

Managing Soybean Rust: Host Resistance, and Chemical Control

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Introduction

The identification of Asian soybean rust in Paraguay in 2001 (Morel and Yorinori, 2002) and its spread to over 90% of the soybean production in Brazil through the 2003 season has heightened the awareness of this disease. The rapid spread of *P. pachyrhizi* and the potential for severe yield losses makes this the most destructive foliar disease of soybean. Soybean rust, if introduced into the U.S., could have a major impact on both total soybean production and production costs.

The focus of this presentation will be on managing the disease using both host resistance and fungicides. But, to manage the disease we need to understand a little about its biology. Asian soybean rust, caused by *Phakopsora pachyrhizi*, is an obligate parasite; it needs living tissue to survive. Urediniospores are the main spore stage. Teliospores and basidiospores have been produced but are not part of the disease cycle, since there is no known alternate host for basidiospores to infect. The pathogen penetrates directly, unlike the pathogens that cause rusts of wheat and corn. Host surface features do not influence the infection process; stomata are not important in infection. Infection can occur with as few as 12 hours of moisture. The infection cycle is short; new infections can produce urediniospores within 5 to 7 days. Most parts of the soybean plant are infected, including the coleoptiles, leaves, petioles, stems and seedpods. Disease symptoms are primarily observed in the lower canopy until flowering. After flowering the symptoms are noticeable in the mid and upper canopy, where it causes rapid defoliation and yield loss. Yield loss can be due to pod abortion, and smaller and fewer seed. Protein is decreased but oil is not. One important feature is that the symptoms appear and spread rapidly after flowering. Spore production and lesion numbers increase after flowering, thus host age is important in the development of the epidemic as well as in evaluating germplasm for resistance.

The pathogen will not over winter in the Midwest. Like leaf or stem rust of wheat and the rusts of corn, soybean rust spores are wind blown and will most likely blow up from the south. *P. pachyrhizi* has a very broad host range and can infect over 90 species of plants in many genera, including Kudzu. Besides Kudzu, there may be other legume hosts found in areas where the fungus will over winter.

Resistance

Specific resistance and physiological specialization. Specific resistance to *P. pachyrhizi* is known and four single dominant genes have been identified as *Rpp1*

(McLean and Byth, 1980), *Rpp*₂ (Bromfield and Hartwig, 1980b), *Rpp*₃ (Bromfield and Hartwig, 1980a; Bromfield and Hartwig, 1980b; Hartwig and Bromfield, 1983), and *Rpp*₄ (Hartwig, 1986). These four genes condition resistance to a limited set of rust isolates (Table 1). *Rpp*₁ was described as having an immune reaction when inoculated with a few isolates, including India 73-1. Inoculation of most rust isolates on *Rpp*₁ or the other genes produces a resistant red-brown (RB) lesion with no or sparsely sporulating uredinia. The RB lesion type is considered to be a resistant lesion type when compared to a fully susceptible TAN lesion (Fig. 1). However, pustules with an RB reaction can produce urediniospores.

Single gene resistance has not been durable and the usefulness of the single genes was lost soon after the sources were identified. For example, Komata was identified in germplasm evaluations done during 1961-1963 (Bromfield, 1984). By 1966, susceptible lesions were found on plants of Komata in field trails, and by the mid 1970's the line was not considered to be a useful source of resistance (Kochman, 1977). Similarly, the accession PI230970 was identified as resistant in field evaluations in 1971-1973, but by 1976 a few susceptible lesions were observed on plants in the field. In 1978, most of the lesions found on plants in the field were of the susceptible TAN type (Bromfield, 1984). The resistance in Ankur, identified in the early 1970's (Singh et al., 1975) was lost in the late 1970's (Bromfield, 1984), providing another example of the diversity in virulence seen in *P. pachyrhizi* populations overcoming single gene resistance. Only Bing Nang, the source of the *Rpp*₄ gene, has not been reported to be defeated, although our observations both in the field in Paraguay and greenhouse inoculation tests indicate that it is susceptible to some isolates.

Physiological races of *P. pachyrhizi* were first described in 1966 when a set of nine single urediniospore isolates were inoculated onto six soybean and five legume accessions (Lin, 1966). The reactions of the nine isolates were similar on all six of the soybean genotypes, but six pathotypes were identified based upon their reactions on the legume accessions. The first example of virulence diversity on soybean cultivars was described in Queensland, Australia (McLean and Byth, 1976) where one rust isolate was found to be virulent on the cultivar 'Willis' but avirulent on the accession PI 200492, while a second isolate was virulent on both soybean genotypes. Several other studies have also shown considerable variation in virulence among isolates from the same field, as well as isolates collected from wide geographical areas (Poonpolgul and Surin, 1985; Shin and Tschanz, 1986). The summary on virulence is that it is diverse and complex. Not only is there physiological specialization in the interaction with soybean but it is also known to occur within the other legume species as well.

Partial resistance. Partial resistance, or rate reducing resistance, is also known in soybean (Wang and Hartman, 1992). Lines with partial resistance in field evaluations were rated as moderately resistant since fewer lesions developed on plants throughout the season. In greenhouse studies, host-pathogen combinations that resulted in RB reaction types tended to have longer latent periods, lower rates of increase in pustule number over time, and smaller lesions compared with susceptible interactions that resulted in a TAN reaction type (Bromfield et al., 1980; Marchetti et al., 1975). Identification and utilization of partial resistance in breeding programs has been limited. The evaluation methods may be time consuming and difficult to incorporate into breeding programs and therefore

limited to use with advanced generations. These difficulties, at least in part, led to the development of a strategy to select genotypes with what was defined as having tolerance or yield stability despite being heavily infected with *P. pachyrhizi* (Hartman, 1995; Wang and Hartman, 1992).

Yield stability. Yield stability, or tolerance, refers to the strategy of selecting genotypes with high yield potential and less yield loss from soybean rust. Screening for yield stability to soybean rust was started at the Asian Vegetable Research and Development Center (Hartman, 1995), where yields from paired plots, with and without the fungicide Dithane M-45 applied every 2 weeks, were compared for losses due to rust. High yielding genotypes with less yield loss under severe rust conditions were considered to be tolerant. Rust development rates and estimates of rust severity on foliage were not correlated with yield loss in tolerant materials. Using fungicide protected plots as yield checks, tolerant lines from breeding populations were identified without having to take notes on rust severity (Hartman, 1995). Cultivars with yield stability may have some partial resistance that was not characterized or selected for in the breeding program.

Current Research

Since the report of soybean rust in Hawaii in 1994, the USDA-ARS has renewed its support for soybean rust research in the U.S. With support coming from the United Soybean Board, part of the research focus has been to identify resistant germplasm. There are over 16,000 soybean accessions in the USDA Germplasm Collection located at the University of Illinois. These soybean accessions, along with commercial and public cultivars grown in the U.S., are being evaluated for resistance to *P. pachyrhizi* in the USDA-ARS FDWSRU Biosafety Level 3 Containment Greenhouses at Fort Detrick, MD. The evaluations are done on seedlings using a mixture of isolates from Africa, Asia and South America. From the 6000 soybean accessions screened to date, fewer than 100 have been identified as having some level of resistance. None of the U.S. commercial cultivars evaluated were found to be resistant to the mixed inoculum. The soybean accessions showing some level of resistance will be further evaluated using individual isolates to detect race specific and/or partial resistance. They also will be planted in field trials in Brazil, Paraguay, China, Thailand, South Africa and Zimbabwe to be evaluated for adult plant resistance. Additional research is being conducted to determine the best way to evaluate partial resistance and yield stability. Besides soybean, about 1,000 *G. soja* accessions will be screened along with some of the perennial *Glycine* spp. previously reported as having resistance (Hartman et al., 1992). As sources of resistance are identified, crosses will be made to incorporate these resistance traits into adapted backgrounds for commercial use.

Fungicides

In the near future the primary tool in the control of soybean rust will be the use of fungicides. Cultural practices have not been shown to be effective in control of the pathogen; recommendations were inconsistent and varied by location. The most effective practice was avoidance or practices that maximized yields in the absence of the disease.

Fungicide Efficacy. Many fungicides have been evaluated to control soybean rust. Early research from Asia indicated that mancozeb was effective (Hartman *et al.*, 1992). Other compounds available at the time were compared to mancozeb and were effective, but results varied by test (Table 2). More recently, fungicide trials in India (Patil and Anahosur, 1998) and Southern Africa (Levy *et al.*, 2002) have identified several triazole compounds and triazole mixes. The most recent trials in Africa and South America have identified additional triazoles, (eg. tebuconazole and tetraconazole), as well as several strobularins and strobularin mixes including azoxystrobin, pyraclostrobin, pyraclostrobin + boscalid and trifloxystrobin + propiconazole (Miles *et al.*, 2003b). Other compounds have been identified that reduce disease severity, but yield protection has been inconsistent. Further efficacy trials are continuing in both Africa and South America to identify additional products.

Labeled and Section 18 compounds. There are a total of three fungicides that are registered for use on soybean, labeled for soybean rust and are commercially viable (Table 3). These fungicides are Quadris®, Bravo®, and Echo®. Quadris is an azoxystrobin; Bravo and Echo are both chlorothalonils. There has been a Section 18 Emergency Exemption request for seven compounds or mixtures of compounds submitted to the EPA by the Departments of Agriculture of Minnesota and South Dakota (<http://plantsci.sdstate.edu/draperm/SoybeanRustSection18>). Not included on any of the lists are the sulfur, lime, elemental compounds, various oils, and other organic products that are not viable in a large commercial operation.

Timing and Number of Applications. The most recent experiments evaluating the timing and number of applications for chemical control of soybean rust have come from Zimbabwe and South Africa (Levy *et al.*, 2002). Early experiments evaluated the number of applications needed to protect the crop (Fig. 2). Treatments differed by date of first application and all treatments, except the non-protected control, received the last, or 108 days after planting (DAP), application. Applications were made at 20-day intervals starting at 28 DAP for the five-application treatment. There were no differences in yields when fungicide application started 28 DAP (five-applications) or 48 DAP (four-applications). There was a slight yield loss when the first spray was applied at 68 DAP (three-applications). Delaying fungicide application until 88 (two-applications) and 108 DAP (single late application) resulted in significant yield losses. Flowering of both cultivars occurred between 50 and 60 DAP. When fungicides were applied during the vegetative growth stages (28 DAP), yields did not increase compared to applications that protected the crop from flowering through grain fill, 48 and 68 DAP.

Experiments that evaluated the timing of applications in post flowering soybean were completed using two cultivars, Sonata and Soprano, treated with 50 g flusilazole + 100 g carbendazim (Punch Xtra®) in single applications at either 50, 60, 70, 80 or 90 DAP, and two-application treatments at 50+70 dap, 60+80 dap or 70+90 DAP. A three-application treatment (50+70+90 DAP) simulated the recommendation being made to farmers, and a four-application treatment was included to provide total rust control. Data indicate that most single applications did not protect yield (Fig. 3). However, if properly timed, a single application has been seen to protect yields when compared to treatments with two or more applications. The timing of the application was critical, as applications

10 days earlier or later showed significant yield losses. All treatments with two-applications had yields similar to treatments with three or four applications. Late applications had slightly less protection in Soprano, the indeterminate cultivar.

Recommendations. In Southern Africa, the recommendation was made to use a program with two or three fungicide applications (Levy *et al.*, 2002). Three applications were considered necessary in high disease situations, while two applications were recommended when disease severities were light. For best yield protection the first application was recommended at 50 DAP, at or just ahead of flowering. Subsequent applications 20 days apart were sufficient to control the disease. These recommendations were made in an attempt to limit the exposure of the crop to the disease due to difficulties in obtaining exact timing of a single application. This recommendation was supported by limited data from Paraguay where a single application at flowering had less yield protection than two applications, one at flowering with the second 20 days later (Miles, unpublished data).

The production practices in Brazil are changing from a single fungicide application at growth stage R3 to R5 used to protect against late season diseases to a two-application program with the first application at R3. Their recommendations differ from the recommendations in Southern Africa. As the scenario plays out in South America next year we will learn more.

The number and timing of applications are critical for the control of soybean rust. The most efficient were applications during early reproductive growth that allowed protection through to crop maturity. The exact number of applications will depend on the length of the reproductive phase of the crop, duration of the compound and severity of the epidemic. Fungicide applications in early vegetative stages, although effective in reducing disease severity, have not been shown to be effective in protecting yield.

Application methods. One of the more critical application challenges for protecting the soybean crop from yield losses due to soybean rust is to penetrate the canopy and deliver the fungicide into the middle third of the canopy. Fungicides are not used in most soybean production areas, so little work has been done to develop fungicide application programs for the crop. Both aerial and ground applications are used in South America. Multiple application methods are being used in Southern Africa, with the most effective methods being those where penetration and canopy coverage are the greatest. Examples of effective methods include air assist and high pressure lateral discharge equipment, increased pressure delivery and increased water volume per hectare.

Currently, there is a multi-state project to evaluate high and low volume application in aerial and ground systems using predominantly 30-inch row spacing. Within the ground application program are different nozzle types that would be available on a commercial basis today. Included are the flat fan nozzle that would be used for Round up® application, as well as air induction and twin jet nozzles. Preliminary data from both aerial and ground application show the need for high volume (10 gal. aerial and 20 gal. ground applied) to penetrate the canopy into the middle third. There is need for additional experimentation before a fungicide application method can be developed to economically protect the soybean crop.

Future Management of Soybean Rust

Control of soybean rust can be accomplished through utilization of fungicides (Miles et al., 2003a). To minimize the use of fungicides and to reduce the production risk with misapplied fungicides, producers will need to avoid planting the most susceptible cultivars. The easy solution; single gene resistance, may not be part of the overall picture for control. It may be possible that with the right combination of single genes, they could play a role in an overall resistance management program. Partial resistance may also be effective in that it will slow down the epidemic, decreasing the build up of rust. Fewer spores produced over time could effectively reduce the need for multiple fungicide applications. Yield stability may be the most effective resistance mechanism. Cultivars with yield stability may have single gene resistance or partial resistance traits, but were selected due to yield performance with and without the disease.

Acknowledgements

We thank the United Soybean Board, St. Louis, MO, U.S.A., and several of the companies whose products were tested for financial support. A CSREES Critical Needs Research Grant and the USDA-ARS also funded portions of the research.

Names are necessary to report factually on available data; however, the USDA neither guarantees nor warrants the standard of the product, and the use of the name by the USDA implies no approval of the product to the exclusion of others that may also be suitable.

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Table 1. Named single genes, original sources and *Phakopsora pachyrhizi* isolates used in studies of the inheritance of resistance to soybean rust

Named single gene	Accession number and cultivar name of original source	<i>Phakopsora pachyrhizi</i> isolates ^a	
		Resistant reaction	Susceptible reaction
<i>Rpp</i> ₁	PI200492 Komata	IN 73-1 ^{bc}	TW 72-1, TW 80-2 (Hartwig and Bromfield, 1983; McLean and Byth, 1980) ^d
<i>Rpp</i> ₂	PI230970	AU 72-1 ^c , IN 73-1 ^c , PH 77-1 ^c , TW 72-1 ^c	TW 80-2 (Bromfield and Hartwig, 1980a; Hartwig and Bromfield, 1983; McLean and Byth, 1980) ^d
<i>Rpp</i> ₃	PI462312 Ankur	IN 73-1 ^c	TW 72-1, TW 80-2 (Hartwig and Bromfield, 1983) ^d
<i>Rpp</i> ₄	PI459025 Bing Nang	IN 73-1 ^c , TW 72-1 ^c , TW 80-2 ^c	(Hartwig, 1986) ^d

a. AU = Australia, IN = India, PH = Philippines, TW = Taiwan.

b. Immune reaction type.

c. Isolates used in original inheritance studies to examine segregation patterns.

d. Reference.

Table 2. Summary of fungicides evaluated for control of soybean rust caused by *Phakopsora pachyrhizi*

Active ingredient	Products evaluated	Country where test were done	Summary of application trials and recommendations in the literature	References
Triadimefon	Bayleton®	India, Japan, Philippines, Taiwan, Thailand	Protection inconsistent when compared to Dithane M45, although it was used as a control in yield loss studies. EDBC's appear to be more effective but in limited testing up to 33% yield increases were seen. First application at flowering, 10 to 20 day intervals.	Hartman <i>et al.</i> , 1992; Patil and Anahosur, 1998
Thiabendazole	Benlate®, Topsin M®	Thailand	Off registration in US, not as effective as Dithane M45, effective only when used with Plantvax, but no yield increase. Phytotoxic as a seed treatment.	Hartman <i>et al.</i> , 1992
Chlorothalonil	Bravo®, Echo®	Brazil, India, Paraguay	Limited data available yield protection similar to or less than Mancozeb. Not as effective as other compounds in some studies.	Hartman <i>et al.</i> , 1992; Miles <i>et al.</i> , 2003; Patil and Anahosur, 1998
Ethylenebisdithio-carbamates (EDBC)*	Dithane-M45®, Mancozeb, Manzate D®, Zineb®, Maneb®	Australia, China, India, Philippines, Paraguay, Taiwan	The EDBC products have been effective in controlling soybean rust when applied 7 to 21 days apart, with the first applications as early as three weeks after planting and as late as flowering. Not all studies showed control of yield increases.	Hartman <i>et al.</i> , 1992; Miles <i>et al.</i> , 2003
Oxycarboxin	Plantvax®	India, Taiwan, Thailand	Not as effective as Dithane M45 or Manzate D, did not always control rust, yield protection varied by study. Apply when lesions first appear, then at 7 day intervals.	Hartman <i>et al.</i> , 1992;
Hexaconazole	Contaf®	India	Effective in reducing disease and protecting yield, 25% yield increase in limited testing.	Patil and Anahosur, 1998
Propiconazole	Tilt®, Propimax®	Brazil, India, Paraguay	Effective in reducing disease and protecting yield, 33% yield increase in limited study. Two applications, 15 days apart, starting at flowering.	Miles <i>et al.</i> , 2003; Patil and Anahosur, 1998

Difenoconazole	Score®	India, South Africa, Zimbabwe	Yield protection varied by study, more effective than Mancozeb. Two or three applications needed, starting at flowering.	Levy <i>et al.</i> , 2002
Triadimenol	Shavit®	India, South Africa, Zimbabwe	Extremely effective in reducing disease incidence. Highest yielding treatment. Two or three applications needed, starting at flowering.	Patil and Anahosur, 1998
Flusilazole+ carbendazim	Punch Xtra®	South Africa, Zimbabwe	One of most effective fungicides in Africa. Two or three applications needed, starting at flowering	Levy <i>et al.</i> , 2002
Tebuconazole	Folicur ®	Paraguay, Zimbabwe	Limited data, yield protection variable by location within studies.	Levy <i>et al.</i> , 2002; Miles <i>et al.</i> , 2003
Azoxystrobin	Quadris®	Brazil, Paraguay	Limited data, good control but single, late application did not control rust or protect yield.	Miles <i>et al.</i> , 2003
Tetraconazole	Eminent®	Brazil, Paraguay	Limited data,	Miles <i>et al.</i> , 2003
Pyraclostrobin	Headline®	Paraguay	Limited data, good rust control with yield benefits	Miles <i>et al.</i> , 2003
Boscalid	Endura®	Paraguay	Limited data	Miles <i>et al.</i> , 2003
Pyraclostrobin + boscalid	Pristine®	Paraguay	Limited data, good rust control with yield benefits	Miles <i>et al.</i> , 2003
Trifloxystrobin + propiconazole	Stratego®	Paraguay	Limited data, good rust control with yield benefits	Miles <i>et al.</i> , 2003
Fenbuconazole	Enable®	Paraguay	Limited data	Miles <i>et al.</i> , 2003
Myclobutanil	Eagle®, Laredo®	Paraguay	Limited data	Miles <i>et al.</i> , 2003

Table 3. Fungicides registered for used on soybean, labeled for Asian soybean rust or on a Section 18 Emergency Exemption request.				
Compound	Product	Company	Registration status	
			Soybeans	Soybean rust
Azoxystrobin	Quadris®	Syngenta	Yes	Labeled
Chlorothalonil	Bravo®	Syngenta	Yes	Labeled
	Echo®	Sipcam Agro	Yes	
Myclobutanil	Laredo®	DAS		Section 18
Propiconazole	Tilt®	Syngenta		Section 18
	Propimax®	DAS		
	Bumper®	Makhteshim-Agan		
Pyraclostrobin	Headline ®	BASF		Section 18
Pyraclostrobin + boscalid	Pristine ®	BASF		Section 18
Tebuconazole	Folicur®	Bayer		Section 18
Tetraconazole	Eminent®	Sipcam Agro		Section 18
Trifloxystrobin + propiconazole	Stratego®	Bayer		Section 18

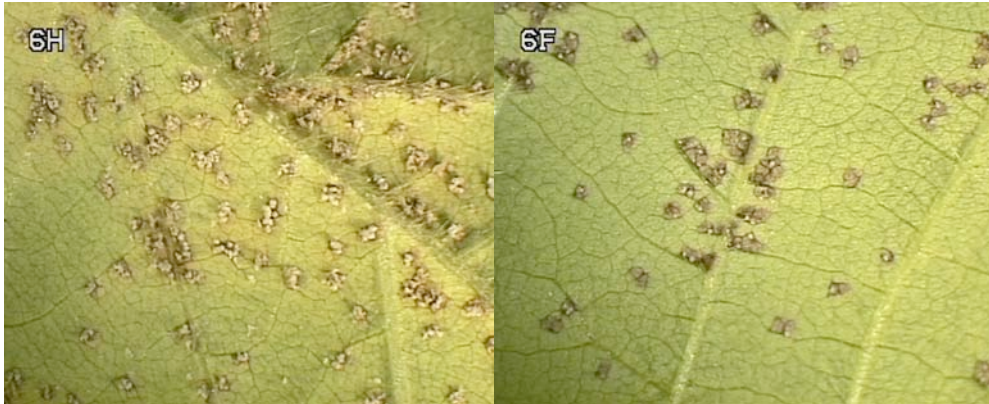


Figure 1. Soybean leaves infected by *Phakopsora pachyrhizi* on a (left) susceptible soybean with susceptible (TAN) lesions and (right) red brown (RB type) lesions.

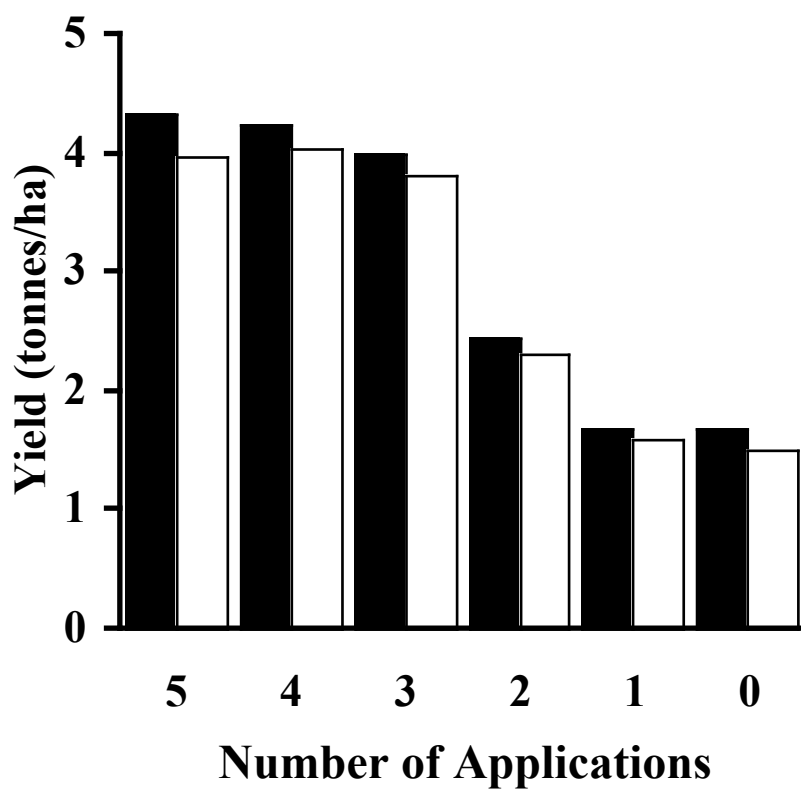


Figure 2. Kernel yield (t ha^{-1} , at 11% moisture content) of two soybean cvs ('Soprano': ■; 'Sonata': □) either sprayed with flusilazol + carbendazim, or left unsprayed at various dates after planting at the Rattray Arnold Research Station, Enterprise, Zimbabwe, in the 2000/2001 season (Levy *et al.*, 2002).

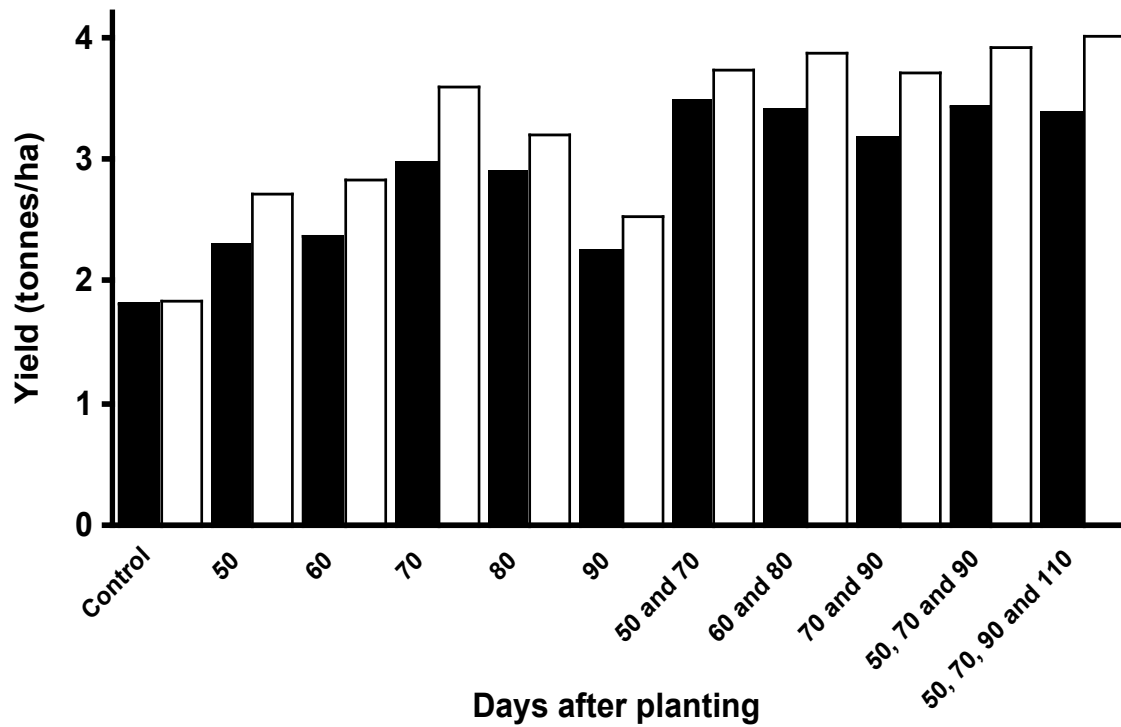


Figure 3. Kernel yield (t ha^{-1} , at 11% moisture content) of two soybean cvs ('Soprano': ■; 'Sonata': □) either sprayed with flusilazol + carbendazim, or left unsprayed at various dates after planting at the Rattray Arnold Research Station, Enterprise, Zimbabwe, in the 2000/2001 season (Levy *et al.*, 2002).