

## The correlation between the transmission of passionfruit ringspot virus and populations of flying aphids

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### Abstract

The percentage of passionfruit ringspot virus infection in batches of young passionfruit seedlings exposed in the open in a plot of diseased fully grown passionfruits was significantly correlated with the number of winged *Aphis spiraecola* trapped in yellow watertrays during the time of exposure.

Of all aphids trapped, 98% were *A. spiraecola*, colonizing predominantly *Eupatorium conyzoides* (Compositae). This plant was found colonized everywhere in the forest area of Ivory Coast, which emphasizes the importance of *A. spiraecola* as a potential virus vector in this part of Africa. The numbers of trapped *A. spiraecola* were positively correlated with the preceding rainfall in mm with an interval of two weeks before the assessment. The influence of a period of rainfall lasted four to six weeks.

### Introduction

In the forest area of Ivory Coast, passionfruit, *Passiflora edulis* var. *flavicarpa*, is always found with symptoms of passionfruit ringspot virus (PRV), cryptogram \*/\* :\*/-(6):E/E:S/Ap. (De Wijs, 1974). Since the crop is easily grown from seed and the virus is not seed-transmitted, there have to be widespread virus source plants and vectors.

The rapidly increasing disease incidence in 24 young passionfruit plants near 12 fully grown diseased plants suggested an abundance of insect vectors. The virus, a member of the potyvirus group, could be transmitted experimentally by *Aphis gossypii* and *A. spiraecola* after brief acquisition feeds (De Wijs, 1974). Therefore a field experiment was set up to find out which aphids were responsible for the spread of PRV in the field.

### Materials and methods

*Experimental site.* The experimental fields of the ORSTOM at Adiopodoumé, Ivory Coast, are situated 17 km west of Abidjan at about 50 m above sea level and surrounded to the North and East side by secondary rain forest and to the South and West side by African cultivation.

*The passionfruit plots.* The site of the two passionfruit plots in the experimental fields of the ORSTOM can be seen on Fig. 1. Passionfruit plot No 1 consisted of three rows

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Fig. 1. Location of the two passionfruit plots in the experimental fields of the ORSTOM: 1 = plot No 1 with three rows of 12 plants; 2 = plot No 2 with two rows of 12 plants; 3 = site of meteorological apparatus; A = secondary rain forest, border is indicated by the broken line; B = African cultivation; Champs d'essais = experimental fields.

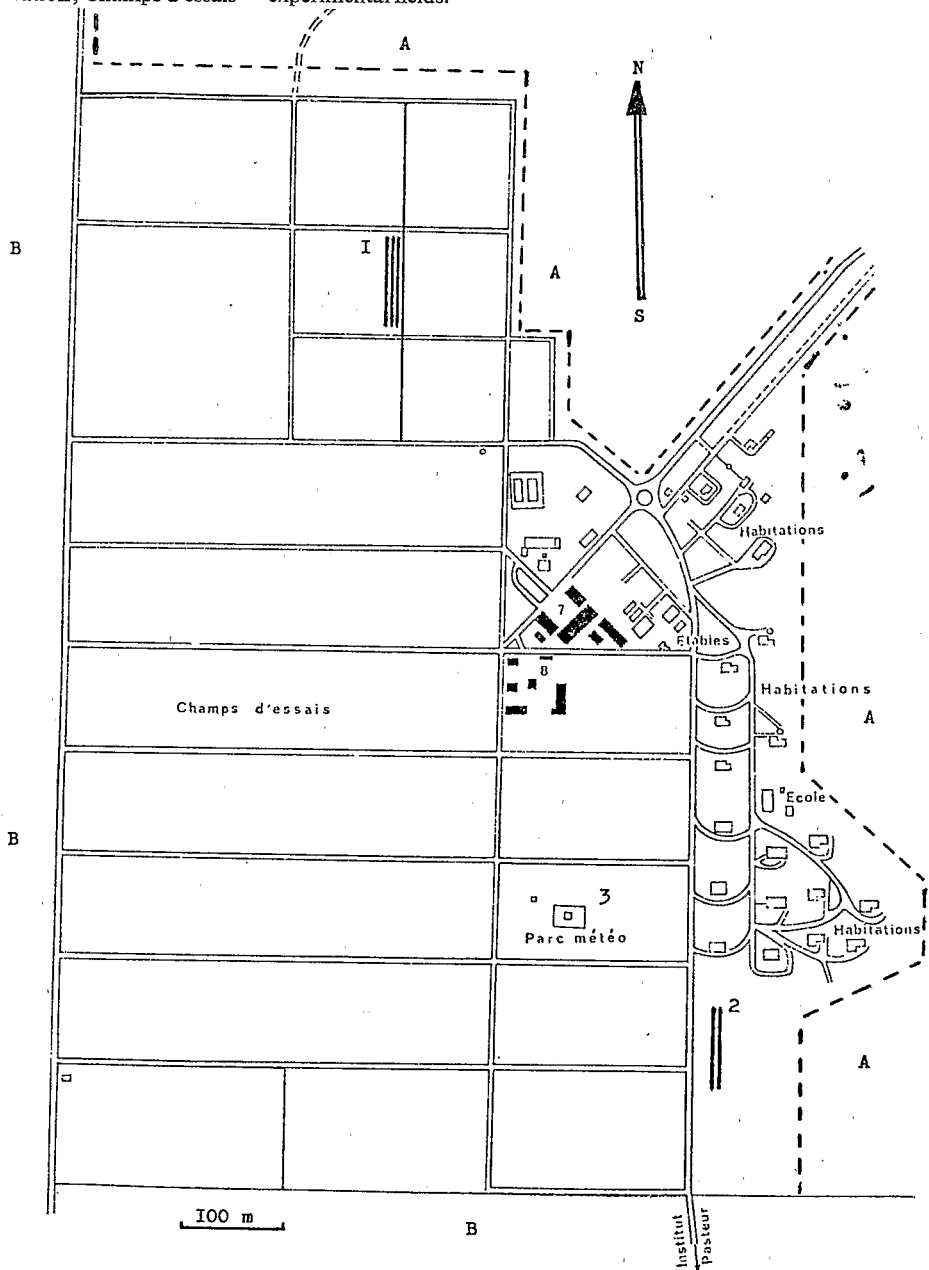


Fig. 1. Ligging van de twee *Passiflora* velden temidden van de proefvelden van het ORSTOM: 1 = veld no 1 bestaande uit drie rijen van 12 planten; 2 = veld no 2 bestaande uit twee rijen van 12 planten; 3 = veld met meteorologische apparatuur; A = secundair regenbos, de bosrand is aangegeven met behulp van een gebroken lijn; B = land bewerkt door Afrikanen; Champs d'essais = proefvelden.

of 12 plants each with 4 m between the rows and 5 m between the plants in the rows. The rows formed hedges of 1.80 m high, directed N-S with cross links each 5 m. The twelve southern most plants were already fullgrown and diseased with PRV in June 1971 when the 24 others were planted as healthy seedlings. The soil cover of this field consisted of a mixture of *Pueraria phaseloides* and wild Gramineae and was cut bi-monthly. The plot was surrounded by experimental fields of *Panicum maximum* and other Gramineae. The second plot consisted of two rows, each of 12 plants, directed N-S and situated in an orchard with Gramineae as soil cover. This plot was planted in May 1971.

*Assessment of the abundance of flying aphids.* Two yellow watertraps of  $38 \times 38 \times 6$  cm were placed 100 cm above the ground. One tray was placed in the shelter of the rows at about 25 m N of the south border; the second was placed at the south border, the windward side of plot No 1. The second tray was only a little sheltered by the passionfruit rows if winds came from S-SE or W-SW. The trapped insects were collected daily, usually between 8 and 9 h a.m.

*Assessment of the PRV transmission rate.* Every month, from November 1971 until October 1972, 100 passionfruit seedlings were planted as 'virus traps' in the two plots, see Fig. 1. The first three to four weeks the young plants were sheltered with palm leaves against sunshine. After three months the plants were checked for PRV infection by visual observation and by control inoculation to *Passiflora foetida* when no obvious symptoms were present. At the date of assessment, the young plants which were too retarded in development because of cricket damage, root-knot attack or causes other than PRV infection, were eliminated and not tested.

*Weather data.* Weather data were obtained from the 'Laboratoire de Bioclimatologie' of the ORSTOM, which has its recording apparatus at a little distance of the passionfruit plots (see Fig. 1). Prevailing winds came from the South.

## Results

*Abundance of flying aphids.* Table 1 shows the aphids trapped in the yellow watertraps during 1971 and 1972. Although the number of species other than *A. spiraecola* trapped during the first four months is not quite certain, about 98% of all aphids trapped were *A. spiraecola*. This is in agreement with field observations throughout the year.

Tray No 1 trapped 1-1.5 times as many aphids as tray No 2. This can be explained by its position in the shelter of the passionfruit rows in comparison to the less sheltered position of tray No 2 (Lewis, 1965).

*Aphid populations detected on their host plants.* As completion of the data obtained with the yellow watertraps, a survey of the flora for the presence of aphid colonies was undertaken in the neighbourhood of the experimental fields throughout the year. The species found are mentioned hereafter.

*Aphis spiraecola* usually occurred in dense colonies on *Eupatorium conyzoides* Vahl (Compositae). This perennial herb was found everywhere at the forest borders and

Table 1. Winged aphids trapped during 1971/1972.

Species	1971							1972							
	Month	D <sup>1</sup>	J <sup>1</sup>	F <sup>1</sup>	M <sup>1</sup>	A	M	J	J	A	S	O	N	D	
<i>Aphis spiraeicola</i> Patch		950	910	286	644	1004	1327	2509	3615	2142	1982	1665	2214	2254	20.552
<i>Aphis gossypii</i> Glover						— <sup>2</sup>	—	12 <sup>3</sup>		1	12	5	28	17	75
<i>Aphis nerii</i> Fonsc.						5	4	7	10	6	—	—	46	13	91
<i>Aphis craccivora</i> Koch						—	—	—	—	—	—	—	1	1	1
<i>Cerataphis variabilis</i> H.R.L.						—	—	2	—	1	2	—	—	3	3
<i>Geoica lucifuga</i> (Zehnter)						—	—	—	—	—	—	—	—	1	1
<i>Longiunguis sacchari</i> (Zehnter)						3	4	5	12	4	2	1	1	1	33
<i>Pentalonia nigronervosa</i> Coquerel				1	2	2	1	3	5	2	3	—	4	42	65
<i>Rhopalosiphum maidis</i> Fitch						—	—	42 <sup>3</sup>		3	—	—	5	—	50
<i>Schoutedenia bougainvilleae</i> Theobald						1	2	—	4	1	—	—	1	—	9
<i>Sitobion congolense</i> Doncaster & H.R.L.						—	—	—	—	—	—	—	—	1	1
<i>Tetraneura nigriabdominalis</i> Sasaki					1	1	7	4	8	11	7	5	8	8	60
<i>Toxoptera aurantium</i> B.D.F.						—	—	2	—	—	—	—	—	—	2
<i>Toxoptera citricidus</i> Kirkaldy						—	—	1	13	3	1	1	15	1	35
unknown			7	20	4										31
total number of winged aphids trapped other than <i>A. spiraeicola</i> :															462
total number of winged <i>A. spiraeicola</i> trapped:															20.552

<sup>1</sup> No exact data of most species are available from December through March.

<sup>2</sup> = species not trapped.

<sup>3</sup> Number of aphids trapped in June and July.

Tabel 1. Gevleugelde bladluizen gevangen in 1971/1972

along roads and tracks and was very abundant at the south and east side of the OR-STOM fields at a distance of 300–1000 m from passionfruit plot 1 and at the forest border near plot 2. The aphids caused a leaf curling by which an aphid infestation could be easily recognized. *E. conyzoides* was found colonized everywhere in the forest area of Ivory Coast. No PRV or other viruses could be isolated from *E. conyzoides* by control inoculations on *P. foetida*. A few colonies of *A. spiraeicola* were found on *Talinum triangulare* Willd. (Portulacaceae) and *Vinca rosea* L. (Apocynaceae).

A few important colonies of other aphid species were found: *Aphis gossypii* Glover on Malvaceae and *Commelina* sp., *Hysteroneura setariae* Thomas on Gramineae (*Eleusine indica* and *Chrysopogon aciculatus*), *Rhopalosiphum maidis* Fitch on maize and *Panicum maximum*, *Toxoptera citricidus* Kirkaldy on *Citrus* sp. Of these *H. setariae* was not detected in the watertraps although colonies with alatae were found beneath.

Of the regularly trapped species (Table 1), *T. nigriabdominalis* was not found by its living habit on the roots of grasses and *A. nerii*, *L. sacchari* and *P. nigronevosa* were not found because their host plants, resp. Apocynaceae, Sorghum and banana do not occur or are to scarcely present in the neighbourhood of the experimental fields.

The predominant position of *A. spiraeicola* in the field is in agreement with the numbers trapped in relation to the number of other aphid species found in the field and in the watertraps.

*Relation between weather data and the abundance of winged aphids.* Table 2 results show that the amount of rainfall and the number of winged aphids trapped are significantly correlated with an interval of at least two weeks and that the effect of a period of rainfall can last for several (4–6) weeks. Two weeks at minimum are needed under the local climatic conditions for a better growth of *Eupatorium conyzoides* and multiplication of the apterous *A. spiraeicola*. This results in crowding in the colonies, which is the most important determining factor for the production of alatae (Lees, 1967).

The data given in Fig. 2 show the numbers of *A. spiraeicola* trapped each week and the weekly rainfall in mm. The shape of the diagrams suggests that periods of rain are followed by periods of aphids. This is especially prominent from Dec. 1971 until April 1972: three separate periods of rain (a, b and c) gave rise to three periods of aphid flights (A, B and C). The rain effect lasted for about six weeks. The aphid flight in August–September resulted from the rainfall in July. That this flight is much more important than the flights A, B and C is due to the good condition of the host plants under less severe climatic conditions (Table 3). This was true even though rainfall was lower in comparison to the period from Dec. 1971 until April 1971. The effect of rain on aphid numbers is less obvious during the rest of the year.

*Disease incidence in plot 1 and 2.* The increase in disease incidence in plots 1 and 2 and the percentage of infection in the batches of seedlings in plot 2 are shown in Fig. 3.

The chance that an infective aphid alights and feeds on one of the trap plants depends on several factors: the abundance of winged aphids, the amount and proximity of virus source plants and the size of the trap plants (Nelson and Tuttle, 1969). Plot 1 contained already 12 fully grown and diseased plants while plot 2 had no adjacent virus sources. This explains the high rate of infection plot 1 in 1971. The first infection of one plant in plot 2 was found in January 1972 (primary infection). The rest

Table 2. Correlation coefficients of rainfall in mm, aphid numbers and PRV percentage in the batches of passionfruit seedlings.

A. Cumulative data of 12 weeks, time of exposure of the passionfruit seedlings:			
rainfall on aphids	0,3243		
rainfall on percentage PRV	0,0738		
aphids on percentage PRV	0,8063	S p = 0,01	
aphid numbers converted to log (n + 1) on percentage PRV	0,8353	S p = 0,001	
(n = 12; p (0,01) = 0,708; p (0,001) = 0,823)			
B. Weekly totals of rain and aphid numbers (18/12/71 until 6/1/73).			
rain on aphids.	-0,0505		
rain of 1st preceding week on aphids	0,2332		
rain of 2nd preceding week on aphids	0,4654	S p = 0,001	
rain of 3rd preceding week on aphids	0,3755	S p = 0,01	
rain of 4th preceding week on aphids	0,4669	S p = 0,001	
rain of 5th preceding week on aphids	0,4246	S p = 0,01	
(n = 12; p (0,01) = 0,34; p (0,001) = 0,43)			
C. Cumulative data of two weeks rain and aphids (8/1/72-6/1/73)			
rain on aphids	0,1290		
rain of 1st preceding 2 weeks on aphids	0,5561	S p = 0,01	
rain of 2nd preceding 2 weeks on aphids	0,5632	S p = 0,01	
rain of 3rd preceding 2 weeks on aphids	0,5001	S p = 0,01	
rain of 4th preceding 2 weeks on aphids	0,5346	S p = 0,01	
(n = 12; p (0,01) = 0,49; p (0,001) = 0,61)			
D. Cumulative data of 4 weeks rain and aphids (1/1/72-31/12/72)			
rain on aphids	0,1816		
rain of first preceding 4 weeks on aphids	0,7254	S p = 0,01	

Tabel 2. Correlatiecoëfficiënten tussen de regenval in mm, de aantallen gevangen bladluizen en het percentage PRV in de groepen *Passiflora* zaailingen.

of the increase in disease incidence must be attributed to spread of PRV from this first plant to others in the plot (secondary spread). The percentage of PRV infection in the batches of passionfruit seedlings in plot 2 (Fig. 3, curve 3) depends obviously on the disease incidence in plot 2. PRV transmission from the fully grown plants to the trap plants starts only when the virus is spreading rapidly in this plot.

*Relation between infection percentage and numbers of winged A. spiraeicola trapped.* There was a significant positive correlation between the numbers of winged *A. spiraeicola* trapped and the PRV infection percentage in the trap plants in plot 1. Aphids were counted for the twelve weeks during which the passionfruit seedlings developed until checked for PRV infection (Table 2, Fig. 4).

Of the seven other aphid species trapped regularly (total numbers ranging from 33 to 91, see Table 1) the numbers trapped are too low to permit with reliability the calculation of a correlation as done for *A. spiraeicola*. At least four of these are known to be able to transmit non-persistent viruses. These four species are *A. gossypii*, *P. nigronervosa*, *R. maidis* and *T. citricidus* (Fritsche et al., 1972). Even when the strong attraction of *A. spiraeicola* for yellow is taken in to account according to O'Loughlin (1963), *A. spiraeicola* outnumbered *A. gossypii* with a factor 20. *A. gossypii* is as efficient a vector of PRV as *A. spiraeicola* (De Wijs, 1974). *T. citricidus* is attracted as much as *A. spiraeicola* to yellow. This species transmitted only occasionally the virus (De Wijs

Fig. 2. Weekly totals of rainfall in mm and alate *A. spiraeola* trapped at the ORSTOM in 1971/1972. Data on aphid numbers trapped in 1971 are corrected since at that time a less effective bright yellow colour was used for the water pans. Periods of rainfall a, b and c are followed by corresponding periods of aphid flights A, B and C.

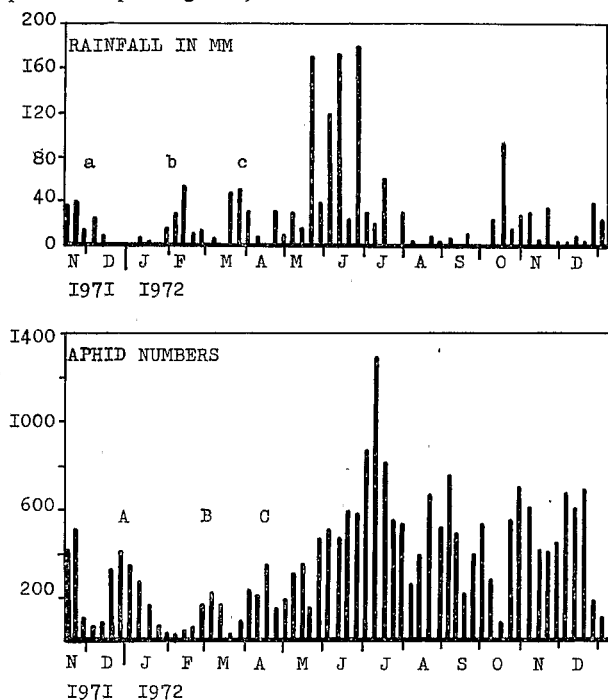


Fig. 2. De wekelijkse totalen van de regenval in mm en de aantallen gevangen gevleugelde *A. spiraeola* op het ORSTOM in 1971|1972. De aantallen gevangen gevleugelde bladluizen in 1971 zijn gecorrigeerd omdat toen een minder effectieve lichtgele kleur gebruikt is voor de vangbakken. Drie periodes van regenval a, b en c worden duidelijk gevolgd door drie periodes van bladluisvluchten A, B en C.

Table 3. Monthly weather data as registered by the 'Laboratoire de Bioclimatologie' of the ORSTOM.

Year	Month	Rainfall in mm	Hours sunshine	Average maximum temp. (°C)	Average minimum temp (°C)
1971	D	36	206	30.1	22.6
1972	J	9	207	30.4	22.9
	F	120	214	30.9	23.4
	M	101	226	30.7	23.2
	A	83	189	30.6	22.9
	M	253	170	29.8	23.3
	J	518	120	28.5	22.8
	J	145	79	26.9	22.5
	A	17	74	26.2	21.5
	S	20	90	27.3	22.2
	O	186	178	29.1	23.2
	N	101	166	29.8	23.3
	D	82	178	30.2	22.8
1973	J	0	220	30.8	23.6

Fig. 3. Increase in disease incidence in plots 1 and 2 (curve 1 and 2, respectively and percentage of infection in the batches of young seedlings in plot No 2 (curve 3) after three months of growth.

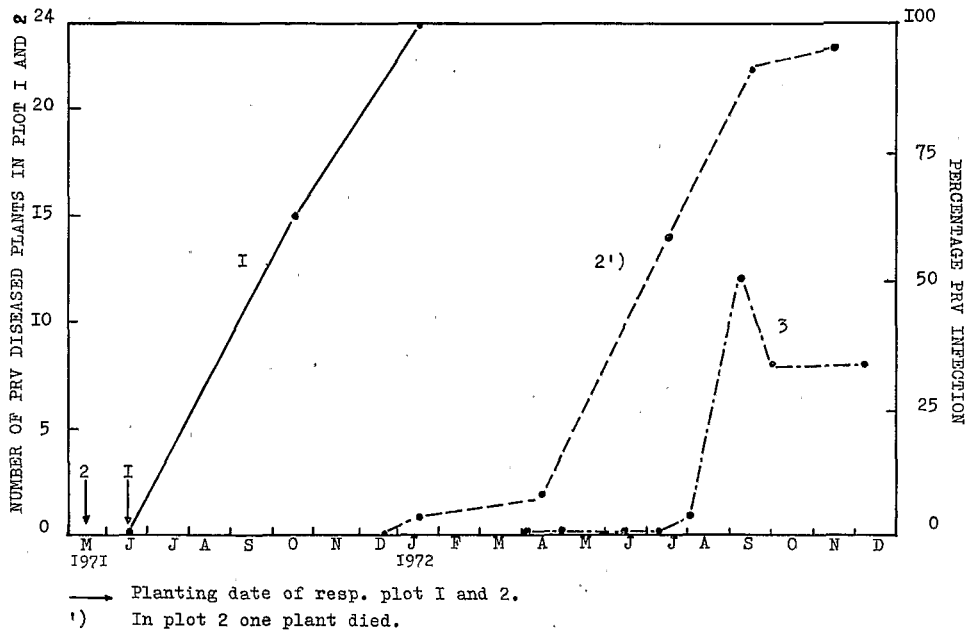


Fig. 3. Toename van het aantal PRV-zieke planten in veldjes 1 en 2 (resp. curve 1 en 2) en het percentage PRV-zieke planten in de groepen zaailingen na drie maanden groei in veld 2 (curve 3).

and De Vrijer, unpublished data). *R.maidis* and *P.nigronevosa* belong to the grass and sedge feeding aphids, which are in general not attracted by yellow. Large numbers of these species might therefore be overlooked. The irregular distribution of *R.maidis* over the year, related to maize cultivation on the experimental fields of the ORSTOM, excludes however a possible relation with the constant virus transmission in the passionfruit fields. The scarce presence of *Musa* and *Colocasia* spp., main host plants of *P.nigronevosa* in relation to the abundance of *A.spiraecola* infested *E.conyzoides* suggest that alatae of *A.spiraecola* must amply outnumber those of *P.nigronevosa* in the air.

Although more than one aphid species might have attributed to the transmission of PRV in the field, the present data suggest that *A.spiraecola* is the main vector under the local conditions.

*The influence of the growth rate of the virus source plants on the PRV infection percentage of the trap plants in plot 1.* The growth rate of the passionfruits showed to be correlated with the changes in weather. Monthly weather data are given in Table 3. The first four months of 1972 were dry and hot which inhibited new growth of the virus source plants. PRV infection percentages were low during this period (Fig. 4). Rainfall in April-May caused a sudden growth in May. This can explain the high infection percentage found in June since only a twofold rise in aphids trapped does not



Fig. 4. The changes in percentage PRV infection in the batches of seedlings after a three months period of exposure in plot 1 in relation to the numbers of trapped alate *A. spiraeicola* during the 12 weeks preceding the week of testing for virus infection.

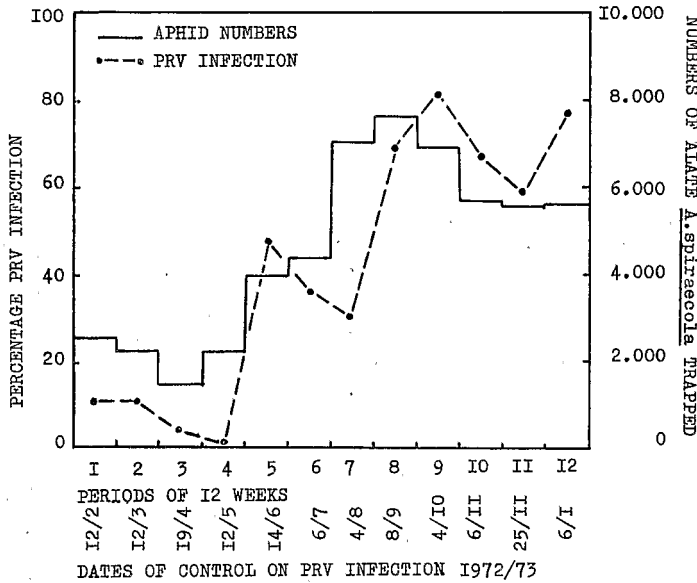


Fig. 4. Het verloop van de percentages PRV-zieke planten in de groepen zaailingen na 3 maanden verblijf in veldje 1 ten opzichte van de aantallen gevangen gevleugelde *A. spiraeicola* gedurende 12 weken voorafgaande aan de week van toetsing op virusinfectie.

explain the sharp rise in infection percentage. New growth was scarce in June and July, causing lower infection percentages in July and August than expected on the ground of the numbers of aphids trapped. New growth started again in August and continued until the end of the year. It was most abundant in November. Although the rainfall in August and September did not reach the level of the first four months of the year, the less severe climatic conditions permitted nevertheless the growth of young shoots which was favourable for maintaining a high infection percentage by the numerous winged aphids present (Fig. 2 and 4).

## Discussion

A significant correlation could be demonstrated between the numbers of winged aphids trapped and the percentage of PRV infection found in batches of young passionfruit seedlings. One aphid species, *Aphis spiraeicola*, appears to be the only important vector under our conditions since 98% of all aphids trapped consisted of this species. *A. spiraeicola* does not colonize *P. edulis*, neither do other aphid species but alatae have been detected visiting it.

Broadbent et al. (1950) demonstrated likewise that potato virus Y was transmitted by alate *Myzus persicae* coming from diseased potato fields to plantings of young

potatoes during the summer flight when the infection percentage of surrounding fields is high.

*A. spiraeicola* may have recently been introduced in West Africa since Eastop (1961) and A'Brook (1968) do not mention it. Hall et al. (1972) first recorded *A. spiraeicola* and found it associated with *Eupatorium odoratum* in Ghana. De Wijs (1973) first recorded the species as a virus vector in Africa.

The primary infection of a crop with a non-persistent virus does not depend on the abundance of winged aphids as much as on the abundance and proximity of virus source plants (Nelson and Tuttle, 1969). This is confirmed by the difference in infection rate of passionfruit plot 1 and 2. The disease increase in plot 1 (Fig. 3, curve no 1) is the result of multiple entries of the virus in the plot in contrast to the increase in plot 2 (Fig. 3, curve no 2) where the lag phase represents the primary infection and the subsequent rapid increase represents the secondary spread inside the plot.

Under natural conditions the primary infection of a crop by a non-persistent virus will be the 'threshold' before secondary spread is possible. This spread is cut down when the crop is also a breeding host for the vector of the virus (Watson and Healy, 1953; Nelson and Tuttle, 1969). Secondary spread is favoured when the vector does not breed on the crop as in our case. The rapid spread of viruses in cantaloups (Nelson and Tuttle, 1969) and of lettuce mosaic virus in lettuce (Gonzalez and Rawlins, 1969) could therefore be related to flights of *Myzus persicae*.

Different factors of plant growth might be involved in virus transmission. Mueller (1964) has demonstrated that a yellow cultivar of lettuce attracted more aphids than a reddish-brown one. Young passionfruit leaves are more yellowish-green than older leaves. Aphids might thus be more attracted to actively growing passionfruits, which results in more landings and probes, thus increasing the possibility of transmission. Moreover the virus might reach a high titer in the outgrowing leaves which increases the likelihood of transmission. Availability of potato virus Y, cucumber mosaic virus (Simons, 1958), bean yellow mosaic virus (Swenson, 1962) and alfalfa mosaic virus (Matisova, 1971) to aphids was correlated with virus concentration as assayed by serology or with indicator plants.

Temperature and rainfall are the most important regulating factors in moist tropical climates on the seasonal cycle of insects (Broadbent, 1967). The temperature at the ORSTOM reaches the extreme values of about 19–25°C as minimum at sunrise and 25–32°C as maximum during the day. Hence it is not a limiting factor for aphid reproduction and plant growth. The growth of the most important host of *A. spiraeicola*, the perennial herb *Eupatorium conyzoides*, is ensured all the year round since there is usually some rainfall during the dry season. Nevertheless, differences in rainfall affect obviously this plant and therefore the multiplication and flights of *A. spiraeicola*. Rainfall is thus the most important environmental factor for the reproduction of *A. spiraeicola* at the ORSTOM.

No exact observations were made on predators and parasites of *A. spiraeicola*. Although larvae of Coleoptera and Neuroptera were found preying in the apterous aphids, the predation did not seem sufficient to interfere with rainfall and growth of the host plant as intermediate regulating factor in aphid flight frequency.

A positive correlation between rainfall and flight of *A. spiraeicola* has also been mentioned by Van Hoof (1962) in Surinam, but he made no mathematical calculations and did not indicate a time interval.

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## Samenvatting

*De correlatie tussen populaties gevleugelde bladluizen en de overdracht van 'passionfruit ringspot virus'*

'Passionfruit ringspot virus' (PRV) werd in Ivoorkust in het veld zeer snel verspreid van oudere zieke naar gezonde jonge zaailingen van *Passiflora edulis*, de passievrucht. *P. edulis* zelf bleek geen waardplant te zijn voor eventuele vectoren. De aantallen gevleugelde bladluizen, gevangen in gele vangbakken gedurende een bepaalde periode, bleken echter significant gecorreleerd te zijn met de percentages zieke planten van groepen jonge *Passiflora* zaailingen die een overeenkomstige periode aan infectie in het veld hadden bloot gestaan.

Van het totaal aantal gevangen gevleugelde bladluizen bleek *Aphis spiraeicola* 98% uit te maken. Alleen deze bladluis werd verantwoordelijk gesteld voor de overdracht van het virus in het veld. Als waardplant voor *A. spiraeicola* dient voornamelijk het meerjarig onkruid *Eupatorium conyzoides* (Compositae), dat zeer algemeen voorkomt in de omgeving van de proefvelden van het ORSTOM. Ook elders in de regenbos-zone van Ivoorkust wordt deze plant algemeen aangetroffen met kolonies van *A. spiraeicola*, wat deze bladluis tot een belangrijke potentiële virusvector in dit deel van Afrika maakt.

De aantallen gevangen gevleugelde exemplaren van *A. spiraeicola* waren significant gecorreleerd met de twee weken daaraan voorafgaande neerslaghoeveelheden. De invloed van een regenperiode hield 4-6 weken aan.

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