Research on the use of different insecticides for the control of the green peach aphid (Myzus persicae) (vector of beet mild yellowing virus (BMYV)) in the Netherlands in 2012

12P08

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CONTENTS

Summary

Virus yellows is an economical important disease in sugar beet. It is caused by the viruses BYV or BMYV. In diagnostic samples of recent years, only beet mild yellowing virus (BMYV) is detected at IRS. This virus is mainly transmitted by the green peach aphid (*Myzus persicae*). The spread of the virus in a sugar beet field can be controlled by controlling this aphid species by means of insecticides. This is done since many years. The insecticides can be applied on the seed pellet, in granular form or by spraying. In order to investigate new insecticides, research was done to control the spread of the virus by controlling the vector. Different methods were compared with a treatment without insecticide.
Therefore field trials were conducted in Westmaas and Colijnsplaat. In these trials green peach aphids, infected with BMYV were inoculated nine weeks after sowing (end of May). It is important to realise that this resulted in a higher infection level in comparison with a natural situation. Normally, less than 1% of the green peach aphids is infested with virus, in the field trials it was 100%.
There was no effect of the insecticides on the number of aphids one week after inoculation. This was due to the very bad weather conditions. Therefore, the number of aphids was too low to see any effect.
On 25 July the number of plants with virus yellow symptoms was observed. All insecticides had a better effect on the control of BMYV than the untreated control. The seed treatments (IRS 658, IRS 663, IRS 664 and IRS 711) gave the best results. Only the seed treatment IRS 711 controlled BMYV better than the granular (IRS 709) and the spraying (IRS 721) treatment. These results show that time of spraying of insecticides is a crucial factor in controlling aphids.
There was no effect of insecticide treatments on sugar yield or financial yield. However, there was an effect on sugar content. The seed treatments (IRS 658, IRS 663, IRS 664 and IRS 711) resulted in the highest sugar content. Only the seed treatments resulted in a higher sugar content compared to the untreated control. The higher the number of plants with virus yellows symptoms, the lower the percentage of sugar and sugar yield.

Samenvatting

Vergelingsziektevirus wordt overgebracht door de groene perzikbladluis (*Myzus persicae*). Er zijn twee soorten vergelingsziektevirussen: BYV en BMYV. De laatste jaren wordt bij de afdeling Diagnostiek van IRS alleen maar BMYV aangetroffen. Het virus kan worden aangepakt door de bladluizen te bestrijden. Doel van dit onderzoek is de effectiviteit bepalen van verschillende soorten insecticiden op pillenzaad, als granulaat en als bespuiting voor de bestrijding van de groene perzikbladluis en daarmee ook vergelingsvirus.
Daarvoor werden in Westmaas en Colijnsplaat proefvelden aangelegd, waarbij groene perzikbladluizen, die vooraf geïnfecteerd waren met het virus, ruim negen weken na zaai (eind mei) werden uitgezet. Hierbij is het wel belangrijk te realiseren dat dit tot een veel hogere druk heeft geleid, dan die we normaal in het veld tegenkomen. Normaal is maximaal 1% van de bladluizen besmet met virus, terwijl dat nu 100% was.
Eén week na infecteren is het aantal bladluizen geteld. Door het slechte weer, waren er toen nog te weinig aanwezig om enig effect van de insecticiden te kunnen meten. Op 25 juli is het percentage planten met vergelingsziekte geteld. Bij de veldjes met insecticiden was dit lager dan bij de veldjes zonder insecticiden. De beste bestrijding werd behaald met de zaadbehandelingen (IRS 658, IRS 663, IRS 664 en IRS 711). IRS 711 had minder planten met vergelingsziekte dan de bespuiting met een insecticide twee dagen na infectie (IRS 721). Dit laat zien dat de timing bij een bespuiting ontzettend belangrijk is. Bij de zaadbehandelingen zijn de bladluizen direct gedood, waardoor de verspreiding van vergelingsziekte beperkt is gebleven ten opzichte van de bespuiting.
De zaadbehandelingen hadden tevens een hoger suikergehalte in vergelijking met de behandeling zonder insecticiden. Hoe hoger het aantal planten met vergelingsziekte, hoe lager het suikergehalte en de suikeropbrengst.

1. Introduction

Virus yellows is an economical important disease in sugar beet. In diagnostic samples of recent years, only beet mild yellowing virus (BMYV) is detected at IRS. This virus is mainly transmitted by the green peach aphid (*Myzus persicae*). The spread of the virus in a sugar beet field can be controlled by controlling this aphid species by means of insecticides. This is done since many years. The insecticides can be applied on the seed pellet, in granular form or by spraying. In order to investigate new insecticides, research was done to control the spread of the virus by controlling the vector. Different methods were compared with a treatment without insecticide.

2. Materials and methods

2.1 Trial sites

Two locations were selected for these trials: one field in Westmaas and one in Colijnsplaat.

2.2 List of products

In table 1 the products used in the trials have been listed. All seed treatments (also the untreated controls) were treated with the fungicides Proseed (6.5 g thiram) and Tachigaren (14 g hymexazol) to prevent influences of fungi on plant establishment.

Table 1. Products used.

treatment	product	way of applying
1	untreated control	-
2	IRS 658	seed treatment
3	IRS 711	seed treatment
4	IRS 663	seed treatment
5	IRS 664	seed treatment
6	IRS 709	granular
7	IRS 721	spraying

2.3 Drilling

Drilling was done with a standard precision sowing machine (Monozentra). This sowing machine is equipped with a system to change seed batches quickly. The granular was applied in the seed furrow at sowing. Sowing distance within the rows in both trials was 18.5 cm and 50 cm between rows. The field trials of Westmaas and Colijnsplaat were sown on 22 and 26 March 2012, respectively. All trials were designed as randomised blocks in four replications. Gross plot size: six rows and 21 meters long. Nett plot size: six rows of 14.5 meters long.

2.4 Inoculation with aphids

In autumn 2011 sugar beets with BMYV were taken from a naturally infested field in Zonnemaire, defoliated and stored in a refrigerator at 6^{o}C. In February 2012, these sugar beets were potted in a mixture of sand and potting soil, watered and placed in the laboratory at room temperature to stimulate leaf growth. Meanwhile sugar beets were sown in the climate chambers (23^{o}C for sixteen hours in light (20,000 lux) and 16^{o}C for eight hours in dark). About 100 plants were sown. About four weeks before inoculation of the plants in the field, green peach aphids were obtained from Koppert Biological Systems in Berkel en Rodenrijs

(the Netherlands), where they were cultured on sweet pepper. They were transferred to the naturally infested sugar beet plants for one day. Leaves with aphids were cut off and transferred to the young sugar beet plants (8-10th leaf stage). Just before transferring, the plants were taken out of the climate room and transferred to the laboratory at room temperature. The aphids, bearing the virus, were multiplied on those plants.
For field inoculation, leaves with aphids from these plants were cut off and carefully transported to the field trials. It was planned to inoculate the plants about seven to eight weeks after sowing. Due to cold and wet circumstances in spring, the fields were inoculated later. The field trial in Westmaas was inoculated on 29 May (nine weeks and five days after sowing - BBCH 18-20 [1]) and the field in Colijnsplaat on 30 May (nine weeks and two days after sowing - BBCH 18). In Westmaas every tenth plant of the four central rows of each plot was inoculated with two to four aphids per plant. In Colijnsplaat seven plants of the four central rows of each plot were inoculated.

2.5 Spraying

Two days after inoculation treatment 7 (IRS 721) was sprayed. One week after inoculation the whole field trial (including the surrounding sugar beet field) was sprayed with Pirimor (pirimicarb, 0.4 kg/ha) to prevent spread of infected aphids.

2.6 Assessment of efficacy

The effect of the different treatments and formulations on green peach aphids was measured by counting the number of aphids on five randomly chosen infected plants per plot. This was done one week after inoculation, just before the field was sprayed with Pirimor. The effect on BMYV was measured by counting the number of plants with yellowing symptoms per plot eight (25 July) and sixteen weeks (19 September) after inoculation. This was done in all rows. In addition, the field trial in Westmaas was harvested in order to measure the effect on yield. This was done on 11 November. The gross weight of the plot was measured and of each plot three subsamples were taken to the tare house of the IRS for analysis of sugar beet quality.

2.7 Analysis of data

Data were analysed by using Analysis of Variance (ANOVA). Analyses were done with Genstat Software Package 15.0.

3. Results and discussion

3.1 Effect on aphids

At both field trials green peach aphids and black bean aphids (*Aphis fabae*) were observed. The black been aphids were not inoculated, but were naturally occurring aphids. There was no interaction between field trial location and treatments on the number of green peach aphids (Probability (P) = 0.154) and on the black bean aphids (P = 0.403). There was also no effect of treatment on both numbers of green peach aphids (P = 0.074) and on black bean aphids (P = 0.142). Data are shown in Annexes B and C. No conclusions can be made, since the number of aphids was very low.

3.2 Effect on beet mild yellowing virus (BMYV)

The effect on beet mild yellowing virus (BMYV) was measured on 25 July and 19 September. On 25 July there was no interaction between treatment and location (P = 0.451). There was a significant difference between the treatments (P<0.001; Figure 1; Annexes B and C). All insecticide treatments had a significantly lower percentage of plants with virus yellows in comparison with the untreated control. There was no significant difference between the seed treatments (IRS 658, IRS 663, IRS 664 and IRS 711). Only the seed treatment IRS 711 had a significantly lower percentages of plants with virus yellows in comparison with the granular (IRS 709) and the spray application (IRS 721).

For the seed treatments it was expected that the insecticides work for about ten weeks after sowing. On the other hand, for the granular (IRS 709) it was expected that it would only work for about eight weeks. The granular might have worked better if the infection with aphids was done earlier. Moreover, the spray application (IRS 721) might have worked better if spraying was done shortly after (at the same day) or one day after infection. This might indicate that for a good effect of a spray application of insecticides, the timing of application is a crucial factor. By using seed treatments, the grower will not face this problem.

It is important to keep in mind that the percentage of plants with virus yellows will hardly ever have been so high as in the field trials, since in practice only a small percentage (less than 1% [2]) of the aphids will carry BMYV.

Data of the measurement on 19 September are only shown in Annexes B and C, since these data do not give a good indication of the direct effect of the different insecticide treatments. The percentage of plants with virus yellows has increased enormously between 25 July and 19 September and this might be due to a secondary infection.

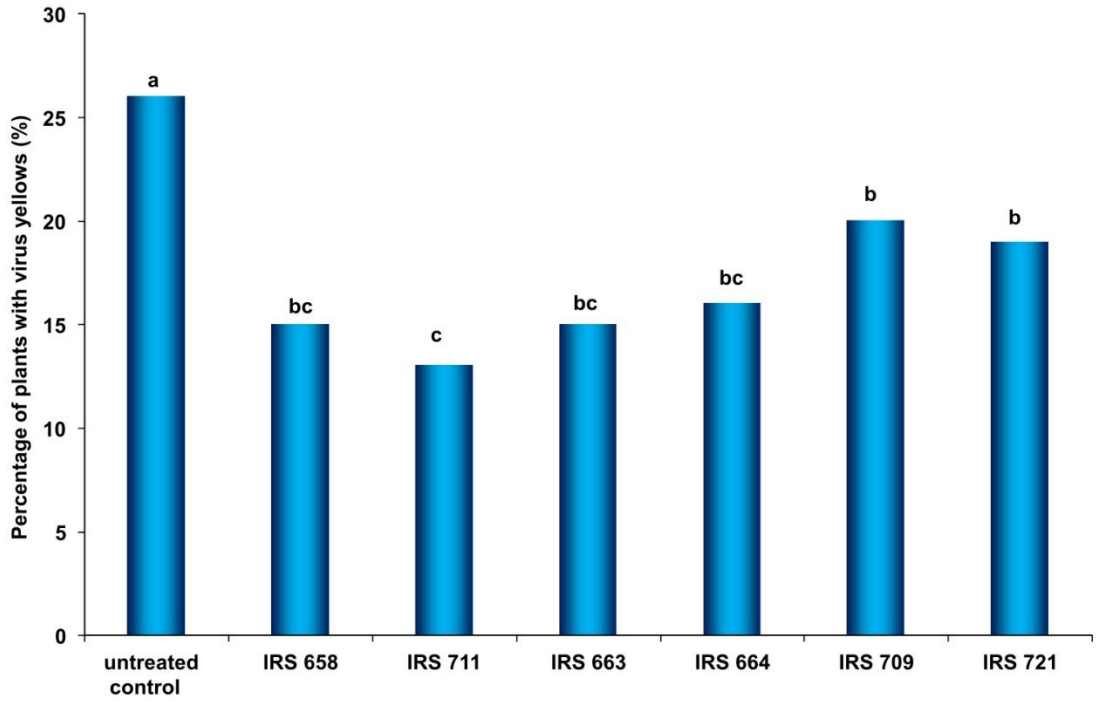


Figure 1. Mean percentage of plants with virus yellows (BMYV) of the field trials in Westmaas and Colijnsplaat (25 July, 2012) (LSD[1] 5% = 5.4; P<0.001). There was no interaction between treatment and location (P = 0.451). IRS 658, IRS 711, IRS 663 and IRS 664 were seed treatments. IRS 709 was the granular and IRS 721 the spray application of insecticides.

[1] LSD = least significant difference.

3.3 Effect on yield

There was no effect of treatment on sugar yield (P=0.06) or financial yield (P=0.18), only on sugar content (P=0.01) (see Annex B). The seed treatments (IRS 658, IRS 663, IRS 664 and IRS 711) had significantly the highest sugar content (figure 2). Only the sugar contents of the seed treatments were significantly higher than the untreated control. There was a negative relation between the percentage of plants with virus yellows on 25 July and the sugar content (R^2=0.58; figure 3) and sugar yield (R^2=0.69; figure 4). The higher number of plants with virus yellows, the lower the percentage of sugar and sugar yield.
It might be due to the very high infection pressure that no effect on sugar and financial yield could be detected.

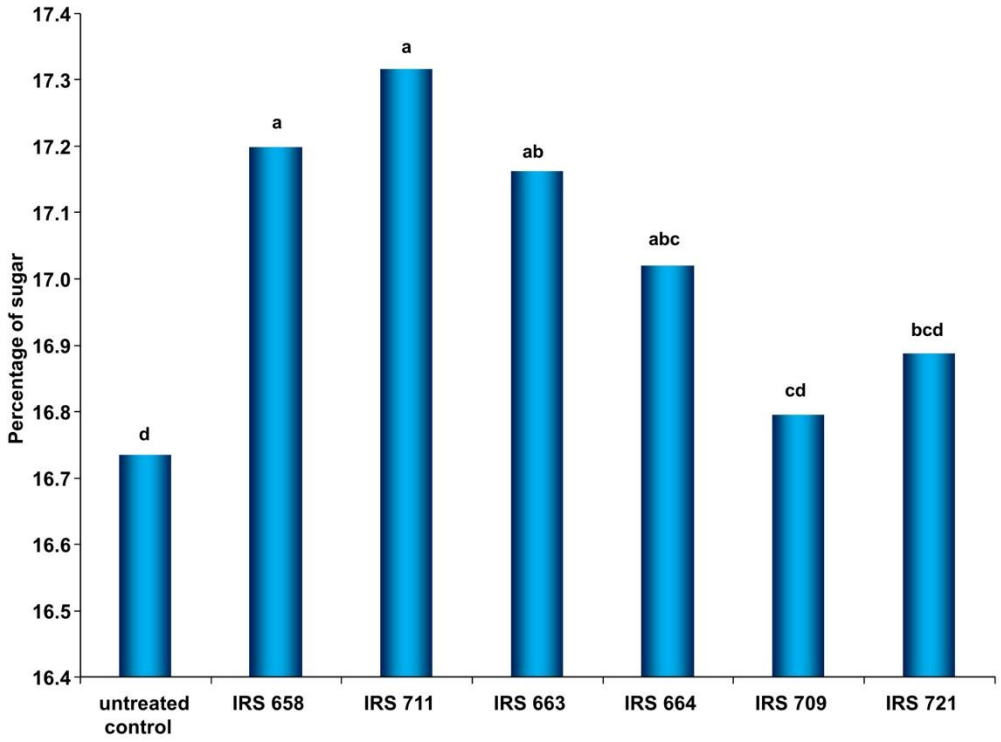


Figure 2. Mean sugar content of different treatments in the field trial in Westmaas (LSD[1] 5% = 0.314; P[2]=0.01).

[1] LSD = least significant difference.
[2] P = probability.

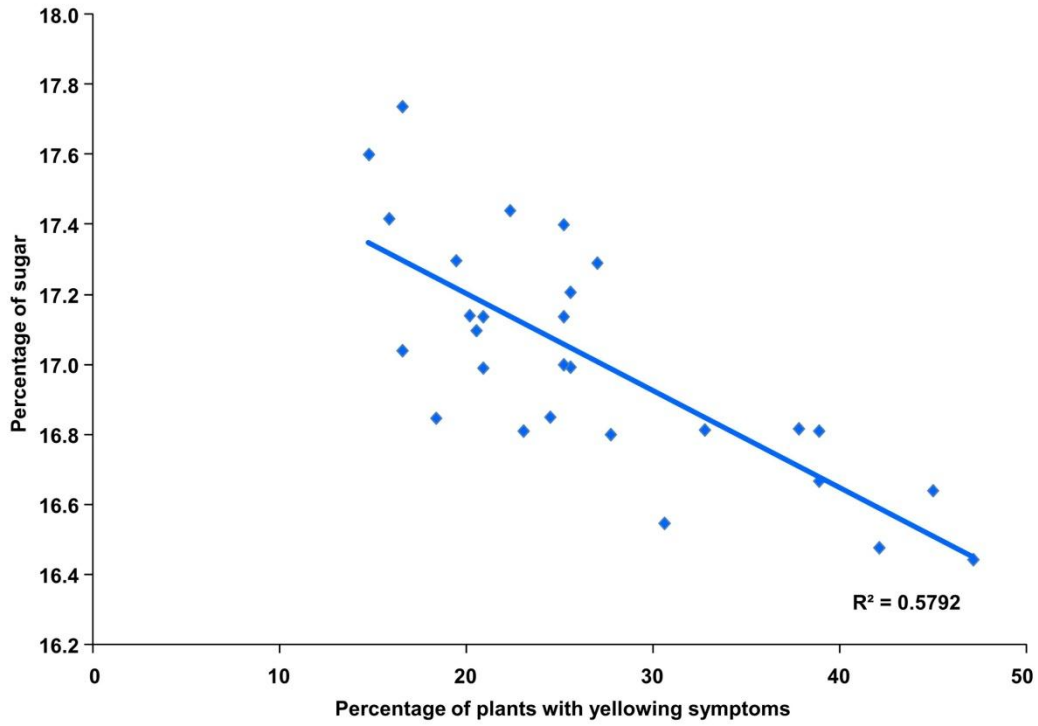


Figure 3. Relation between percentage of plants with symptoms of virus yellows (25 July 2012) and sugar content at harvest (9 November 2012) at the field trial in Westmaas.

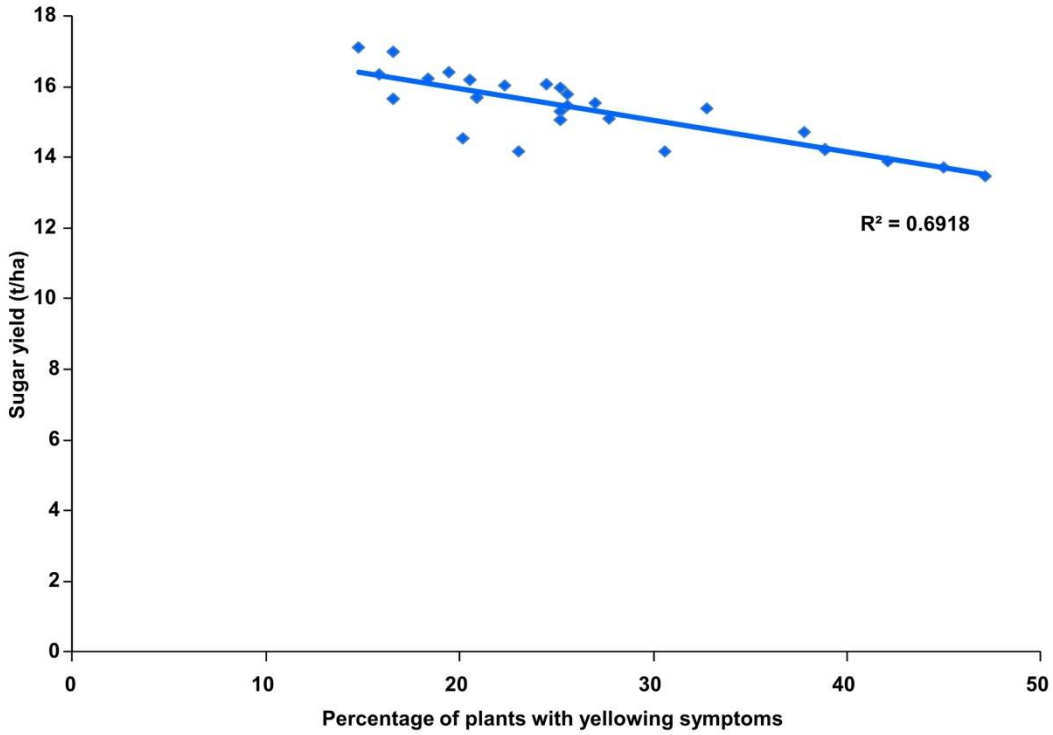


Figure 4. Relation between percentage of plants with virus yellows symptoms (25 July 2012) and sugar yield at harvest (t/ha; 9 November 2012) at the field trial in Westmaas.

4. Conclusions

The aim was to study the efficacy of different insecticides on the control of beet mild yellowing virus (BMYV) and its vector, the green peach aphid (*Myzus persicae*).
There was no effect of the insecticides on the number of aphids. This implies that only a few aphids can do a lot of damage.
All insecticides had a better effect on the control of BMYV than the untreated control. The seed treatments (IRS 658, IRS 663, IRS 664 and IRS 711) gave the best results. Only the seed treatment IRS 711 controlled BMYV better than the granular (IRS 709) and the spraying (IRS 721) treatment. These results show that time of spraying of insecticides is a crucial factor in controlling aphids.
There was no effect of insecticide treatments on sugar yield or financial yield. However, there was an effect on sugar content. The seed treatments (IRS 658, IRS 663, IRS 664 and IRS 711) resulted in the highest sugar content. Only the seed treatments resulted in a higher sugar content compared to the untreated control. The higher the number of plants with virus yellows symptoms, the lower the percentage of sugar and sugar yield.

5. Literature

1. Meier U. (2001).
   Growth stages of mono- and dicotyledonous plants.
   BBCH Monograph, 2.
2. Stevens M, H.G. Smith, P.B. Hallsworth, L.A Haylock & A.M. Dewar (1994).
   Detection of viruses and insecticide resistance in sugar beet aphids caught in suction traps.
   Crop Protection Conference, Pest and Diseases; Brighton: 917-922.

Annex A GEP CERTIFICATE IRS

Ministerie van Landbouw, Natuur en Voedselkwaliteit

landbouw, natuur en voedselkwaliteit

This is to declare that, in conformity with the request of March 4, 2008

Stichting IRS

Residing Van Konijnenburgweg 24, Bergen op Zoom, the Netherlands

HAS OFFICIALLY BEEN RECOGNISED AS AN ORGANISATION FOR EFFICACY TESTING

as has been laid down in the 'Regeling gewasbeschermingsmiddelen en biociden' (Regulation Crop Protection Products and Biocides) of September 26, 2007 (Staatscourant 2007, 386)

This recognition will commence on June 19, 2008 and expire on June 19, 2014

Wageningen, June 9, 2008

For the Minister of Agriculture, Nature and Food Quality,

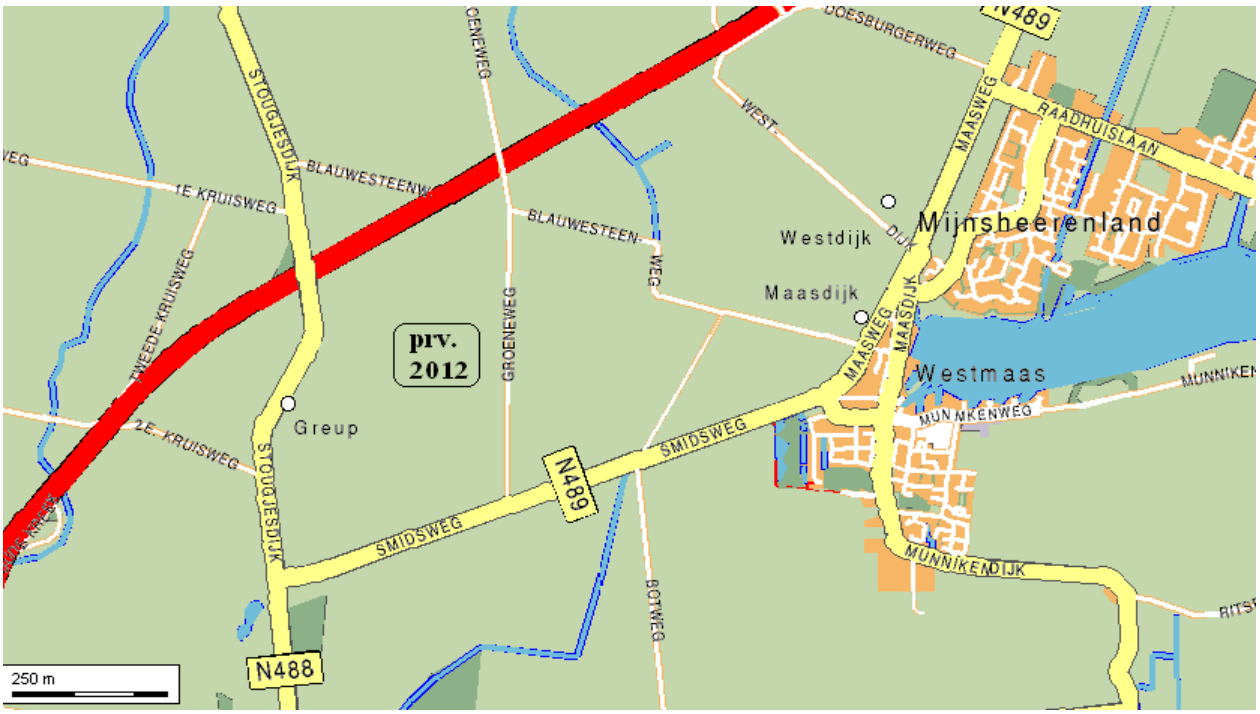
Mr. ing. H.A. Harmsma

Acting Director Plant Protection Service

Annexes B Field trial Westmaas

Annex B1 Location

GPS location:
51.78923, 4.44742
N 51 47.354, E 4 26.845
N 51 47 21.2, E 4 26 50.7



Annex B2 Trial scheme

Trial field:	Westmaas		
Number of replications:	4		
Nett size (m):	14.5×3	Gross size (m):	21×3

D							
1	**2**		**3**	**4**	**7**	**6**	**5**
			extra brede bruto				
6	**4**		**7**	**1**	**5**	**2**	**3**
2	**3**		**5**	**6**	**4**	**7**	**1**
5	**1**	**sp.sp**	**4**	**7**	**2**	**3**	**6**
A							

Annex B3 Treatments

Trial field code: IRS 12-03-01.03
Name of trial field: Beet mild yellowing virus (BMYV), Westmaas
Number of replications: 4
Number of treatments: 7

number	treatment
1	untreated control
2	IRS 658
3	IRS 711
4	IRS 663
5	IRS 664
6	IRS 709
7	IRS 721

Annex B4 General data

soil type:	clay soil	2.5% organic matter
	pH-KCl = 7.3	parts < 16 µm = 34%
	K-value = 24	Pw = 35 mg P_2O_5 per l of soil
	$CaCO_3$ = 9%	
preceding crop:	2011 wheat	
	2010 barley + English ryegrass	
	2009 wheat	
	2008 potatoes / Brussels sprouts	
	2007 barley	
	2006 fallow / potatoes / onions	
drilling date:	22 March 2012	
variety:	Bever	
distance in row:	18.5 cm	
distance between rows:	50 cm	

Annex B5 Efficacy

Trial field code: IRS 12-03-01.03
Name of trial field: Beet mild yellowing virus (BMYV), Westmaas

Assessment Number of green peach aphids/plant
Date of assessment 5 June 2012

number	treatment	A	B	C	D	average
1	untreated control	0	0.4	1.0	0.2	0.4
2	IRS 658	0	0	0.2	0.6	0.2
3	IRS 711	0	0	0	0	0.0
4	IRS 663	0	0	0	0	0.0
5	IRS 664	0.4	0	0	0	0.1
6	IRS 709	0	0	0.4	0	0.1
7	IRS 721	0.2	0.4	0.8	0.2	0.4
	LSD[1] 5%					0.3492
	P[2]					0.104
	significance[3]					NS

Assessment Number of black bean aphids/plant
Date of assessment 5 June 2012

number	treatment	A	B	C	D	average
1	untreated control	0.6	1.6	0	0.2	0.6
2	IRS 658	0	0.2	0	0	0.1
3	IRS 711	0	0	0	0	0.0
4	IRS 663	0	0	0	0	0.0
5	IRS 664	0	0	0.2	0	0.1
6	IRS 709	2.0	2.4	0	0	1.1
7	IRS 721	0.4	0	0.2	0.8	0.4
	LSD[1] 5%					0.823
	P[2]					0.085
	significance[3]					NS

[1] LSD = least significant difference.
[2] P = probability.
[3] NS = not significant.

Assessment Percentage of plants with virus yellows
Date of assessment 25 July 2012

number	treatment	A	B	C	D	average
1	untreated control	38	45	39	33	39
2	IRS 658	15	24	25	26	22
3	IRS 711	21	21	22	17	20
4	IRS 663	20	28	16	27	23
5	IRS 664	26	31	25	21	26
6	IRS 709	17	42	18	39	29
7	IRS 721	23	47	25	19	29
	LSD[1] 5%					9.9
	P[2]					0.02
	significance[3]					S

Assessment Percentage of plants with virus yellows
Date of assessment 19 September 2012

number	treatment	A	B	C	D	average
1	untreated control	98.9	99.3	98.1	95.5	98.0
2	IRS 658	87.9	93.9	88.7	92.2	90.6
3	IRS 711	87.6	91.9	89.9	89.5	89.7
4	IRS 663	93.4	94.4	89.5	93.0	92.6
5	IRS 664	92.9	93.5	88.7	92.5	91.9
6	IRS 709	93.0	97.2	92.9	95.1	94.5
7	IRS 721	95.1	97.2	91.7	91.8	94.0
	LSD[1] 5%					2.4
	P[2]					<0.001
	significance[3]					VS

[1] LSD = least significant difference.
[2] P = probability.
[3] S = significant, VS = very significant.

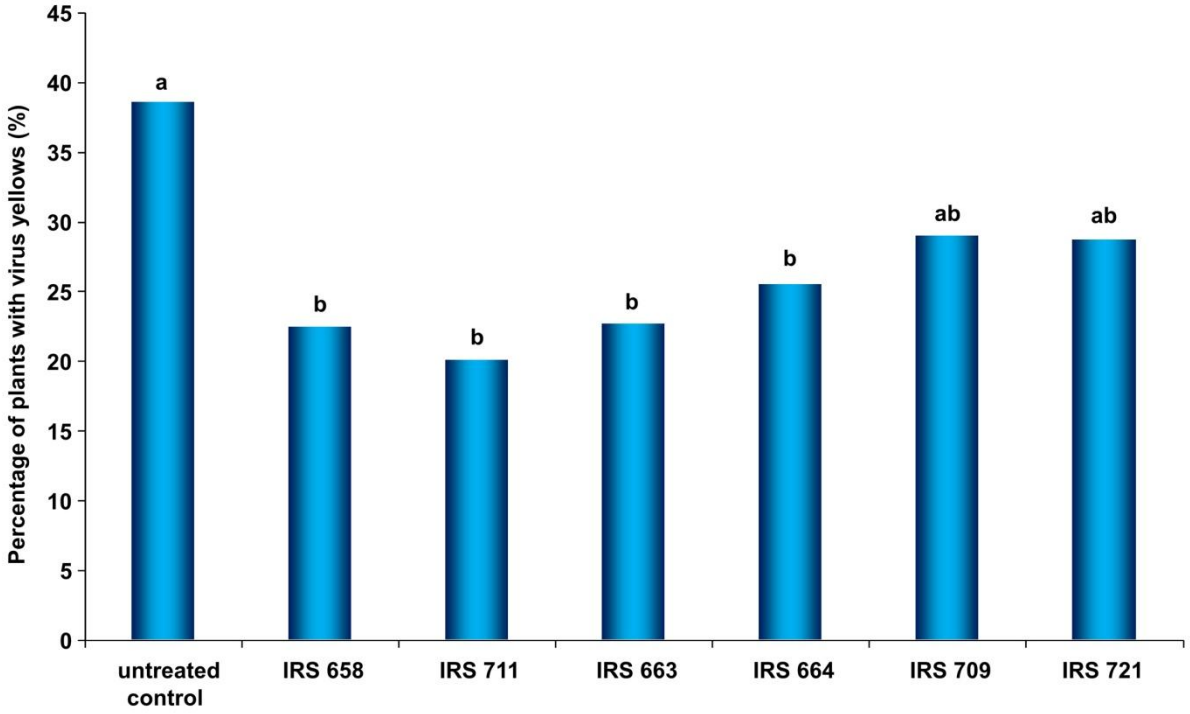


Figure. Mean percentage of plants with virus yellows (BMYV) of the field trial in Westmaas (25 July 2012) (LSD[1] 5% = 9.9; P[2]=0.020).

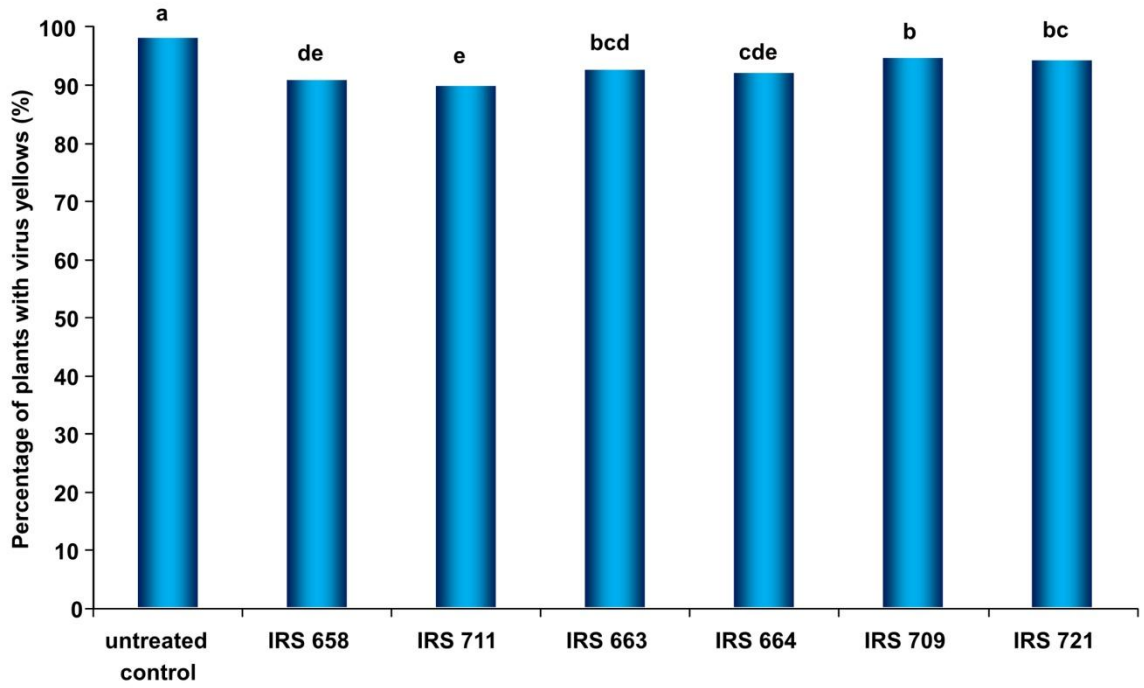


Figure. Mean percentage of plants with virus yellows (BMYV) of the field trial in Westmaas (19 September 2012) (LSD[1] 5% = 2.4; P[2]<0.001).

[1] LSD = least significant difference.
[2] P = probability.

Yield
12-03-01.03: Westmaas

Table. Yield data

treatment	root yield (t/ha)	sugar content (%)	sugar yield (t/ha)	soil tare (%)	plants (%)	financial yield (€/ha)
untreated control	86.8	16.73	14.5	15.5	86.2	3,237
IRS 658	93.5	17.20	16.1	16.5	88.4	3,622
IRS 711	93.8	17.32	16.2	16.3	89.2	3,683
IRS 663	89.7	17.16	15.4	18.4	87.2	3,424
IRS 664	90.1	17.02	15.3	17.2	87.3	3,428
IRS 709	89.4	16.79	15.0	12.8	89.2	3,395
IRS 721	87.5	16.89	14.8	17.1	85.6	3,294
LSD[1] 5%	6.1	0.31	1.2	7.0	3.6	373
P[2]	0.17	0.01	0.06	0.74	0.29	0.18
significance[3]	NS	VS	NS	NS	NS	NS

[1] LSD = least significant difference.
[2] P = probability.
[3] NS = not significant.

Annexes C Field trial Colijnsplaat

Annex C1 Location

GPS location:
51.59257, 3.86759
N 51 35.554, E 3 52.055
N 51 35 33.3, E 3 52 3.3

Annex C2 Trial scheme

Trial field:	Colijnsplaat		
Number of replications:	4		
Nett size (m):	14.5×3	Gross size (m):	21×3

D						
1	**6**	**4**	**2**	**3**	**7**	**5**
7	**3**	**5**	**1**	**4**	**2**	**6**
6	**4**	**2**	**3**	**5**	**1**	**7**
5	**1**	**6**	**7**	**2**	**3**	**4**
A						

Annex C3 Treatments

Trial field code: IRS 12-03-01.04
Name of trial field: Beet mild yellowing virus (BMYV), Colijnsplaat
Number of replications: 4
Number of treatments: 7

number	treatment
1	untreated control
2	IRS 658
3	IRS 711
4	IRS 663
5	IRS 664
6	IRS 709
7	IRS 721

Annex C4 General data

soil type:	clay soil pH-KCl = 7.3 K-value = 29 $CaCO_3$ = 7.0%	1.8% organic matter parts <16 µm = 36% Pw = 34 mg P_2O_5 per l of soil
preceding crop:	2011 flax 2010 potatoes 2009 maize 2008 grass 2007 grass 2006 grass	
drilling date:	26 March 2012	
variety:	Bever	
distance in row:	18.5 cm	
distance between rows:	50 cm	

Annex C5 Efficacy

Trial field code: IRS 12-03-01.04
Name of trial field: Beet mild yellowing virus (BMYV), Colijnsplaat

Assessment Number of green peach aphids/plant
Date of assessment 5 June 2012

number	treatment	A	B	C	D	average
1	untreated control	0.2	0	0	0.4	0.2
2	IRS 658	0	0	0.2	0.6	0.2
3	IRS 711	0	0	0	0	0.0
4	IRS 663	0	0	0	0	0.0
5	IRS 664	0	0	0	0.2	0.1
6	IRS 709	0.2	0.2	0	0.8	0.3
7	IRS 721	0	0	0	0	0.0
	LSD[1] 5%					0.2254
	P[2]					0.065
	significance[3]					NS

Assessment Number of black bean aphids per plant
Date of assessment 5 June 2012

number	treatment	A	B	C	D	average
1	untreated control	9.0	3.6	0.2	1.0	3.5
2	IRS 658	2.4	14.8	0.2	0.4	4.5
3	IRS 711	1.0	0.2	0.0	0.4	0.4
4	IRS 663	0.2	0.6	0.0	0.8	0.4
5	IRS 664	0.2	0.0	2.0	2.6	1.2
6	IRS 709	13.4	9.6	0.0	0.0	5.8
7	IRS 721	9.2	10.2	1.8	1.4	5.7
	LSD[1] 5%					5.636
	P[2]					0.216
	significance[3]					NS

[1] LSD = least significant difference.
[2] P = probability.
[3] NS = not significant.

Assessment Percentage of plants with virus yellows
Date of assessment 25 July 2012

number	treatment	A	B	C	D	average
1	untreated control	8	15	13	18	14
2	IRS 658	6	5	10	9	8
3	IRS 711	2	9	6	8	6
4	IRS 663	3	6	9	7	6
5	IRS 664	3	8	8	5	6
6	IRS 709	9	9	13	12	11
7	IRS 721	9	10	10	7	9
	LSD[1] 5%					3.2
	P[2]					<0.001
	significance[3]					VS

Assessment Percentage of plants with virus yellows
Date of assessment 19 September 2012

number	treatment	A	B	C	D	average
1	untreated control	31	37	32	38	35
2	IRS 658	17	14	28	15	19
3	IRS 711	17	16	21	18	18
4	IRS 663	16	19	23	15	18
5	IRS 664	16	18	23	17	19
6	IRS 709	28	36	41	30	34
7	IRS 721	38	31	29	22	30
	LSD[1] 5%					6.3
	P[2]					<0.001
	significance[3]					VS

[1] LSD = least significant difference.
[2] P = probability.
[3] VS = very significant.

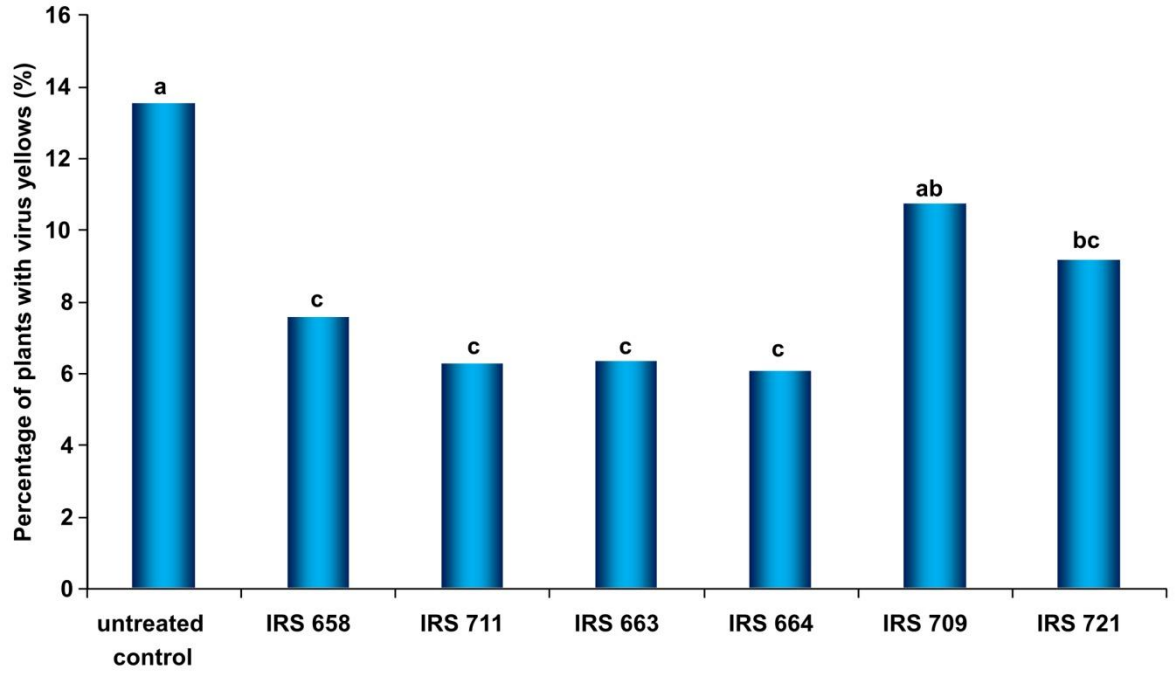


Figure. Mean percentage of plants with virus yellows (BMYV) of the field trial in Colijnsplaat (25 July 2012) (LSD[1] 5% = 3.2; P[2]<0.001).

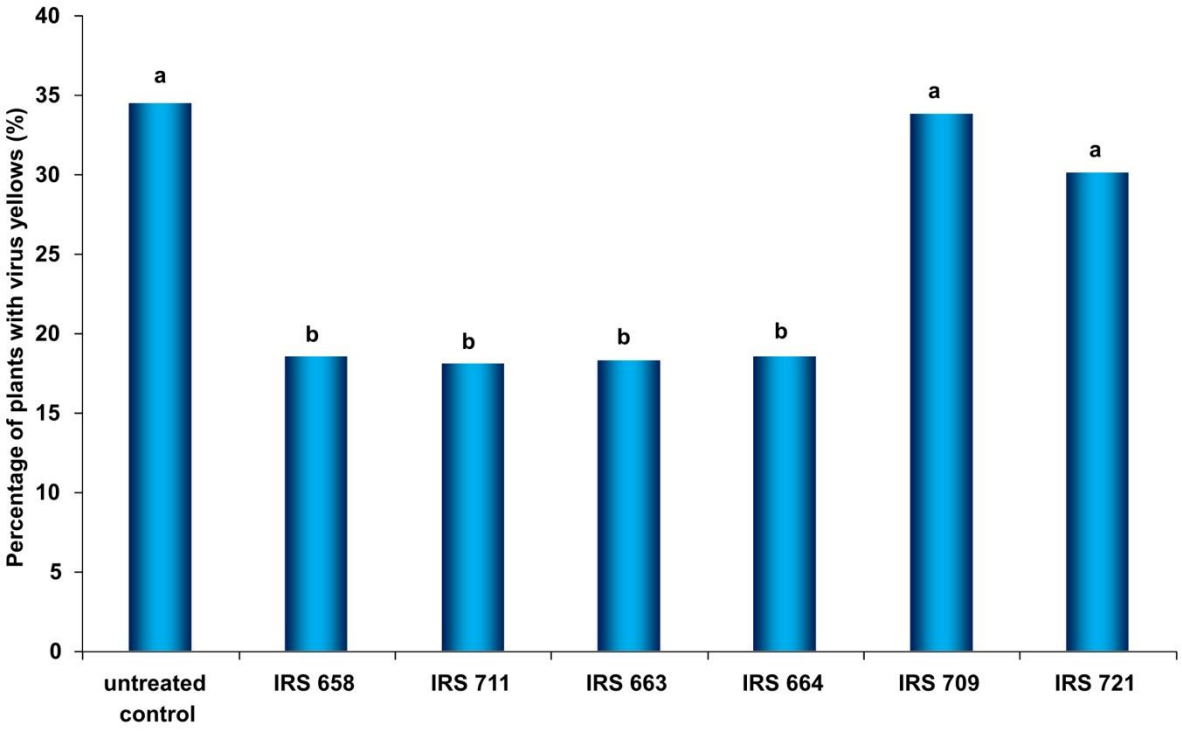


Figure. Mean percentage of plants with virus yellows (BMYV) of the field trial in Colijnsplaat (19 September 2012) (LSD[1] 5% = 6.3; P[2]<0.001).

[1] LSD = least significant difference.
[2] P = probability.