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Pachyrhizus toxicity: Genetic variability of mature and immature seeds and its effects on *Sclerotium rolfsii* and *Ralstonia solanacearum*

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***Pachyrhizus* seeds-induced toxicity is very well-known to have developed in humans, fishes, insects, and some micro-organisms. Yet, there is a paucity of studies addressing it in their germplasm, as well as its effects on some phytopathogens. This work aimed to assess the *Pachyrhizus* genetic variability on toxicity and to determine *Pachyrhizus* extract toxicity on *Sclerotium rolfsii* and *Ralstonia solanacearum in vitro*. In the findings, it was shown to only have toxicity genetic diversity in immature seeds. Its toxicity in mature seeds was very high and its genetic variability was not detected. There was a toxic effect on *S. rolfsii* and *R. solanacearum*. In *S. rolfsii* it was determined that the most toxic dose was 1: 1000 with P40 genotype. It decreased the mycelial growth by 72% following 5 days. In *R. solanacearum* it was determined that the most toxic dose was 1:200 with P40 genotype. It reduced bacterial multiplication by 57% between 24 and 48 h.**

Key words: *Pachyrhizus* extract, genetic variability, toxicity, *Sclerotium rolfsii*, *Ralstonia solanacearum.*

INTRODUCTION

The development of biopesticides that will make organic food production more feasible and combat domestic pests is a new trend in the market. There is currently a worldwide shortage of this type of pesticide, which has shown to be even more acute in Europe, since it is where more strict legislation for reducing the use of synthetic pesticides is being enforced (Amanatidis, 2022). Thus, research is needed to assess the toxic potential of different plant species that could be used as biopesticides.

The Amazon holds an immense flora, which could be characterized in the search for these biopesticides; though very few efforts have been made to do it. A quick way to explore these resources would be the use of underutilized cultivated plants according to the traditional knowledge. Using plant biopesticides directly without

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estimating their dose or identifying the genotype can be as harmful to health as synthetic pesticides. For example, tobacco extracts that are currently widely used in the Amazon region, are toxic to humans both when inhaled and in contact with the skin. Thus, they have been banned in several countries (Gonzalez-Coloma et al., 2010), on account of it.

Initially, pesticides were always obtained from plants. Gonzalez-Coloma et al. (2010) showed Eugenol to be obtained from laurel (*Laurus* species), azaridachtina from the neem tree (*Azadirachta indica*), nicotina from *Nicotiana tabacum*, and karanjina from *Derris indica* Karanjina. It also showed insecticidal vegetable oils to be obtained from *Brassica napus, Brassica campestris, Capsicum* species, *Tagetes erecta, Thymus vulgaris* and *Gaultheria procumbens*, insecticidal acids from *Varroa destructor* and *Pyrethrins, chrysanthemats* and pyrethrates from *Tanacetum cinerariaefolium* flowers. Rotenone from the genera *Derris, Lonchocarpus* and *Tephrosia*. Natural commercial insecticidal extracts were obtained from the stem of *Ryania speciosa*, crushed seeds of *Schoenocaulon officinale*, potato and corn starch syrup (Gonzalez-Coloma et al., 2010). Also, soluble powder of *Derris* is marketed with different concentrations of rotenone (Undersun Biomedtech, 2022; Yates, 2022).

Yam bean (*Pachyrhizus* species, Fabaceae), also called jacatupé in southeastern Brazil, jícama in Mexico and Central America and bengkoang in Indonesia is a promising Amazonian plant that could quickly be used as a natural pesticide. It is well adapted and domesticated to the humid tropics due to having been cultivated for the fresh consumption of its white, slightly sweet roots. However, little importance has been given to its seeds, stem and leaves toxic properties. Currently, toxicity is known to be more highly concentrated in seeds and, tests on living beings have shown toxicity to be present on cells *in vitro* (Estrella-Parra et al., 2014), humans (Sorensen, 1996; Yu et al., 2020), insects (Bejar et al., 2000), mites (Bejar et al., 2000)), fungi (Barrera-Necha et al., 2004), fishes (Sorensen, 1996), and viruses (Phrutivorapongkul et al., 2002).

This bean's seeds hold 20 iso-flavonoids, mainly rotenoids, with rotenone (Bejar et al., 2000), showing to be the most toxic one among them. This substance has been well known since the beginning of the 20th century on account of its insecticidal action (Roark, 1945a, 1942b, 1943c, 1943d, 1944e, 1945f) and, has already been widely used to control ants (*Dolichoderus bidens*) and the Mediterranean fruit fly in the USA (*Ceratitis capitata*) (De Moura and Schlichting, 2007). However, it was extracted from the roots of timbó (*Derris* species = *Lonchocarpus* species) and from *Tephrosia* (Dutta et al., 2019).

Rotenone is a lethal substance due to two reasons: (i) it inhibits electron transport in complex I of the mitochondrial respiratory chain, by blocking the ATP (Catteau et al., 2013) manufacture, and (ii) produces reactive oxygen species, causing oxidative stress (Mohammed et al., 2020).

There are some advantageous aspects in the use of yam bean seeds. First, the ease to be produced under Amazonian conditions, since it needs neither fertilization nor disease and pest control. Second, it has a moderate rotenone content, which ranges from 1.13 to 2.76 mg g^{-1} (Dutta et al., 2019). Third, the toxicity of oral rotenone in animals and humans is low. In rats, orally, the LD50 was 60 mg kg^{-1} body weight; intravenously, LD50 was 0.2 mg kg^{-1}, and intra-peritoneally, LD50 was 1.6 mg kg^{-1}. In humans, orally, LD50 ranges from 300 to 500 mg kg^{-1} body weight (Gonzalez-Coloma et al., 2010). Reports of accidental poisoning in humans have been observed when 40 to 80 fresh seeds are eaten (Yu et al., 2020; Fu and Wang, 2012). Only death was observed when 100 seeds were eaten (Narongchai et al., 2005).

The few *Pachyrhizus* spp. genotypes agronomically and physic-chemically (Silva et al., 2016) characterized (Vasconcelos et al., 2018) in the Amazon are found in the germplasm bank of the National Amazonian Research Institute (INPA), which conserves 64 progenies. Genetic variability was observed in them, which indicates that there could be genetic variability for toxicity. Therefore, it should be studied in this aspect, as well as tested about its effect on some important phytopathogens.

There are two important phytopathogens in the Amazon that cause death at any stage of growth. They attack the neck of the plant, which prevents nutrients from being taken to the aerial part and processed into photo-assimilates. These are the fungus *Sclerotium rolfsii* in cubiu (*Solanum sessiliflorum* Dunal) and the bacterium *Ralstonia solanacearum* in tomatoes. In both cases, control is difficult, and even in resistant varieties, some plants may die.

This work aimed to evaluate toxicity genetic variability in both mature and immature seeds using *Pheidole* species ants as well as test the toxicity of mature seeds on *S. rolfsii* and *R. solanacearum.*

MATERIALS AND METHODS

The genetic variability and toxicity experiments on *S. rolfsii* and *R. solanacearum* were carried out at the Phytopathology and Vegetable Breeding Laboratories, of the National Amazonian Research Institute (INPA).

Pachyrhizus toxicity genetic variability

Two experiments were carried out using mature and immature seeds flour. This flour was obtained from seeds milled in Wilye Type Micro Knife Mill. The extracts were prepared by flour maceration in filtrated water and then filtered using grade 4 filter paper. The final concentration was adjusted at 1:1000 (1 mg L^{-1}). The immature seeds, from 22 progenies, were obtained from green pods and, were green-colored as well. In contrast, mature seeds were obtained from dry pods and their color varied from brown to light beige.

Each experiment followed the completely randomized design with three repetitions. One Petri plate (diameter=10 cm) per experimental unit. Each plate was filled with 15 ml of extract then 20 ants were put in it.

Ants (*Pheidole* species) were picked up using plastic cups with sausage slices (Estrella Alimentos ®) outside buildings in INPA/Campus 3. After 0.5 to 1 h cups were taken up to the laboratory. Toxicity was assessed by counting the dead ants every 10 min for 60 min. Lethality percentage was calculated as [1-SN/TN] × 100, where SN= survivor number and TN= Total number of ants.

Toxicity on *S. rolfsii*

Six potato-dextrose-agar (PDA) culture media were prepared using 200 g potato, 10 g dextrose (Biotec Reagentes Analíticos) and 20 g agar (Agargel) to 1 L of distilled water. This was mixed with aqueous extracts from mature seeds of genotypes P8, P40 and P49. The final concentrations were for P8 1:500 (2g L^{-1}), P8 1:1000, P40 1:1000, P49 1: 1000, PDA (control), and Cabrio® Top (4g L^{-1}) (control).

This was followed by a completely randomized design with treatments in a 6×5 factorial scheme (six culture media) and five observation times (two, three, four, five and seven days) with three replications. The experimental unit was a Petri dish (9 cm) holding approximately 15 ml of solution. *S. rolfsii* sclerotia was placed in the center of the plate with a forceps. The mycelium diameter was measured on the fifth and seventh days.

Toxicity on *R. solanacearum*

Yeast-peptone-glucose (YPG) liquid culture was prepared using 5 g yeast extract (Kasvi), 5 g peptone (BD Difco), and 5 g glucose (Biotec Reagentes Analíticos) to 1 L of distilled water, pH=7 (Kpêmoua et al., 1996). This was then mixed with aqueous extracts of the genotype P40. The mixing was carried out so the final concentrations would be 1: 200, 1: 1000 and 1: 2000. Three *R. solanacearum* isolates, were used: V1 (Biovar 3), V13 (Biovar 2), and V15 (Biovar 1).

Thus, the experiment followed a completely randomized design, with treatments in a 5×3 factorial design (three extract concentrations + two controls) × (three *R. solanacearum* isolates) with 24 repetitions. The experimental unit was a 96-well Elisa microtiter plate. The two controls mentioned earlier were YPG+Tetracycline® (240 mg L^{-1}) and YPG by itself. 50 µl of each solution were placed in each well with the aid of a micropipette and the bacterium *R. solanacearum* was added. Absorbance was assessed through the iMark Microplate Reader spectrophotometer, every 24 h for three days.

Statistical analyses

All data were submitted to analysis of variance to determine the significant effect of the extracts. The averages of the treatments were compared using the Duncan test ($P<0.05$). The SAS 9.0 software, PROC GLM procedure was used (SAS Institute, Cary, NC). To observe the genetic diversity of yam bean toxicity, standardized averages of immature seed toxicity in *Pheidole* were used to construct a dendogram. This standardization was done for each time (10 to 60 min). For this purpose, genotypes were clustered based on Euclidean distances (bootstrap=1,000 samples) using unweighted pair group method with arithmetic mean (UPGMA) method. The Darwin 6.0 software was used (Perrier and Jacquemoud-Collet, 2006).

RESULTS AND DISCUSSION

Many rotenone and rotenoid toxicity tests on insects have been carried out since the early 20th century (Roark, 1945a, 1942b, 1943c, 1943d, 1944e, 1945f). These toxins were extracted mainly from the *Derris* genera. However, the rotenoids from *Pachyrhizus* sub-species were not tested sufficiently on plant's pests and diseases. In this work, the toxicity genetic variability of genotypes *Pachyrhizus* was assessed on *Pheidole* spp. ants, as well as, the effect of *Pachyrhizus* extracts over the fungus *S. rolfsii* and the bacterium *R. solanacearum*.

Yam bean toxicity genetic variability

Usually, toxicity tests have been carried out on several species such as the arthropod *Artemia salina*, zooplankton *Daphnia magna*, the worm *Eisenia andrei* and the collembola *Folsomia candida* (Danabas et al., 2020; Bandeira et al., 2020). There are still other species being used to determine chronic or acute toxicity in different environments. Yet, these methods need that these species be bred. In this work, *Pheidole* spp. ants were chosen for two reasons. First, because they are abundant in the Amazon region and are easy to capture. Second, they are abundant around the world and their control is difficult (Ali and Ali, 2020).

The results showed the mature seeds to present 100% lethality on *Pheidole* spp. after 10 min of treatment (Table 1). However, immature seeds had approximately the same lethality after 60 min. This indicates mature seeds to be six times more toxic (Tables 2 and 3).

The analysis of variance showed a significant effect for genotypes, which indicates there to be toxicity genetic variability in immature seeds at 10 to 60 min (Table 1). This result was observed in a dendogram (Figure 1). With 50% of dissimilarity, only two clusters were observed, but with 20% of dissimilarity, six clusters were observed. P9, P58, P7, P10, P61 and P28 showed no genetic variability for toxicity. However, water, P5 and P50 are a distant cluster from others. All these results indicate that the most toxic genotypes could be selected following 10 min.

Average lethality of immature seeds at 10 min of exposure ranged from 5.5 to 100.0% (Table 2). This indicates there would be genetic variability for rotenone or other toxic substances. P9, P58, P7, P10, P61 and P28 showed to be the most toxic genotypes all with 100% lethality. Conversely, P50 (5.5%) and P5 (8.5%) were the least toxic ones. Thus, P50 and P5 could be edible as long as they are cooked (Bidwell, 2020), dried or roasted (Catteau et al., 2013), or soaked by changing the water as it is usually in lupine (*Lupinus* spp.) preparation.

The analysis of variance also showed significance for the 'extracts versus control' contrast (Table 1). It suggests immature seeds to have a toxic action. This toxicity was observed on humans who had eaten 40 to 100 cooked

Table 1. Analysis of variance of the lethality of extracts of immature, *Pachyrhizus* seeds (1: 1000) on ants (*Pheidole* species).

Source of variation	DF †	Mean square (%)[2]					
		10 min	20 min	30 min	40 min	50 min	60 min
Treatments	22	6461.82**	6342.57**	6180.16**	5587.52**	4630.94**	3763.66**
Genotype (G)	21	3121.70**	2000.80**	1231.22**	1021.81**	742.50**	494.90**
G vs. Water	1	76604.54**	97519.80**	110108.10**	101467.50**	86288.48***	72408.18**
Error	88	197.66	231.72	239.29	308.87	347.34	430.10
Total	110						
CV ‡ (%)		33.92	28.25	25.10	26.33	26.17	28.00
General average		41.44	53.87	61.62	66.73	71.21	74.06
Genotype average		63.13	78.34	87.62	91.69	94.50	95.15
Water average		9.63	17.98	23.48	30.12	37.45	43.13

*, **, ns: significant at P<.05, P<.01 and non-significant by F test, respectively. †Degrees of free. ‡CV: Coefficient of variation.
Source: Authors (2022)

Table 2. Lethality averages of immature *Pachyrhizus* seeds for ants (*Pheidole* species) *in vitro* following 10 to 60 min of exposure.

Genotype	Lethality (%)					
	10 min†	20 min†	30 min†	40 min†	50 min†	60 min†
P5	8.5^{g}	25.76^{ef}	48.4^{b}	54.5^{b}	63.1^{bc}	71.7^{b}
P7	100.0^{a}	100.0^{a}	100.0^{a}	100.0^{a}	100.0^{a}	100.0^{a}
P8	65.7^{d}	80.9^{abc}	93.6^{a}	95.3^{a}	100.0^{a}	100.0^{a}
P9	100.0^{a}	100.0^{a}	100.0^{a}	100.0^{a}	100.0^{a}	100.0^{a}
P10	100.0^{a}	100.0^{a}	100.0^{a}	100.0^{a}	100.0^{a}	100.0^{a}
P17	30.4^{efg}	45.7^{de}	83.8^{a}	97.7^{a}	100.0^{a}	100.0^{a}
P20	66.8^{cd}	90.4^{ab}	95.2^{a}	97.6^{a}	100.0^{a}	100.0^{a}
P28	100.0^{a}	100.0^{a}	100.0^{a}	100.0^{a}	100.0^{a}	100.0^{a}
P39	53.3^{de}	76.6^{abc}	90.0^{a}	93.3^{a}	100.0^{a}	100.0^{a}
P40	64.9^{d}	93.0^{ab}	95.4^{a}	97.6^{a}	100.0^{a}	100.0^{a}
P41	24.2^{fg}	56.7^{cd}	87.7^{a}	100.0^{a}	100.0^{a}	100.0^{a}
P42	94.2^{ab}	94.2^{ab}	98.1^{a}	100.0^{a}	100.0^{a}	100.0^{a}
P48	29.6^{efg}	68.5^{cd}	80.4^{a}	82.7^{a}	87.7^{ab}	87.7^{a}
P49	52.3^{de}	76.1^{abc}	85.7^{a}	90.4^{a}	90.4^{ab}	90.4^{a}
P50	5.5^{g}	5.5^{f}	12.96^{c}	22.2^{c}	34.2^{c}	45.8^{b}
P51	73.7^{cd}	94.4^{ab}	96.4^{a}	98.2^{a}	100.0^{a}	100.0^{a}
P52	54.0^{de}	78.1^{abc}	88.9^{a}	94.5^{a}	97.6^{a}	97.6^{a}
P58	100.0^{a}	100.0^{a}	100.0^{a}	100.0^{a}	100.0^{a}	100.0^{a}
P59	26.1^{fg}	61.9^{cd}	83.3^{a}	92.8^{a}	100.0^{a}	100.0^{a}
P61	100.0^{a}	100.0^{a}	100.0^{a}	100.0^{a}	100.0^{a}	100.0^{a}
P62	48.7^{def}	78.3^{abc}	87.5^{a}	100.0^{a}	100.0^{a}	100.0^{a}
P63	90.3^{abc}	96.9^{ab}	100.0^{a}	100.0^{a}	100.0^{a}	100.0^{a}
Water	9.6^{g}	17.9^{f}	23.5^{c}	30.1^{bc}	37.4^{c}	43.1^{b}
Average	60.77	75.68	84.81	88.99	91.75	92.88

†Different letters in the column state a significant difference in averages by the Duncan test (P <0.05).
Source: Authors (2022)

Pachyrhizus immature seeds (Yu et al., 2020; Narongchai et al., 2005; Silva et al., 2016). They had diarrhea, dyspnea, unconsciousness or death (Silva et al., 2016).

Mature seeds presented no significant difference on their genotype-induced lethality following 10 min (Table 3). Yet, a difference was detected between genotypes and

Table 3. Averages of lethality of extracts from mature *Pachyrhizus* seeds on ants (*Pheidole* sp.) following 10 min of exposure.

Genotype	Lethality† (%)	Genotype	Lethality † (%)	Genotype	Lethality† (%)
P5	100.0[a]	P40	100.0[a]	P52	100.0[a]
P7	100.0[a]	P41	100.0[a]	P58	100.0[a]
P8	100.0[a]	P42	100.0[a]	P59	100.0[a]
P9	100.0[a]	P48	100.0[a]	P61	100.0[a]
P17	100.0[a]	P49	100.0[a]	P62	100.0[a]
P20	100.0[a]	P50	100.0[a]	P63	99.3[a]
P28	100.0[a]	P51	100.0[a]	Água	27.0[b]
P39	100.0[a]				

†Different letters in the column state a significant difference in averages by the Duncan test (P <0.05).
Source: Authors (2022)

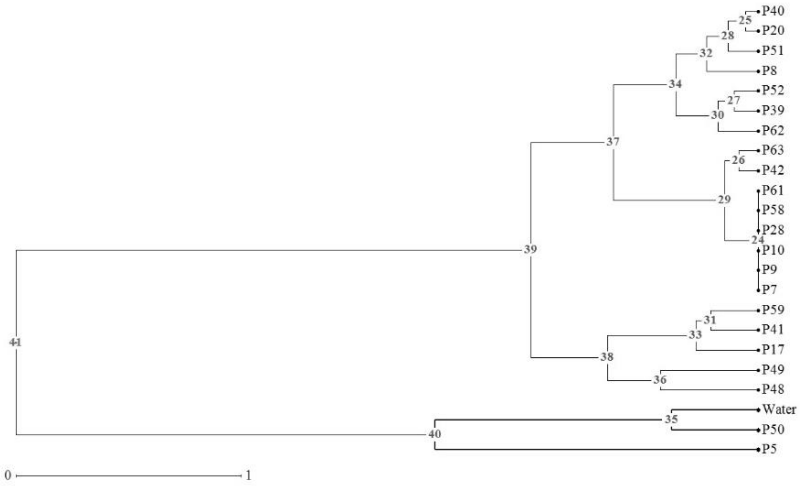

Figure 1. Dendogram based on Euclidean distances (bootstrap = 1,000 samples) and unweighted pair group method with arithmetic mean (UPGMA) showing the genetic variability of *Pachyrhizus* toxicity in ants (*Pheidole* species).
Source: Authors (2022)

control. Lethality averages for genotypes and control were 100 and 27%, respectively. This indicates all these genotypes to bear high toxicity.

Pachyrhizus seeds bear isoflavonoids and coumarins (Jiménez-Martínez et al., 2003). Rotenone and pachyrrhizin are each group's most toxic ones. Thus, the genetic variability can be for these substances, one of them or their interaction.

S. rolfsii

The analysis of variance (Table 4) and Duncan test detected a significant effect of the treatments on the *S. rolfsii* growth. The mycelial diameter after five days ranged between 20 and 60 mm in culture media with *Pachyrhizus* seed extracts (1:1000= 0.1%). Whereas mycelial diameter was 75 and 0 mm in PDA and PDA+Cabrio® Top controls. Thus, *Pachyrhizus* extracts reduced by 73% the mycelial growth.

P40 extract, at 1: 1000 concentration inhibited mycelial growth by 73%, following five days of cultivation (Table 5). P8 extracts at concentrations 1: 500 and 1: 1000 reduced mycelial growth by 32.6 and 48.9%. This shows the less concentrated extract to bear greater toxicity.

This conclusion is reinforced when compared with the *Pachyrhizus* extract concentration used in *Sclerotium cepivorum*, which was 5% (=1:20) (Lautié et al., 2013). In that case, the mycelial growth was reduced by 60% and the sclerotia production was totally inhibited. As a result, field tests could be performed using concentrations of 1: 1000 or even more diluted ones.

Table 4. *Sclerotium rolfsii* mycelium diameter analysis of variance showing significance of treatments (extracts) and days of evaluation (2^{nd}, 3^{th}, 4^{th}, 5^{th} and 7^{th}) for this trait are shown.

Source of variation	DF	Mean square of mycelial diameter (mm^{2} ‡)
Treatments (T)	5	2582.75**
Error 1	5	362. $86^{n.s}$
Days (D)	4	1966.60**
Error 2	4	$16.35^{n.s}$
T x D	18	941.38**
Error 3	97	108.39
Total	134	
Average		46.61
Coefficient of variation (%)		22.33

*, **, ns: significant at P<0.05, a P<0.01 and no significant by F test. †Degrees of free.
Source: Authors (2022)

Table 5. Mycelia diameter averages of *Sclerotium rolfsii* of treatments (*Pachyrhizus* extracts and controls) fifth and seventh day after inoculation.

Treatments/Concentrations	Mean square of mycelial diameter $(mm)^{2}$	
	Fifth day	Seventh day
PDA + P8 extract (1:500)	50.52 c	85.84 a
PDA + P49 extract (1:1000) ‡	59.89 b	85.85 a
PDA + P8 extract (1:1000)	38.30 d	75.09 b
PDA + P40 extract (1:1000)	20.28 e	49.38 c
PDA¶	74.97 a	85.84 a
PDA + Cabrio® Top (0,4 $g.L^{-1}$)	0.00 f	0.00 d

†Different letters in the column state a significant difference in averages by the Duncan test (P <0.05). ‡Imature *Pachyrhizus* seeds. ¶ Potato-dextrose-agar.
Source: Authors (2022)

On the other hand, Cabrio® Top was shown to have inhibited the mycelium growth. Thus, its use could be recommended to control this fungus. Even though there is no recommendation for doing so. Cabrio® Top is an agricultural pesticide indicated against various fungal diseases. Its composition holds two pesticides: (i) pyraclostrobin which inhibits the mitochondrial respiratory chain, and (ii) metiram that reacts non-specifically with fungus sulfhydryl enzymes (Registration 01303, from the Ministry of Agriculture, Livestock and Supply, Brazil).

Rotenone and Cabrio® Top bear some similarities in action mechanism (Catteau et al., 2013). Both prevent the mitochondrial respiratory chain and the production of ATP in the fungus. Thus, *Pachyrhizus* seeds could be used to control this soil fungus. They could be applied in the pit as extracts or powder prior to placing the seedlings.

R. solanacearum

The best way to assess bacterial multiplication is by absorbing light. To standardize the absorbance readings, decision was made to estimate the difference between the 48 and 24 h ones (Table 6). The results showed the absorbance differences for the extracts to have ranged from 0.29 to 0.51. In controls they were 0.06 (Tetracycline) and 0.68 (without extracts and no Tetracycline). This indicates these extracts to have reduced the multiplication of *R. solanacearum* by 17 to 39%. The extract holding the concentration of 1: 200 showed to be the one which exerted the most toxic effect for *R. solanacearum*.

Likewise, biovar 3 showed the largest difference in absorbance (0.52), suggesting it to be the most virulent. In contrast, biovar 1 showed the smallest difference in absorbance, which indicates it to be less virulent.

It is very difficult to cultivate tomato in the Amazon region, mainly due to the *R. solanacearum*. Yet, there was the singular case of the success of tomato production in Central Amazon. It was carried out at the Agro-industrial Adventist Institute in the 1970s and showed to yield up to 53 $t\ ha^{-1}$ (Prance, 1989). According to Dr. Noda, methyl bromide was used, in this institute to sterilize the soil (Oliveira, 2015). But this substance was

Table 6. Absorbance difference averages of *Ralstonia solanacearum* multiplication cultivated with *Pachyrhizu*s extracts *in vitro*.

Treatments	Absorbance difference between 24 and 48h†	Biovar	Absorbance difference between 24 and 48h†
PYG+ P40 extract 1:200	0.29[d]	Biovar 3	0.52[a]
PYG + P40 extract 1:1000	0.45[c]	Biovar 2	0.37[b]
PYG + P40 extract 1:2000	0.51[b]	Biovar 1	0.29[c]
PYG ‡	0.68[a]		
PYG +Tetracycline 240 mg.L^{-1}	0.06[e]		

†Different letters in the column state a significant difference in averages by the Duncan test (P <0.05). ‡Peptone-Yeast-Glicose. Source: Authors (2022)

prohibited because it was harmful to human health and the environment.

Therefore, *Pachyrhizus* seeds can be used as bactericide and insecticide in tomato cultivation mainly to control *R. solanacearum* and the mole cricket (*Gryllotalpa brachyptera*), which seems to be its vector.

Conclusions

There is genetic variability for toxicity in *Pachyrhizus* spp. It opens the possibility of screening all germplasm of this genus. The most toxic genotypes can be selected and included in a genetic improvement program. The aim of which should be to maximize seed and rotenoids yield.

Pachyrhizus seeds control *R. solanacearum* and *S. rolfsii*. Thus, *Pachyrhizus*-based products can be used for preventing soil diseases. Nevertheless, some novel methods, for treating the soil through the use of these biopesticides, will have to be developed.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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