

Using a procedure developed in the laboratory for artificial inoculation in controlled conditions under greenhouse, young rubber plants were infected as follows : plant-growth tubs were filled with soil, whose humidity level was controlled with neutron probes. Sticks of Hevea wood, sterilized in Roux flasks and seeded with one of the fungi, constituted the inoculum. Because of the different degradative powers of these fungi, the incubation before inoculation lasted five months for *P. noxius* and eleven months for *R. lignosus*. For each one-month-old plant, five of these sticks were applied against the tap root, 20 cm deep in the soil. Each isolate was inoculated onto 30 plants in a single tub. The experiment lasted 5 months but 10 randomly chosen plants were harvested early, 2 months after inoculation.

In order to quantify the infection of each inoculated plant, the lengths of the stem and the root, the appearance of symptoms on the leaves, the date of death of the plant, the presence of rhizomorphs on the root, any reactive rhizogenesis, and the degree of colonization of the tissues (local penetrations and proportion of necrosis) were recorded. A plant health scale from 0 to 9 was established, incorporating these various criteria, so that each plant and each isolate could be given a score representing the severity of the attack (table 2).

PATHOTYPES OF *RIGIDOPORUS LIGNOSUS*

* Infectious processes appraisal (after 2 months)

From the sample at 2 months it is possible to appraise the initial stages of pathogenesis, namely rhizomorphogenesis, which conditions the contamination and initial penetrations.

The results obtained with each batch of 10 plants were collected in table 3. The strains are classified in order of decreasing phytosanitary scores (PS). For all the isolates, all the inoculated plants were contaminated, and all the tap roots, except those of certain plants with isolates 1 and 37, were penetrated by the fungal hyphae. For the first four isolates, nearly 50 % of the plants inoculated were dead or had leaf symptoms of impending death. The examination of their tap roots showed that on average they were 70 % necrosed from apex to collar. The pathogenicity of the four other isolates seems, on the other hand, to have been notably lower, especially that of strain 37, with which the mortality level was 0. Correspondingly, the reactional secondary rhizogeneses of the plants infected by this isolate were clearly perceptible. Finally this sampling clearly demonstrates that as early as two months there were real differences in pathogenicity between the strains tested. These differences were not dependent on the quality of the inocula, that is, on the initial capacity of each isolate to contaminate plants via rhizomorphs, since all the plants were contaminated a few weeks after inoculation.

Fungal attack affected plants' growth, since weekly measurements of the stem heights of the plants showed a delay in growth at two months that became more marked up to the end of the experiment (figure 1).

does not yield any new information. The most pathogenic isolates lie on axis 1 negative. As an indication, in this group of five strains, three were isolated from *Hevea*, one from *Tectona grandis*, one from a forest tree, in Cameroon and in various regions of the Ivory Coast...

PATHOTYPES OF *PHELLINUS NOXIUS*

A similar experiment, also comparing eight isolates, was carried out with *P. noxius*. In order to understand this analysis, it is important to bear in mind that the biology of this fungus is different from that of *R. lignosus*. *P. noxius* has no mycelial propagation structures comparable with rhizomorphs and so, contaminates the roots by means of a mycelial sleeve. The contamination is therefore a critical phase in the achievement of artificial infection.

* Infectious processes appraisal (after 2 months)

Indeed, and unlike *R. lignosus*, *P. noxius* contaminated plants very unequally after two months ; this affected the later development of the infection with regard to penetration and mortality (table 6). With all but three isolates (numbers 2, 32, and 45, which killed yet some of the inoculated plants) the infectious process was just initiated. This is corroborated by the absence of signs of anatomical reaction. At this stage the impact of the parasitism on the growth of the plants was still barely perceptible, but it increased significantly during the following months and conveyed the development of the infection into the roots tissues (figure 4).

* Pathogenesis analysis (after 5 months)

Overall 7 out of the 8 isolates tested demonstrated their parasitic ability after the 5 months of confrontation but the previously noticed differences became more marked (table 7). The variability in the parasitic behavior of each isolate can be illustrated by the following examples :

- strain 45 showed great ability both to contaminate and to colonize tap roots, right from the start of the experiment. The severity of its attack, shown by the rate of 65 % dead or dying plants, was inversely correlated with the ability of *Hevea* to establish secondary rhizogenesis.

- isolate 4 is similar to the preceding isolate in its abilities to contaminate and penetrate tap roots. However it caused clearly lower mortality, while paradoxically the frequency of host reactions remained low. It can be assumed that there were real differences in aggressiveness between these two isolates, since in both cases the contamination rates were excellent. But, on the other hand, what conclusions can be drawn about strain 31, whose contamination rate was particularly low ?

Informations about the scatter of the results for this isolate are given in table 8. In fact it can be seen that only two of the twenty inoculated plants were contaminated. This homogeneity rules out any technical error at the time of inoculation and one may justifiably deduce that this strain has a low contamination capacity. On the other hand, the fact that the plants infected with the very aggressive isolates (45, 32, 35, 4 and 2) had phytosanitary scores that were scattered between 3 and 9, reveals a substantial intra-isolate variability. At first sight this could be attributable to the fact that contamination of the plants was not synchronous and the impact of the infection was thus to some extent delayed.

The overall development of infestation of the plants by each isolate between two and five months is shown in figure 5. Contrary to the results with *R. lignosus*, differences in behavior between isolates of *P. noxius* take the form of differences in slope between straight line segments. Thus, for example, isolate 35 made up for the delay at the time of inoculation, whereas strain 45 scarcely progressed between two and five months. The behavior of the other isolates was intermediate between these two extremes.

The kinetics of the mortality of the plants adds to the analysis. Two types of behavior can be distinguished in the figure 6:

- that of strains 39, 4 and 35 for which pathogenesis began slowly and remained low ;

- that of strains 45, 32 and 2, whose mortality kinetics were bimodal, with an initial ascending phase that was particularly marked for isolate 45 producing a mortality of 40 % in onemonth. This was followed by a second, plateau phase observable from the third month. To explain this phenomenon of stagnation of infection three possible hypotheses can be suggested : first, exhaustion of the fungus due to impoverishment of its trophic reserves in the sticks that carried the inoculum ; second, an abrupt arrest of the progression of the fungus in the tissues by host cellular and/or anatomical reactions ; third, delayed contamination of a fraction of the inoculated plants in the tub, whose level of infestation because of this delay scarcely reached that of the plants contaminated right from the beginning of the experiment.

Integration of all the data by principal-component analysis established a clear hierarchy of isolates (figure 7). This type of analysis made it possible to discriminate between isolates 45 and 32 even though their average phytosanitary scores were very similar. It is therefore reasonable to suppose that the modalities of infection are different for these two most aggressive isolates. Isolates 4, 35, and 2 then form one set distinct from isolates 7, 39 and 31.

Finally it is interesting that strains 31 and 32, which differ greatly in pathogenicity were nevertheless isolated from the same plantation.

N° ISOLATE		COUNTRY CRIGIN		HOST	ISOLATION DATE
RIGIDOPORUS LIGNOSUS	1	South East	IVORY COAST	<i>Hevea brasiliensis</i>	1978
	9	East	IVORY COAST	Primary Forest	1978
	13	West	IVORY COAST	Primary Forest	1978
	21	South East	LIBERIA	<i>Hevea brasiliensis</i>	1979
	37	South East	IVORY COAST	<i>Hevea brasiliensis</i>	1981
	38		CAMEROON	<i>Hevea brasiliensis</i>	1981
	42	South West	IVORY COAST	<i>Hevea brasiliensis</i>	1981
	52	East	IVORY COAST	<i>Tectona grandis</i>	1981
PHELLINUS NOXIUS	7	West	IVORY COAST	Primary Forest	1978
	45	East	IVORY COAST	<i>Cedrela odorata</i>	1981
	31	South West	IVORY COAST	<i>Hevea brasiliensis</i>	1980
	4	South East	IVORY COAST	<i>Hevea brasiliensis</i>	1978
	35	East	IVORY COAST	<i>Hevea brasiliensis</i>	1981
	39		CAMEROON	<i>Hevea brasiliensis</i>	1981
	2	South West	IVORY COAST	<i>Hevea brasiliensis</i>	1977
	32	South West	IVORY COAST	<i>Cedrela odorata</i>	1980

TABLE 1

ORIGINS OF ISOLATES

RIGIDOPORUS LIGNOSUS		PHELLINUS NOXIUS	
NOTE		NOTE	
0	no mycelium on roots	0	no mycelium on roots
1	non aggregate hyphae	1	mycelium crust in formation
2	rhizomorphs	2	well - formed mycelium crust
3	rhizomorphs and ponctual penetration	3	mycelium crust and ponctual penetration
4	rhizomorphs and localized necrosis	4	mycelium crust and localized necrosis
5	rhizomorphs and partial root decay <20%	5	mycelium crust and partial root decay <20%
6	rhizomorphs and 20 to 50% root decay	6	mycelium crust and 20 to 50% root decay
7	rhizomorphs and root decay >50%	7	mycelium crust and root decay >50%
8	foliar symptoms	8	foliar symptoms
9	plant death	9	plant death

TABLE 2

PHYTOSANITARY SCALE

- % CONTAMINATION : note 1 to 9
- % PENETRATION : note 3 to 9
- % MORTALITE : note 9

Pathogenicity variability in 8 *Rigidoporus lignosus* strains

Phytosanitary scale												PS
		0	1	2	3	4	5	6	7	8	9	
N°	Strains											
52	<i>Teak</i> IC East								5		95	8,9
42	<i>Hevea</i> IC South West						5		5		90	8,7
13	Forest IC South West								15		85	8,7
38	<i>Hevea</i> CAM						5	5	15	5	70	8,3
21	<i>Hevea</i> LIB South East						10	10	10		70	8,1
1	<i>Hevea</i> IC South East								10	15	75	8,4
9	Forest IC East			5		10		25	10	5	45	7,2
37	<i>Hevea</i> IC South East		15			5	20	35	15		10	5,5

TABLE 5

PHYTOSANITARY CLASSIFICATION FOR EACH HOST ISOLATE COMBINATION

Results in % of infected rubber trees for each phytosanitary class (0 to 9)

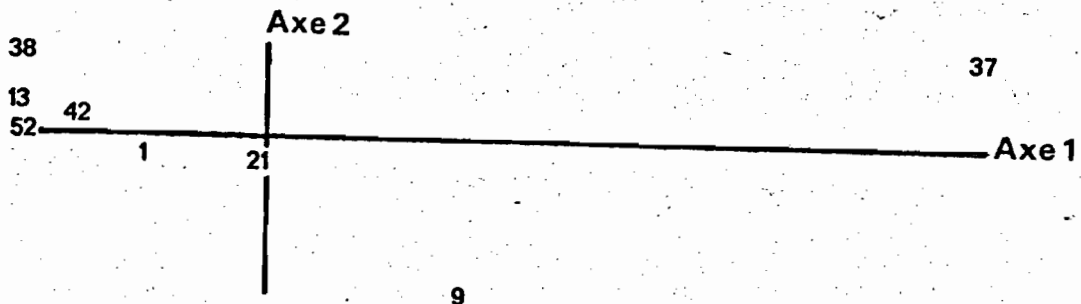


FIGURE 3

POSITION OF EACH ISOLATE ON THE PLAN 1 - 2 OF THE PRINCIPAL COMPONENT ANALYSIS , FIVE MONTHS AFTER INOCULATIONS WITH RIGIDOPORUS LIGNOSUS.

Pathogenicity variability in 8 Phellinus noxius strains

Isolates	N°	PS	Contamination (%)	Penetration (%)	Foliar symptoms + Mortality (%)	Necrosis (%)
<i>Cedrela</i> IC East	45	6,3	100	100	50	40
<i>Cedrela</i> IC South West	32	3,6	80	60	20	21
<i>Bevea</i> IC South West	2	3,4	50	50	20	28
<i>Bevea</i> IC South East	4	3,0	70	70	0	0
<i>Bevea</i> IC East	35	1,2	30	0	0	0
Forest IC South West	7	0,8	30	10	0	3
<i>Bevea</i> CAM	39	0,3	10	10	0	0
<i>Bevea</i> IC South West	31	0,2	20	0	0	0

TABLE 6

INFECTIOUS PROCESSES DESCRIPTION 2 MONTHS AFTER INOCULATION

Pathogenicity variability in 8 Phellinus noxius strains

Isolates	N°	PS	Contamination (%)	Penetration (%)	Foliar symptoms + Mortality (%)	Necrosis (%)	Secondary rhizogenesis (%)
<i>Cedrela</i> IC East	45	6,5	90	80	65	53	5
<i>Cedrela</i> IC South West	32	6,2	95	95	40	24	25
<i>Bevea</i> IC East	35	5,1	85	80	30	30	30
<i>Bevea</i> IC South East	4	4,8	85	80	15	22	5
<i>Bevea</i> IC South West	2	4,5	65	65	25	16	25
Forest IC South West	7	2,6	60	60	5	7	5
<i>Bevea</i> CAM	39	1,5	35	30	5	5	10
<i>Bevea</i> IC South West	31	0,4	10	10	0	0	0

TABLE 7

INFECTIOUS PROCESSES DESCRIPTION 5 MONTHS AFTER INOCULATION

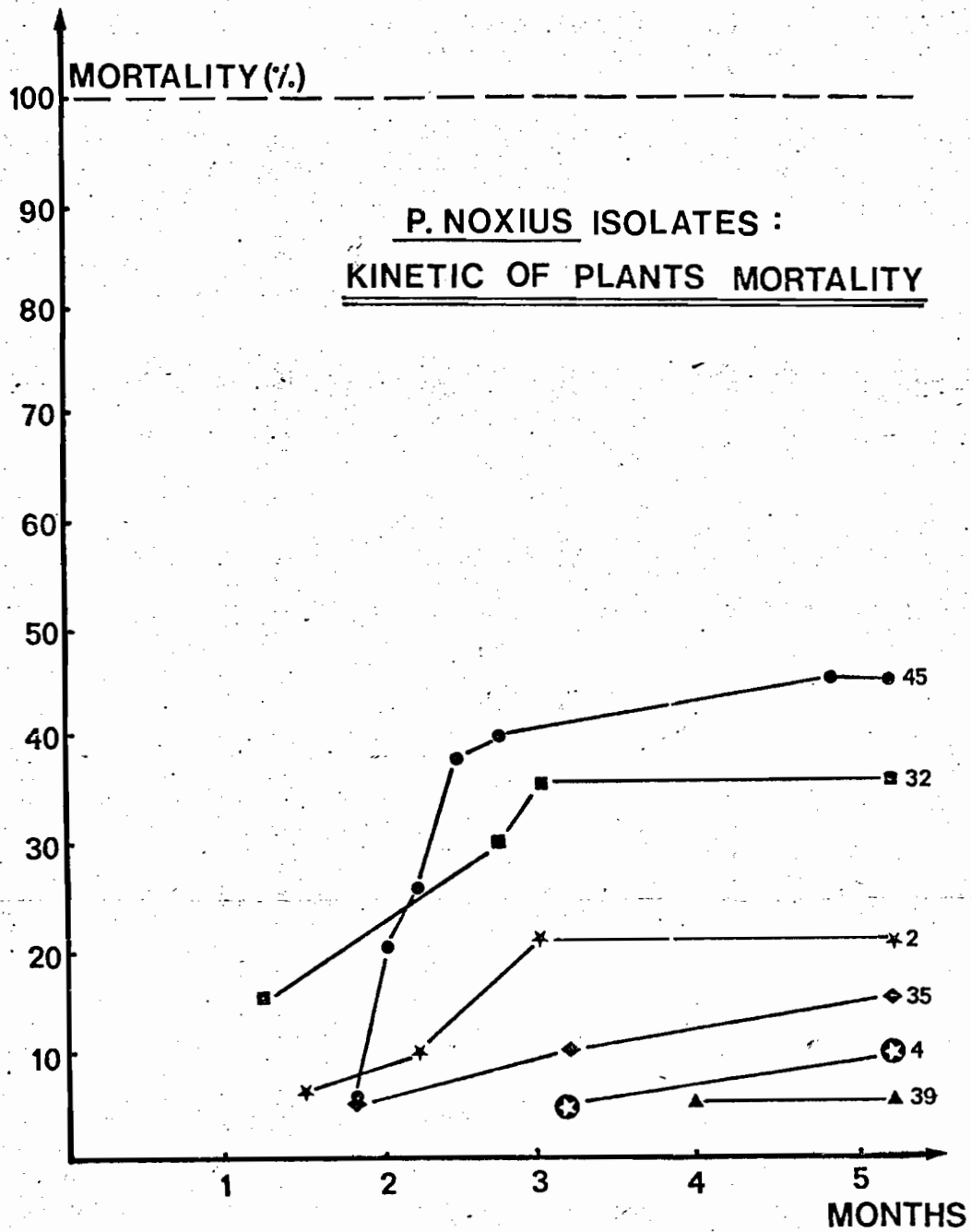


FIGURE 6

Pathogenicity variability in 8 *Phellinus noxius* strains

Phytosanitary scale Strains		0	1	2	3	4	5	6	7	8	9
		45	<i>Cedrela</i>	IC East	10			5	10		
32	<i>Cedrela</i>	IC South West				20	30		5	5	35
35	<i>Hevea</i>	IC East	15	5		5	10	20	15		15
4	<i>Hevea</i>	IC South East	15	5			10	40	5	10	5
2	<i>Hevea</i>	IC South West	35				5	10	10	15	5
7	Forest	IC South West	40			10	35	10			5
39	<i>Hevea</i>	CAM	65	5			20	5			
31	<i>Hevea</i>	IC South West	90			5	5				

TABLE 8 PHYTOSANITARY CLASSIFICATION FOR EACH HOST ISOLATE COMBINATION

Results in % of infected rubber trees for each phytosanitary class (0 to 9)

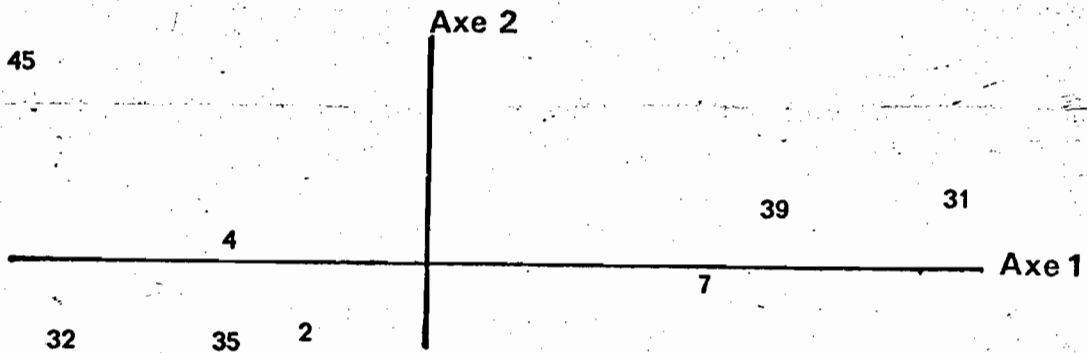


FIGURE 7 POSITION OF EACH ISOLATE ON THE PLAN 1 - 2 OF THE
PRINCIPAL COMPONENT ANALYSIS, FIVE MONTHS AFTER INOCULATION
WITH PHELLINUS NOXIUS.