DE LA CARPOGENESE<br>(6) |                   |
|--|----------------------------|----------------------------|----------------------------|---------------|------------------------------------|-------------------|
|  | 2<br>(+11 mois)            | 3<br>(+ 16 mois)           | 1<br>2 A<br>-70            | 2 B<br>+70    | 1<br>2 (-70)<br>+11                | 2 (+70)<br>+16    |
| Nombre théorique<br>Nbre sclérotés réels   | 108<br>58 à 75             | 96<br>16 à 54              | 216<br>-                   | 54<br>29 à 46 | 54<br>23 à 46                      | 54<br>16 à 54     |
|  | Présents en % de l'essai 1 |                            | En % de la population      |               | En %                               |                   |
| a produits peu<br>efficaces (4)<br>Intervalle de<br>confiance 0,05   | 62<br>± 6,75               | 50<br>± 8,5                | 45<br>± 6,03               | 23<br>± 7,65  | 9,62<br>± 5,05                     | 10,9<br>± 3,95    |
| m témoins<br>Produits efficaces n°9<br>Produits efficaces n°10   | 68<br>83<br>84             | 50 (1)<br>71 (2)<br>81 (3) | 50<br>0,05<br>0,05         | 11<br>2<br>0  | 37<br>9<br>24                      | 31<br>35<br>29    |
| Taux sclérotés détruits<br>Produits inefficaces<br>Témoin<br>Produit efficace n°9<br>Produit efficace n°10 | 38<br>32<br>17<br>16       | 50 (5)<br>50<br>29<br>19   |                            |               | 1,7<br>0,005<br>0,009              | 1<br>7<br>0       |
|  |                            |                            |                            |               | 5,1<br>14<br>9,5                   | 2<br>5,63<br>6,27 |

(1) 16 sur 32 sclérotés observés

(2) 23 sur 32

(3) 52 sur 64

(4) Données établies à partir d'un échantillon de 8 produits

(5) de l'échantillon initial

(6) Définie comme le rapport du nombre maximum d'apothécies observées sur le nombre de sclérotés fructifères.

catégories de poids moyen connu (0,09 - 0,19 - 0,38g), sont disposés par quatre ( la catégorie 2 étant représentée deux fois) de façon repérée dans des récipients à fond grillagé appelés tamis. Dix huit de ces tamis sont placés dans des bacs (50\*30\*5cm) formant la parcelle élémentaire. Chaque traitement est répété trois fois. Les bacs, placés au champ, sont soumis aux conditions climatiques naturelles à partir du 20 novembre 1984.

Lorsque les sclérotés présentent leurs premiers stipes, on applique les traitements chimiques sur le sol à raison de 500l/ha (6/05/85). Les bacs sont alors introduits sous le couvert d'une culture de colza.

Les premières apothécies apparaissent le 31/05; les observations se font une fois par semaine, et portent sur le nombre d'apothécies présentes par sclérote. Elles se poursuivent jusqu'au 27 juin.

Les bacs sont laissés au champ après la récolte du colza. La moitié de la population de sclérotés de chaque bac est prélevée le 16/10/85 (soit 108 sur 216). Cette moitié est elle-même subdivisée en deux échantillons égaux placés en bacs à germination remplis de sable, l'un mis en présence de graines de tournesol (essai 2B), l'autre non (essai 2A). Ces bacs sont disposés dans des salles à 20°C et régulièrement arrosés.

La même manipulation est reproduite en fin d'hiver, sans nouvelle subdivision, le 26/03/86 sur la dernière moitié des sclérotés traités avec les produits inactifs, sur le reliquat de la population de sclérotés récupérables pour les témoins et les produits actifs. Les observations sont de même nature que celles des essais précédents: nombre de sclérotés ayant fructifié, nombre d'apothécies présentes par sclérote à chaque date d'observation.

Notons que des difficultés liées à un orage qui a endommagé l'essai ont entraîné une réduction de la population de certains lots des témoins et des produits efficaces dans cette dernière partie de l'essai.

## RESULTATS:

### ETUDE EN FONCTION DU TEMPS DE L'ACTIVITE DES PRODUITS SUR LA VIABILITE DES SCLEROTES.

#### a) Evolution au cours du temps de la population de sclérotés étudiés.

Alors que le premier essai s'est déroulé avec les sclérotés présents dans le sol, les essais suivants ( 2 A et B, 3) ont été précédés de l'extraction des sclérotés ce qui a permis leur dénombrement.

Les sclérotés présents pouvaient être sains ou bien déjà attaqués par des microorganismes: dans ce cas ils sont mous au toucher sur une fraction ou sur leur totalité. Les résultats portent sur le nombre total.

Le tableau (1) indique l'évolution de la population des sclérotés pour un échantillon de produits classés inefficaces, pour le témoin et les deux produits classés très efficaces.

L'évolution de la population témoin montre que 32% des sclérotés a disparu après 11 mois passés dans le sol, et 50% après 16 mois. Le taux de la population de sclérotés

traités avec des produits peu efficaces est le même (test t). Cette population a donc un comportement identique à celui des sclérotés non traités alors que les sclérotés initialement traités avec le mélange CIANAMIDE + PERLUREE sont présents en nombre plus élevé que dans les cas précédents et ce de manière significative (risque 0,05).

On peut donc conclure que les mélanges de produits efficaces, s'ils inhibent la formation des apothécies se comportent comme des "conservateurs" de sclérotés.

#### b) Evolution de l'aptitude des sclérotés à la carpogénèse.

Les sclérotés sont placés en conditions favorables à la carpogénèse, soit naturellement (essai 1), soit artificiellement (essais 2 et 3). Les résultats expriment donc leur aptitude à la fructification à un moment donné.

L'évolution des témoins montre que 50% seulement des sclérotés présents dans le sol a été induit à la carpogénèse au cours de l'hiver dans les conditions de l'essai. A l'automne suivant, seulement 11% des 68% des sclérotés encore présents est apte à émettre des apothécies. Il y a donc eu perte de l'induction au cours de l'été qui a suivi l'apparition de la première génération d'apothécies. Les conditions de l'hiver suivant provoquent une remontée de l'induction du reliquat des sclérotés présents à 31%.

Il est intéressant de noter que l'aptitude à émettre des apothécies ne dépend pas uniquement des séquences climatiques hivernales: la proximité de graines de tournesol en germination accroît de 11 à 37% le taux de sclérotés aptes. Ce facteur d'induction pourrait être intéressant à étudier. Il indique au moins que le risque sclérotinia ne s'exprime pas uniquement en termes d'aptitude absolue mais aussi relativement à la culture choisie.

L'évolution de la population témoin s'inscrit tout à fait dans celle, statistiquement plus précise, des sclérotés soumis à l'action des produits peu efficaces.

Par contre, l'évolution de la population traitée avec des produits très efficaces montre des différences statistiques bien nettes: perte à peu près totale et durable de l'aptitude la première année du traitement. Il y a néanmoins retour à des taux statistiquement identiques à ceux des autres populations un an après l'application de ces produits sur le sol. Seule la présence des graines en voie de germination montre qu'il s'agit moins d'une perte de l'aptitude que d'une inhibition susceptible d'être levée (0 à 24%). L'inhibition paraît moins forte dans le cas du produit 10, soit en raison de la présence de quintozone, soit, ce qui est plus probable, à cause de la diminution de la quantité de PERLUREE présente.

#### c) Evolution de l'intensité de la carpogénèse.

Cette intensité est définie comme le rapport du nombre maximum d'apothécies observées par scléroté sur le nombre total de sclérotés fructifères de l'échantillon considéré.

L'échantillon témoin présente une diminution de près de la moitié de l'intensité de la carpogénèse au cours de la période automnale. Mais, au même moment, la proximité des graines de tournesol accroît de 5 fois le nombre de sclérotés présents. Au printemps suivant l'intensité de la carpogénèse redevient proche de celle observée un an auparavant.

Cette observation témoigne de ce qu'il y a moins perte de l'induction qu'inhibition de l'aptitude à la carpogénèse, cette inhibition pouvant être levée par d'autres facteurs que ceux qui induisent initialement.

Les produits peu efficaces affectent peu l'intensité de la carpogénèse lors de la première génération d'apothécies. Par contre cette intensité s'accroît considérablement à l'automne par rapport au témoin, ce qui signifie qu'il n'y a pas eu, là non plus, perte de l'induction. Celle-ci est peu sensible à la présence des graines de tournesol, ce qui confirme l'absence d'inhibition. L'intensité diminue au printemps, mais reste plus élevée que celle du témoin et est équivalente à celle des sclérotés traités avec des produits efficaces.

Ces produits très efficaces altèrent l'intensité au moment de la période normale d'apparition des apothécies d'une façon quasi complète. L'addition de quintozène, concomitant d'une diminution de la quantité de PERLUREE prolonge l'effet jusqu'à l'automne, mais cet effet peut être levé par la présence des graines de tournesol en germination. L'intensité de la carpogénèse un an après l'application des produits est double voire triple de celle des témoins et voisine celle observée sur la population de sclérotés soumis à l'action des produits peu efficaces.

#### DISCUSSION-CONCLUSIONS

Les sclérotés issus de plantes de tournesol, placés dans un sol à 1cm de profondeur et soumis aux conditions climatiques hivernales naturelles, sont susceptibles de fournir au moment de la floraison du colza 50% de sclérotés porteurs de 1,7 apothécies en moyenne par sclérote. L'année suivante, sur 50% de sclérotés restants, seulement 31% émettront chacun en moyenne deux apothécies.

L'application de produits très efficaces inhibe l'aptitude à la carpogénèse de façon durable dans la même année. Mais le taux de sclérotés présents est plus élevé l'année suivante et ces sclérotés sont susceptibles de fournir, avec un même taux d'aptitude que pour les témoins, une intensité de carpogénèse 5 à 6 fois supérieure. Un nouveau traitement s'imposera donc dans ces sols s'ils sont à nouveau emblavés d'une culture sensible.

Le traitement du sol avec des produits peu efficaces n'influe ni sur la destruction des sclérotés dans le temps ni sur leur aptitude à la carpogénèse à moyen et à long terme. C'est l'intensité de la carpogénèse qui sera accrue aussi bien à l'automne qu'au printemps suivant.

La proximité des graines de tournesol à l'automne entraîne une levée de l'inhibition de l'aptitude à la carpogénèse et accroît notablement l'intensité de celle-ci.

dans le cas des populations témoins et des populations traitées aux produits très efficaces. Cette observation montre que le risque sclerotinia, s'il dépend en partie du passé de la parcelle, est également lié à la culture en place.

Les produits très efficaces sont:

|                |                                      |   |   |      |
|----------------|--------------------------------------|---|---|------|
| n° 9 CIANAMIDE | à 200 kg/ha du produit titrant 19% N |   |   |      |
| +              | +                                    |   |   |      |
| PERLUREE       | à 100 kg/ha                          | " | " | 16%N |

|                   |                                      |   |   |       |
|-------------------|--------------------------------------|---|---|-------|
| n° 10 CIANAMIDE   | à 200 kg/ha du produit titrant 19% N |   |   |       |
| +                 | +                                    |   |   |       |
| PERLUREE          | à 50 kg/ha                           | " | " | 16% N |
| +                 | +                                    |   |   |       |
| CRYPTONOL SPECIAL | 20 kg/ha                             | " | " | 20%   |

de

quintozène.

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## CONTROL OF FUNGAL DISEASES IN WINTER OIL SEED RAPE

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## ABSTRACT

Experiments showed that two spring treatments using fungicides were effective in the control of oilseed rape fungal diseases. In small plot trials tests were carried out to determine the incidence and importance of downy mildew/ *Peronospora brassicae*/, stem rot/ *Sclerotinia sclerotiorum*/, stem canker/ *Phoma lingam*/, dark pod spot / *Alternaria* spp./ and grey mould/ *Botrytis cinerea*/. The following active ingredients were tested: prochloraz, carbendazim, iprodione, vinclozolin, procymidone, metalaxyl and copper oxychloride. The yield and 1000 seed weight were measured and estimated.

## INTRODUCTION

Observations made by plant protection service in Poland at the beginning of the eighties showed increasing infection of winter oilseed rape with fungal diseases. The fungus *P.lingam* caused important damage . In badly affected cases there was up to 50% crop failure. *S.sclerotiorum*, *Alternaria* spp., *B.cinerea* and *P.brassicae* also damaged rapeseed cultures.

The data concerning disease control with fungicides/ Barnes & Williams 1983, Evans & Gladders 1981, Cox et al. 1981, Gladders 1978, Goetz 1979, Hampel et al. 1981, Mes-  
seliere 1981/ underline the effectiveness of chemical methods. This encouraged us to start with similar experiments in Poland.

## MATERIALS AND METHODS

Experiments were carried out in randomized blocks with 4 replicates in Bąków/ near Opole/. The plot size was 20 m<sup>2</sup>. Oilseed rape plants cv.BKH 485/ '00'/ were naturally infested in field conditions. They were treated using

knapsack sprayer/ 500 l of water/ha /.

Fungicide applications

The first treatment - was performed when inflorescence rose above the level of the rosette/ 23 April, G.S. 3.2/ / Harper & Berkenkamp, 1975/.

The second treatment - was performed when lower pods started to fill/ 20 May, G.S. 4.3/.

The following products were evaluated:

| <u>Active ingredients</u>      | <u>Dose per hectare</u> |
|--------------------------------|-------------------------|
| iprodione                      | 0.750 kg                |
| iprodione + carbendazim        | /0.525 + 0.262/ kg      |
| metalaxyl + copper oxychloride | /0.200 + 1.600/ kg      |
| prochloraz                     | 0.675 l.                |
| prochloraz + carbendazim       | /0.450 + 0.120/ l.      |
| procymidone                    | 0.750 kg                |
| vinclozolin                    | 0.750 kg                |

Disease sampling - the following samples were taken:

leaves - 25 out of each plot were collected at random and they were examined for the presence of downy mildew/ *P.brassicae*/

stems - 50 consecutive stems in the middle row of each plot were assessed for the presence of stem canker / *P.lingam*/ and the incidence of stem rot/ *S.sclerotiorum*/ among the 250 consecutive stems in the two middle rows of every plot was ascertained

Pods - 100 pods per plot were sampled at random and the extent of damage by black pod spot/ *Alternaria* spp./ and grey mould/ *B.cinerea*/ was determined.

Leaves were collected when lower pods started to fill/21 May, G.S. 4.3/. Stems and pods were collected at G.S. 5.3 - the seeds in the lower pods were starting to become mottled green-brown.

The extent of *P.brassicae* was determined using the scale:

0 - no infection

1 - up to 3 sources of fungus growth on a leaf

2 - more than 3 sources of fungus growth on a leaf, less than 25% of leaf area infested

3 - more than 25% of leaf area infested.

Alternaria spp. were assessed using the following scale:

0 - no infection

1 - up to 3 spots on a pod

2 - more than 3 spots on a pod, less than 25% of pod area infested

3 - more than 25% of pod area affected.

Disease noxiousness was calculated and expressed as a mean disease index M.D.I. using the formula:

$$\text{M.D.I.} = \frac{b+2c+3d}{a+b+c+d} \quad \begin{array}{l} a, b, c, d, - \text{ number of leaves/pods in} \\ \text{each degree of disease respectively} \end{array}$$

The incidence of diseases was shown as a mean percent of damaged leaves, stems and pods.

10000 seeds out of each plot were counted/ using a seed counter/, weighed and expressed as a mean 1000 seed weight. The yield was weighed and calculated in tonnes per hectare. The results of the observations were estimated by analysis of variance and by comparing the means by the t-Student test.

#### RESULTS

P.brassicae/ downy mildew/ occurred in the field throughout the season. It appeared on leaves in the autumn and continued to affect the plants till harvest. The fungus occurred even on pods.

The effectiveness of fungicides in the control of disease on leaves is shown in Table 1. The pathogen was effectively controlled using a product containing metalaxyl + copper oxychloride. It reduced the severity of the disease M.D.I. from 1.95 in untreated to 0.51 in treated plots.

TABLE 1

Incidence and importance of P.brassicae on rape leaves

| Treatment                      | % Leaves infected  | M.D.I./ 0-3/ |
|--------------------------------|--------------------|--------------|
| Untreated                      | 98                 | 1.95         |
| vinclorolin                    | 100                | 1.94         |
| metalaxyl + copper oxychloride | 45 <sup>xxxx</sup> | 0.51         |

xxxx- significantly different from untreated at P=0.001



*S.sclerotiorum*/ stem rot/ damaged only 2.5% of stems in untreated plots. However, infested plants were completely destroyed 3 weeks before harvest and no yield was obtained. The experiments showed effective control of disease using iprodione+carbendazim, vinclozolin and procymidone/ Tab.2/

TABLE 2

Incidence of *S.sclerotiorum* and *P.lingam* on rape stems

| Treatment                      | % Stems infected      |                   |
|--------------------------------|-----------------------|-------------------|
|                                | <i>S.sclerotiorum</i> | <i>P.lingam</i>   |
| Untreated                      | 2.5                   | 38.0              |
| prochloraz + carbendazim       | 2.9                   | 20.5 <sup>x</sup> |
| prochloraz                     | 1.4                   | 37.5              |
| iprodione + carbendazim        | 0.7 <sup>xx</sup>     | 24.0              |
| iprodione                      | 1.7                   | 29.5              |
| vinclozolin                    | 0.8 <sup>x</sup>      | 22.5              |
| procymidone                    | 0.9 <sup>x</sup>      | 19.5 <sup>x</sup> |
| metalaxyl + copper oxychloride | 3.4                   | 36.5              |

xx - significantly different from untreated at P=0.05

x - " - " - " - " - " - P=0.10

The first symptoms of *P.lingam*/ stem canker/ were observed about 20th of June and stems were slightly cankered 3 weeks before harvest/ small blotches, lesions encircling less than half the stem/. The application of procymidone and prochloraz + carbendazim reduced disease incidence on stem bases/ Table 2/.

Infections of *Alternaria* spp./ dark pod spot/ were generally slight. M.D.I. was equal to 0.28 and 26% of pods were affected with the disease/ Table 3/. In plots treated with iprodione and procymidone the incidence and extent of disease were lowest of all.

*B.cinerea*/ grey mould/ occurred on pods due to damage caused by *Dasyneura brassicae* and *Ceutorhynchus assimilis*. About 4.7% of pods were affected by this fungus. Treatments with prochloraz, procymidone, vinclozolin and iprodione reduced disease occurrence/ Table 3/.

Under small plot trial conditions, the yield was rela-

tively low/ Table 4/. The highest yield increase was observed in plots treated with metalaxyl + copper oxychloride. Higher 1000 seed weight was stated after treatment using iprodione, vinclozolin, procymidone and prochloraz + carbendazim.

TABLE 3

Incidence of *Alternaria* spp. and *B.cinerea* on rape pods

| Treatment                      | <i>Alternaria</i> spp. |                     | <i>B.cinerea</i>    |
|--------------------------------|------------------------|---------------------|---------------------|
|                                | M.D.I./0-3/            | %Pods infected      |                     |
| Untreated                      | 0.28                   | 26.25               | 4.75                |
| prochloraz + carbendazim       | 0.15                   | 15.00 <sup>xx</sup> | 4.50                |
| prochloraz                     | 0.16                   | 15.75 <sup>xx</sup> | 0.50 <sup>xxx</sup> |
| iprodione + carbendazim        | 0.13                   | 12.75 <sup>xx</sup> | 2.75                |
| iprodione                      | 0.08                   | 8.25 <sup>xx</sup>  | 2.25                |
| vinclozolin                    | 0.18                   | 17.75               | 2.50                |
| procymidone                    | 0.09                   | 8.75 <sup>xx</sup>  | 2.50                |
| metalaxyl + copper oxychloride | 0.23                   | 22.50               | 2.75                |

xxx - significantly different from untreated at P=0.01

xx - " - " - " - " - " - P=0.05

TABLE 4

Yield and 1000 seed weight

| Treatment                      | Yield<br>t/ha | 1000 seed weight<br>g |
|--------------------------------|---------------|-----------------------|
| Untreated                      | 2.49          | 4.67                  |
| prochloraz + carbendazim       | 2.50          | 4.71                  |
| prochloraz                     | 2.67          | 4.66                  |
| iprodione + carbendazim        | 2.51          | 4.62                  |
| iprodione                      | 2.42          | 4.73                  |
| vinclozolin                    | 2.45          | 4.72                  |
| procymidone                    | 2.50          | 4.71                  |
| metalaxyl + copper oxychloride | 2.71          | 4.66                  |

## DISCUSSION

Effective control of stem rot, dark leaf/pod spot and grey mould in oilseed rape crops has already been reported

/ Barnes & Williams 1983, Evans & Gladders 1981, Cox et al. 1981, Hampel et al. 1981, Marshall & Harris 1984, Messeliere 1981/. This has been confirmed by the results obtained in Poland after procymidone, iprodione, vinclozolin and prochloraz treatments.

Yield increase worth noticing/ 0.22 t/ha / was observed in plots treated with metalaxyl + copper oxychloride. However this treatment was found to exert effective control only over downy mildew. The low increase in 1000 seed weight obtained in 4 cases was not followed by a better yield.

In 1986 stem canker appeared on stems/ from soil level up to 20 cm above/ relatively late, i.e. about 20th of June/ a month after the last treatments/. Samples were taken nearly 6 weeks after the last sprays. Although damage caused by fungus was not significant, differences between fungicide treatments were visible. Incidence of *P. lingam* was reduced in plots with procymidone and prochloraz + carbendazim. Analyses made at harvest showed no connection between yield and disease incidence 3 weeks before harvest. These results need further experiment.

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Kosten des Rapsschutzes vor Schädlingen, Krankheiten  
und Unkräutern im Landwirtschaftlichen Forschungsbe-  
trieb Magnice in den Jahren 1970 - 1985

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In den vergangenen 15 Jahren fanden in der VR Polen deutliche Veränderungen im Rapsanbau statt. Vor allen Dingen war eine signifikante Vergrösserung der Anbaufläche mit gleichzeitiger Lokalisierung des Rapsanbaus hauptsächlich im nordwestlichen Teil des Landes zu verzeichnen. Darüber hinaus änderte sich die Anbautechnologie, auch die Rapssorten, die grosse Mengen von Erukasäure enthalten, wurden durch neue Arten, wie Quinta, Jet Neuf ersetzt. Infolge dieser Veränderungen gewann auch der chemische Pflanzenschutz immer grössere Bedeutung.

Die vorliegende Arbeit zielt darauf ab, die Kosten der Pflanzenschutzmassnahmen zu analysieren und diese mit Rapsanbaukosten zu vergleichen, den Verbrauch von Pestiziden beim Rapsschutz zu analysieren und diesbezügliche Veränderungstendenzen sowie den ungefähren Rentabilitätsindex für Schädlings- und Unkrautbekämpfungsmassnahmen zu bestimmen.

In den Forschungen wurden folgende Methoden verwendet: die Methode der Einzelkosten zwecks Bestimmung der Kosten der Schutzmassnahmen und des Rapsanbaus /2/, die vertikale Analyse zwecks

Ermittlung der Veränderungen, die in den Jahren 1970 - 1985 sowohl in Anbaukosten als auch in den Kosten der Schutzmassnahmen vorkamen, und statistische Methoden zwecks Bestimmung der Veränderungstendenzen der untersuchten Merkmale /3/.

### Ergebnisse

Der Landwirtschaftliche Forschungsbetrieb Magnice, der zur Landwirtschaftlichen Akademie Wrocław gehört, umfasst eine Fläche von etwa 520 ha Ackerland. Die Bodenqualität ist in dem Betrieb gut, denn 70% der Ackerfläche nimmt der Boden der II. und III. Klasse ein. Im Anbauverhältnis ist hier der Rapsanteil differenziert, was vor allem auf ungünstige Klimabedingungen zurückzuführen ist /beispielsweise musste in den Jahren 1982 und 1985 ein Teil der Kultur im Frühling umgepflügt werden/.

Im Rapsanbau wird der chemische Schutz ein wichtiges Element des Produktionsprozesses. Im Verlauf von den analysierten 16 Jahren wurden Unkraut- und Schädlingsbekämpfungsmassnahmen sowie Saatgutbeize durchgeführt, wobei die Anzahl der unkrautbekämpfenden Massnahmen um 3% zugenommen und die der schädlingsbekämpfenden um 3% abgenommen haben /Tab.1/. Auch die Arbeitsfläche, an der die erwähnten Massnahmen durchgeführt wurden, vergrösserte sich jährlich im Durchschnitt um etwa 11%, was vor allen Dingen der Unkrautbekämpfung zuzuschreiben ist, da 70% aller Schutzmassnahmen der Unkrautbekämpfung galten.

Darüber hinaus stieg in dem o.g. Betrieb auch der Verbrauch von aktiver Substanz auf 1 ha Ackerfläche und war höher als der Landesdurchschnitt. Auf 1 ha Ackerfläche wurde nämlich pro Jahr durchschnittlich 0,06 kg Aktivsubstanz mehr verbraucht. Für das ganze Land ist der entsprechende Wert zweifach geringer. In dem

Betrieb Magnice wurde in dem erwähnten Zeitabschnitt 0,13 - 2,12 kg Aktivsubstanz auf 1 ha Ackerfläche verbraucht. Das Anstiegstempo betrug hier 0,1 kg Aktivsubstanz pro Jahr. In der Struktur des Pestizidverbrauchs spielten die Herbizide eine ausschlaggebende Rolle. Der Anteil von Insektiziden war hingegen differenziert je nach dem Befall durch wirtschaftlich wichtige Agrophagen. Der Befall durch *Meligethes aeneus* F. betraf 5 - 60% der Knospen, durch *Ceuthorrhynchus napi* Gyll. 30-90% und durch *Ceuthorrhynchus quadridens* Panz. 24 - 98% aller Pflanzen.

Die Beize des Rapssaatgutes ist ein sehr wichtiges Element der Schutzmassnahmen. Es muss jedoch hier unterstrichen werden, dass nicht jeder Produzent entsprechende Bedingungen hat, um die Beize mit eigenen Kräften durchführen zu können.

Die Anbaukosten eines Hektars Raps wurden auf Grund mehrerer Daten bestimmt, die ausser der normalen Rechnungsführung gesammelt wurden. Sie weisen eine steigende Tendenz, nämlich 2 702 zł pro Jahr auf. In dem analysierten Zeitabschnitt lag der Anteil von Pestiziden an der Kostenstruktur zwischen 1% und 8% der Gesamtkosten. Die Tendenzen hierzu werden durch die Gleichung  $y = -0,111x + 5,85$  ausgedrückt, allerdings ist diese Abhängigkeit statistisch nicht signifikant. Der Anteil von Pestiziden an der Kostenstruktur wies also eine wachsende Tendenz auf, obwohl deren Verbrauch deutlich gestiegen ist /Tab. 1/. Dies kann auf eine höhere Preisstabilität chemischer Pflanzenschutzmittel im Vergleich zu anderen Kosten zurückgeführt werden. Wenn man eine einstündige Durchführung einer Pflanzenschutzmassnahme in Betracht zieht, so sind in der Kostenstruk-

tur die Brennstoffkosten besonders auffallend. Der durchschnittliche Anstieg betrug nämlich pro Jahr etwa 2%. Das Tempo des Kostenanstiegs für eine einstündige Durchführung von Schutzmassnahmen betrug 33 z%. In Bezug auf 1 ha Anbaufläche sind es 19 z%. Die Schutzmassnahmen wurden in Betrieb Magnice durch Einzelarbeit ausgeführt.

Im Bereich der Rapsproduktionskosten wiesen die Kosten der chemischen Schutzmassnahmen /Pestizide + Einsatzkosten/ eine grosse Differenziertheit auf und schwankten je nach dem Jahr von 1,5% bis 11,2%. In Bezug auf 1 ha Anbaufläche schwankten die der Unkrautbekämpfung zwischen 947 und 1025 z%, was für ihre hohe Stabilität spricht. Dabei nahmen die Herbizide 59-96% aller Kosten ein. Die meistgebrauchten Herbizide waren Treflan und in den letzten Jahren auch Trifluotox /das Herbizidangebot hat sich in den achtziger Jahren wesentlich vergrössert/. Die Rentabilität der Unkrautbekämpfungsmassnahmen, gemessen nach der Menge des zu schützenden Produktes, die die Schutzkosten decken soll, ist differenziert. Der Rentabilitätsindex verminderte sich jedoch, was für eine hohe Rentabilität der Unkrautbekämpfungsmassnahmen spricht /Tab.3/,/1,4,5/. Die Kosten einer schädlingbekämpfenden Massnahme waren auch differenziert und wiesen eine wachsende Tendenz von 35 z% pro Jahr auf. Der Anteil von Insektiziden an den Schutzkosten war niedriger als derjenige der Herbizide und schwankte zwischen 46 - 92%. Die Massnahmenkosten waren hier durch eine unterschiedliche, zwischen 9 und 94 kg variierende Menge des zu schützenden Produktes rekompensiert, was ebenfalls für die Rentabilität der chemischen Schutzmassnahmen spricht. Zur Schädlingbekämpfung wur-



den die Präparate Bi-58-EC und Gamakorbatox verwendet.

#### Schlussfolgerungen

1. In den Jahren 1970 - 1985 sind die Rapsanbaukosten von 8199 zł auf 43711 zł gestiegen. Der Anteil von Pestiziden an den Produktionskosten schwankte zwischen 1% und 8%. Die hohen, eine jährlich 2%ige Anstiegstendenz aufweisenden Brennstoffkosten sind in Bezug auf 1 Schutzmassnahmenstunde die auffallendsten in der Gesamtkostenstruktur.
2. Im Verbrauch von Pestiziden auf 1 ha Rapsanbaufläche hatten Herbizide einen dominierenden Anteil. Der Pestizidverbrauch auf 1 ha Anbaufläche stieg von 0,3 auf 2,1 kg Aktivsubstanz.
3. Chemische Rapsschutzmassnahmen charakterisieren sich durch eine hohe Rentabilität in Bezug sowohl auf Unkraut- als auch auf Schädlingsbekämpfung. Bei chemischen Schutzmassnahmen wurden die Kosten der Unkrautbekämpfung durch 20 - 143 kg Raps und die der Schädlingsbekämpfung durch 9 - 64 kg Raps kompensiert.

Tabelle 1

## Anbau- und Arbeitsfläsche sowie Pestizidverbrauch im Rapsanbau

| Nr. Aufzählung   | Jahre | 1970/71 | 1972/73 | 1974/75 | 1976/77 | 1978/79 | 1980/81 | 1982/83 | 1983/84 | 1984/85 | 1985/85 |
|--|-------|---------|---------|---------|---------|---------|---------|---------|---------|---------|---------|
| 1. Rapsanteil am Anbauverhältnis in % der Ackerfläsche |       | 10,1    | 7,9     | 9,4     | 14,7    | 11,3    | 12,0    | 7,0     | 13,8    | 7,8     | 13,5    |
| 2. Arbeitsfläsche in % der Rapsanbaufläsche            |       |         |         |         |         |         |         |         |         |         |         |
| a/ Unkräuter   |       | 13,0    | 14,9    | -       | 87,9    | 47,6    | 83,0    | 163,6   | 60,3    | 1100,0  | 1100,0  |
| b/ Schädlinge  |       | 80,0    | 79,4    | 68,6    | 87,9    | 100,0   | -       | 145,4   | 100,0   | 1167,3  | 1150,9  |
| c/ Unkräuter u. Schädlinge                             |       | 93,0    | 94,3    | 68,6    | 175,8   | 147,6   | 83,0    | 309,0   | 160,3   | 1267,3  | 1250,9  |
| 3. Struktur d. Arbeitsfläsche                          |       |         |         |         |         |         |         |         |         |         |         |
| a/ Unkräuter   |       | 13,9    | 15,8    | -       | 50,0    | 32,0    | 100,0   | 53,5    | 35,1    | 37,4    | 40,0    |
| b/ Schädlinge  |       | 86,1    | 84,2    | 100,0   | 50,0    | 78,0    | -       | 46,2    | 64,2    | 62,6    | 60,0    |
| 4. Aktivsubstanz auf 1 ha Rapsanbau /kg/               |       | 0,39    | 0,41    | 0,3     | 1,4     | 1,07    | 0,83    | 1,66    | 1,65    | 1,59    | 2,12    |
| darunter:  |       |         |         |         |         |         |         |         |         |         |         |
| a/ Herbizide   |       | 0,13    | 0,15    | -       | 0,91    | 0,49    | 0,83    | 1,15    | 0,62    | 1,05    | 1,04    |
| b/ Insektizide   |       | 0,26    | 0,26    | 0,2     | 0,45    | 0,52    | -       | 0,40    | 1,00    | 0,54    | 1,08    |
| c/ Fungizide   |       | -       | -       | 0,1     | 0,04    | 0,06    | -       | 0,11    | 0,03    | -       | -       |
| 5. Aktivsubstanz auf 1 ha Ackerfläsche in Magnico      |       | 0,97    | 0,94    | 0,81    | 1,67    | 2,20    | 1,22    | 1,81    | 2,08    | 1,73    | 2,10    |
| 6. Aktivsubstanz auf 1 ha Ackerfläsche in Polen /kg/   |       | 0,39    | 0,31    | 0,64    | 0,58    | 0,51    | 0,49    | 0,86    | 0,77    | 0,76    | -       |

Tabelle 2  
Anbaukosten von 1 ha Raps und Kosten einständiger Durchführung von Schutzmaßnahmen

| Inr  | Aufzählung  | 1970/71 | 1972/73 | 1974/75 | 1976/77 | 1978/79 | 1980/81 | 1982/83 | 1983/84 | 1984/85 | 1985/86 |
|------|---|---------|---------|---------|---------|---------|---------|---------|---------|---------|---------|
| I.   | Struktur d. angewählten Kosten /%                               |         |         |         |         |         |         |         |         |         |         |
| 1.   | Arbeitskosten   | 8,2     | 7,2     | 4,5     | 6,0     | 5,8     | 6,9     | 8,1     | 6,2     | 7,3     | 5,5     |
| 2.   | Kosten d. Mineraldüngers  | 22,2    | 21,3    | 23,9    | 18,6    | 19,6    | 16,8    | 21,9    | 17,2    | 14,9    | 13,4    |
| 3.   | Kosten d. Pflanzenschutzmittel                                  | 2,9     | 5,3     | 1,2     | 7,7     | 8,3     | 5,3     | 5,4     | 2,1     | 3,9     | 2,8     |
| 4.   | Gesamtkosten zI/ha  | 8199    | 9959    | 9172    | 111 678 | 113 124 | 13 658  | 128 859 | 137 824 | 139 794 | 143 711 |
| 5.   | Ertrag dt/ha  | 22,9    | 26,9    | 17,0    | 28,1    | 31,2    | 23,5    | 29,2    | 29,4    | 26,9    | 31,3    |
| 6.   | Produktionskosten von 1 dt /zI/                                 | 345     | 353     | 522     | 403     | 404     | 556     | 947     | 1 210   | 1 410   | 1 329   |
| III. | Kostenstruktur einständiger Durchführung v. Schutzmaßnahmen /%: |         |         |         |         |         |         |         |         |         |         |
| 1.   | Arbeitskosten   | 26,03   | 19,14   | 15,42   | 22,29   | 20,8    | 19,84   | 38,88   | 24,2    | 23,9    | 24,2    |
| 2.   | Kosten d. Schleppereinsatzes                                    | 48,52   | 63,80   | 51,58   | 41,22   | 28,40   | 22,03   | 8,90    | 29,30   | 20,70   | 19,50   |
| 3.   | Kosten d. Spritzeneinsatzes                                     | 3,58    | 2,96    | 3,58    | 3,50    | 4,38    | 2,68    | 2,87    | 5,00    | 12,10   | 12,50   |
| 4.   | Brennstoff  | 16,66   | 10,27   | 26,18   | 28,76   | 42,39   | 52,28   | 42,75   | 32,30   | 33,10   | 35,70   |
| 5.   | Indirekte Kosten  | 5,21    | 3,93    | 3,24    | 4,23    | 3,53    | 4,17    | 6,61    | 9,20    | 10,20   | 10,10   |
| 6.   | Gesamtkosten zI/ha  | 43,80   | 71,05   | 86,23   | 80,31   | 92,32   | 157,80  | 208,40  | 480,00  | 569,00  | 661,00  |
| 7.   | Arbeitsleistung ha/h  | 1,16    | 0,93    | 1,81    | 1,09    | 1,62    | 1,06    | 1,60    | 1,58    | 1,62    | 1,60    |
| 8.   | Kost von 1 ha /zI/  | 37,80   | 76,40   | 48,80   | 73,70   | 57,00   | 148,90  | 130,30  | 303,80  | 351,20  | 413,10  |

Tabella 3

Kosten des chemischen Pflanzschutzes

| Nr | Auszahlung   | Jahre | 1970/71 | 1972/73 | 1974/75 | 1976/77 | 1978/79 | 1980/81 | 1982/83 | 1983/84 | 1984/85 | 1985/86 |
|----|--|-------|---------|---------|---------|---------|---------|---------|---------|---------|---------|---------|
| 1. | Pestizidanteil an Wirtschaftskosten                          |       | 0,83    | 0,97    | 0,81    | 1,62    | 3,26    | 2,29    | 1,53    | 0,95    | 1,12    | 1,20    |
| 2. | Pestizidanteil an Kosten d. Pflanzproduktion                 |       | 2,10    | 2,90    | 1,20    | 7,70    | 8,30    | 5,30    | 5,40    | 2,10    | 3,90    | 2,80    |
| 3. | Anteil d. Massnahmenkosten an Kosten Pflanzproduktion / %    |       | 2,90    | 3,70    | 1,50    | 8,70    | 9,30    | 6,20    | 6,80    | 3,40    | 6,30    | 5,20    |
| 4. | Kosten einer unkrautbekämpfenden Massnahme                   |       | 947     | 984     | -       | 982     | 11 025  | 11 053  | 968     | 952     | 1 012   | 1 017   |
|    | - % der Herbizide  |       | 96,0    | 92,0    | -       | 93,0    | 94,0    | 86,0    | 87,0    | 68,0    | 65,0    | 59,0    |
|    | - Wo x/  |       | 1,28    | 1,37    | -       | 0,96    | 0,98    | 0,89    | 0,39    | 0,20    | 0,23    | 0,23    |
| 5. | Kosten einer Schädlingsbekämpfenden Massnahme                |       | 163     | 201     | 98      | 183     | 672     | -       | 211     | 700     | 889     | 814     |
|    | - % der Herbizide  |       | 76,0    | 62,0    | 50,0    | 60,0    | 92,0    | -       | 46,0    | 57,0    | 61,0    | 49,0    |
|    | - Wo   |       | 0,22    | 0,28    | 0,11    | 0,16    | 0,61    | -       | 0,09    | 0,15    | 0,21    | 0,19    |
| 6. | Durchschnittl. Kosten der Durchführung einer Schutzmassnahme |       | 225     | 307     | 140     | 1028    | 1179    | 846     | 1973    | 1285    | 2497    | 2252    |
|    | - Wo   |       | 0,32    | 0,42    | 0,16    | 1,01    | 1,12    | 0,72    | 0,81    | 0,28    | 0,59    | 0,52    |

x/ Wo = der ungefähre Restabilitätsindex einer Massnahme / 5/

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