

EFFECTS OF FIPRONIL BAIT ON SUGARCANE YIELD IN OKINAWA, JAPAN

By

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**KEYWORDS: Fipronil Bait, Phytostimulatory,
Sorghum, Sugarcane Yield, Wireworm.**

Abstract

FIPRONIL bait (Prince[®] Bait, BASF Agro), newly registered as a sugarcane insecticide in 2006 in Japan, has been shown to reduce sugarcane wireworm, *Melanotus sakishimensis* and *M. okinawawesis* (Coleoptera: Elateridae), damage and increase sugarcane yield. In January 2008, a high sugarcane growth rate was observed in fipronil bait-treated areas in Minami-daitou Island. To confirm this high growth rate, we investigated sugarcane growth and yield in two fields on Minami-daitou Island from March to December 2008. Moreover, as we suspected that fipronil has a phytostimulatory effect that promotes sugarcane growth, we conducted a simple test using sorghum, also a gramineous C₄ crop and a species with a high degree of collinearity with sugarcane. Research on Minami-daitou Island showed that sugarcane growth in the fipronil bait-treated area was significantly higher than the areas treated with ethylthiodemeton granule insecticide (TD[®], Sankyou Agro) or isoxathion granule insecticide (TD[®] Ace, Sankyou Agro). Further, cane yield in the fipronil bait-treated area was also significantly increased as compared with that in the ethylthiodemeton granule and isoxathion granule insecticide-treated areas. Sorghum tests showed that the coleoptile length of sorghum seeds increased when treated with 0 to 10 ppm of fipronil, and then decreased for concentrations over 10 ppm. It was concluded that fipronil bait treatment in sugarcane fields can significantly increase both the growth and yield of sugarcane. Moreover, it was suggested that fipronil at low concentrations may have a phytostimulatory effect on sorghum seedlings.

Introduction

Fipronil (5-amino-1-[2,6-dichloro-4-(trifluoromethyl) phenyl]-4-[(trifluoromethyl) sulfinyl]-1*H*-pyrazole-3-carbonitrile) is a phenylpyrazole insecticide that interferes with GABA (γ -aminobutyric acid)-gated ion channels (Figure 1).

It blocks chloride ions influx by acting on GABA-gated chloride ion channels, and causes excessive neural excitation, paralysis, and eventual death (Gunasekara *et al.*, 2007). Since registration in 1996, fipronil has been widely used in many cropping systems around the world, including Japan, particularly in rice production.

In 2006, fipronil bait insecticide (0.5% active ingredient, Prince[®] Bait, BASF Agro) for sugarcane was registered in Japan. The formulation consists of only 0.5% fipronil and approximately 99.5% grain-based material, and differs from conventional sugarcane insecticides in its mode of action.

This insecticide attracts insects with the grain-based material, which leads to the uptake of fipronil by the insect. In addition, fipronil has systemic activity and can control insects such as boll weevils, rootworm and wireworm (Clive, 2006). Sugarcane wireworm, *Melanotus sakishimensis* and *M. okinawawesis* (Coleoptera: Elateridae), is a serious economic pest in Japan (Nagamine and Kinjo, 1979).

Tarora *et al.* (2007) demonstrated that Prince[®] Bait has an attractant effect and causes high wireworm mortality in laboratory tests. Moreover, the number of ratoon shoots was significantly higher in bait-treated fields than in fields treated with carbosulfan granule insecticide, a conventional insecticide used against wireworm in Japan, on Miyako Island.

However, their research differs from our research in terms of location, insecticide standard and planting season. They studied wireworm damage in three crops: summer planting, spring planting and at ratooning, but the yield research was performed only at spring planting.

As no fipronil insecticides for sugarcane were available in Japan before 2006, there are few reports on the effects of fipronil bait insecticide on sugarcane yield.

The present study was aimed at demonstrating the effects of this bait insecticide on sugarcane yield on Minami-daitou Island, its impact on pest insects, as well as its phytostimulatory effects.

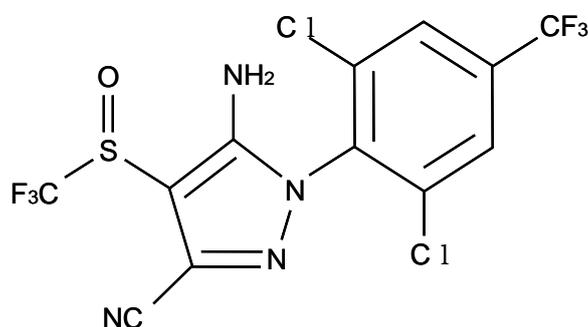


Fig. 1—Chemical structure of fipronil.

Materials and methods

Research on Minami-daitou Island

The research was conducted in two fields, A (1.3 ha) and B (1.6 ha), planted with *Saccharum* spp. cv. Ni26 on Minami-daitou Island in Okinawa.

Fields A and B were treated with fipronil bait, while the control areas in each field were treated with ethylthiodemeton granule insecticide (3.0% a.i., TD[®], Sankyou Agro) and isoxathion granule insecticide (2.0% a.i., TD[®] Ace, Sankyou Agro), respectively.

Ethylthiodemeton granule insecticide and isoxathion granule insecticide are both conventional insecticides used against wireworms in Okinawa. Field studies were conducted on March 21–22, April 12–13, July 12–13, October 8–9, and December 20–21, 2008.

From March 21–22 to October 8–9, growth analysis was conducted. Briefly, 3 blocks were selected in each insecticide-treated area, and the sub-stem length of the main stem, number of tillers and number of dead heart stalks per block were measured. In addition, the number of stalks was determined for an area of 7.0 × 1.5 m. Sub-stem length was defined as the length to the dewlap of the first leaf.

Data on the number of tillers, stalks and dead heart stalks were not recorded on October 8–9 in field A due to severe lodging. Yield sampling was conducted on December 20–21. A 2.2 × 1.5 m plot was cut in each insecticide-treated area, and the diameter of each stem, sub-stem length, and number of stalks per plot was measured.

The crown above the dewlap of the 5th leaf was pruned off, and the length and weight of each stem was measured. Sugarcane yield per 10 a was calculated from the number of stalks per 2.2 × 1.5 m plot and stem weight data. To evaluate sugarcane quality, sucrose content was analysed from 5 juice samples per plot using an HPLC system (LC-10A, Shimadzu). The data for each field were analysed using Student's t-test, with a value <0.05 considered as significantly different.

Phyostimulatory effect

Sorghum seeds (*Sorghum bicolor* cv. FS501) were used to assess potential phyostimulatory effects because sorghum has the same photosynthetic process as sugarcane and a high degree of collinearity exists between the two crops. Adjustment of the fipronil concentration was difficult as it has low water solubility. A suspension concentrate (SC) (5.0% a.i., Prince[®] Flowers, BASF Agro) was therefore used for the laboratory experiments. A 5.0% (50 000 ppm) fipronil SC was diluted to produce 10 concentrations (0, 1, 2.5, 5, 7.5, 10, 25, 50, 75, 100 ppm) in distilled water. A total of 10 seeds per treatment were placed in a vial (50 mL), and 50 mL of each fipronil concentration was added. All vials were capped and left to incubate for 2 h.

Seed samples (one seed per tube) and solutions (4 mL per tube) were then transferred to a liquid-filter paper culture medium in glass tubes. All tubes were covered by parafilm and maintained in the dark for 4 days at 30°C. Germination rate, rooting rate, coleoptile length and root length were measured.

Germination and rooting were considered to have occurred if the coleoptile and root were more than 1 mm in length. The experiment was replicated three times and means compared by Scheffe's test at $p < 0.05$.

Results

Research on Minami-daitou Island

Figures 2 and 3 show sugarcane growth from March to October, 2008, and Figures 4 and 5 are photos taken in March, 2008. In field A, sub-stem length in the fipronil-bait treated area was significantly longer than that in the ethylthiodemeton insecticide-treated area in March and April. In field B, sub-stem length in the fipronil-bait treated area was also significantly longer than that in the isoxathion-treated area in March, April and October.

The number of stalks peaked in March in all areas, except in the isoxathion-treated plots where it peaked in April, and decreased thereafter. This decrease was due to the death of non-productive tillers through competition.

Although the number of tillers peaked in April and thereafter decreased in all plots, no significant differences were detected between the fipronil bait- and conventional insecticide-treated plots.

The number of dead heart stalks in the fipronil bait-treated plots was lower than in the conventional insecticide-treated plots in March, April and October, but this difference was not apparent in July.

Table 1 shows the yield sampling results for December 2008. In the fipronil bait-treated plots, stem length and sub-stem length in field A, and stem length, diameter and weight in field-B were significantly higher than the values obtained in the conventional insecticide-treated plots.

Table 1—Effects of fipronil bait treatments on sugarcane yield and quality.

Field	Treatment	No. of stalks' (2.2×1.5m)	Stem length (cm)	Sub-stem length (cm)	Stem diameter (mm)	Stem weight (kg)	Sucrose %	Yield 10a-1 (t)
A	Fipronil	32	336.5 ± 84.8*	379.5 ± 98.1*	31.05 ± 3.56	1.59 ± 0.57	16.4 ± 3.5	15.45
	Ethylthio-demeton	20	292.5± 59.8	339.2± 69.6	30.88± 5.59	1.44 ± 0.60	18.2± 1.5	8.53
B	Fipronil	31	296.0± 44.2*	337.1 ± 49.1	32.81 ± 3.13*	1.51± 0.46*	18.7± 1.4	13.96
	Isoxathion	29	275.2±46.6	320.7±62.3	29.69±4.41	1.27±0.46	19.3±0.9	11.19

* $P < 0.05$.

There were no significant differences in sucrose content between fipronil treatments. These results show that fipronil bait treatment improved sugarcane growth and yield without reducing cane juice quality.

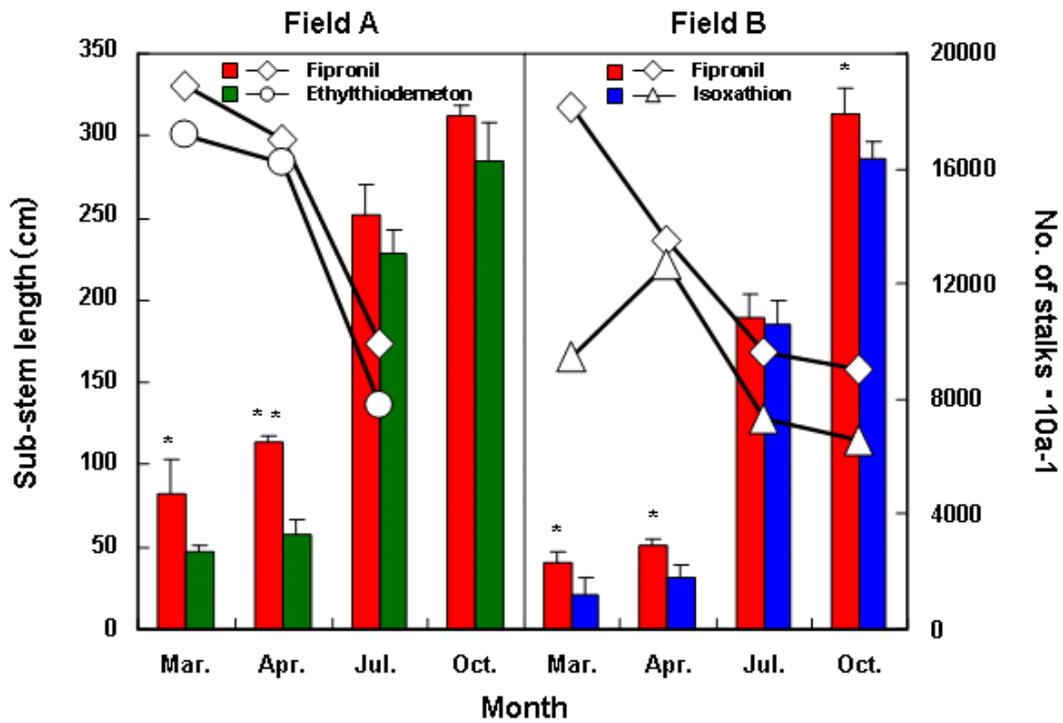


Fig. 2—Effects of fipronil bait treatments on the number of stalks and sub-stem length. Bar and line graphs show sub-stem length and number of stalks per 10 a, respectively. * $P < 0.05$, ** $P < 0.01$.

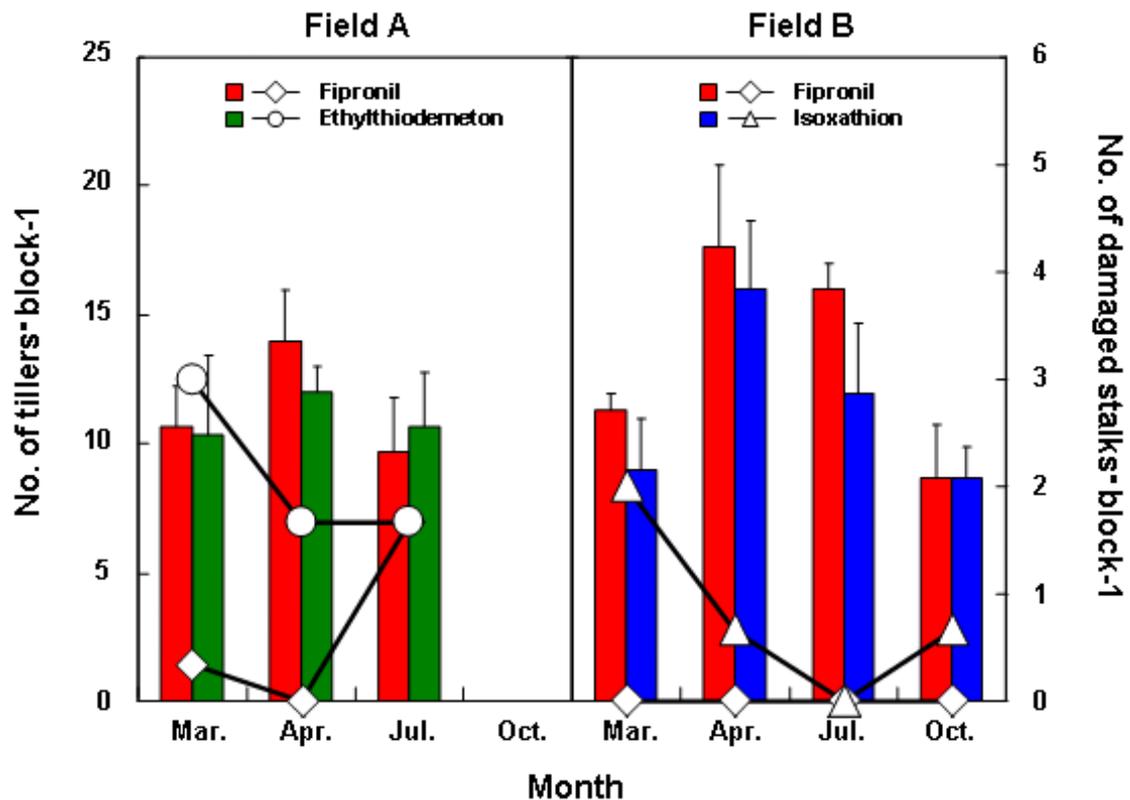


Fig. 3—Effects of fipronil bait treatments on the number of tillers and damaged stalks. Bar and line graphs show tiller number per block and number of damaged stalks per block, respectively.



Fig. 4—Field A in March 2008.

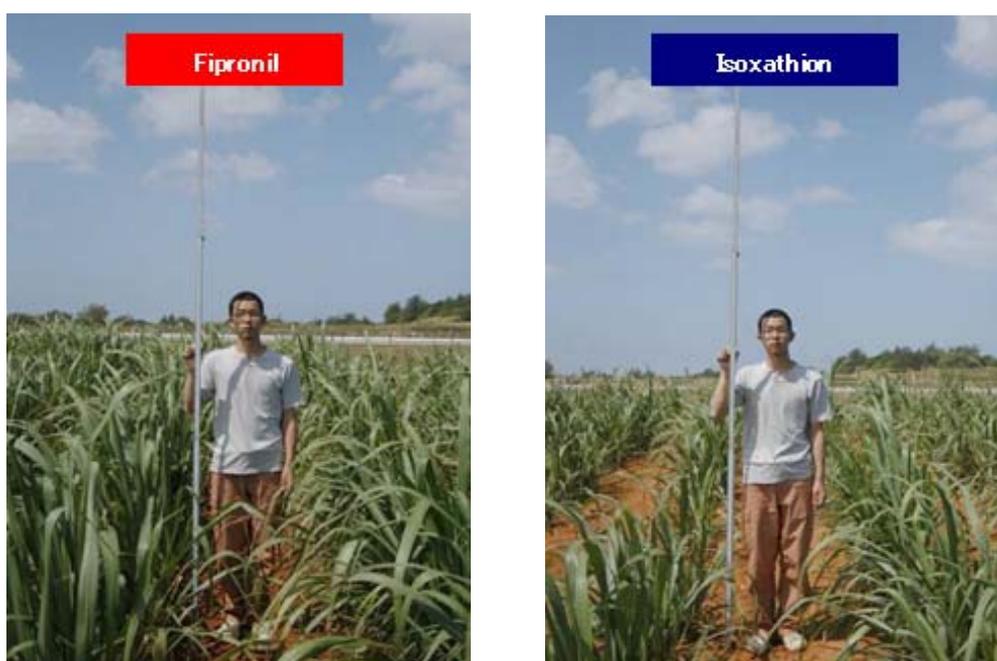


Fig. 5—Field B in March 2008.

Phyostimulatory effect

Figures 6 and 7 show the effects of fipronil on coleoptile (upper) and root (lower) length in sorghum, respectively, and Table 2 shows the results of germination and rooting rates.

From these results, no significant differences were observed between treatment concentrations.

However, there was a tendency for coleoptile length to gradually increase from 0 to 10 ppm, and then to decrease again over 10 ppm. Germination and rooting rates were 85 to 100% regardless of concentration.

These results show that fipronil, at a low concentration of 10 ppm, enhanced sorghum growth.

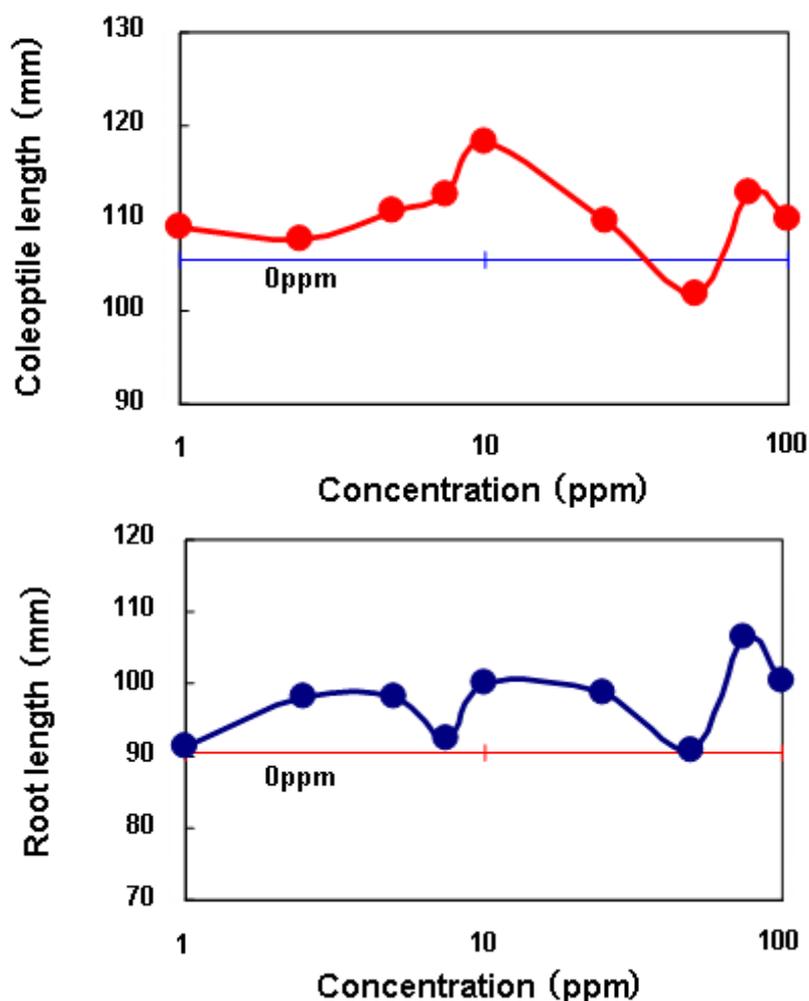


Fig. 6—Effects of fipronil on sorghum coleoptile and root length. The thin lines in the figures show coleoptile (thin red line) and root (thin blue line) length at 0 ppm.

Table 2—Effects of fipronil on the germination and rooting rates of sorghum seeds.

	Concentration (ppmP)								
	0	1	2.5	5	7.5	10	25	75	100
Germination (%)	95	95	95	90	100	95	85	95	95
Rooting (%)	100	95	95	85	100	95	85	95	95

Discussion

The sugarcane wireworm has caused substantial damage for a number of years to sugarcane production in Okinawa. Wireworms damage sugarcane shoots, stems and roots, and shoots are especially damaged as compared with other plant parts. Therefore, non-germination, dead heart, non-ratooning are caused, and sugarcane yield decreases in damaged field (Oshiro *et al.*, 2006). In the 1960s, the incidence of wireworm decreased due to the use of organic chlorine insecticides; however, the subsequent ban of this type of insecticide led to an increase in wireworm numbers after 1971 (Azuma, 1977). Sugarcane production, therefore, decreased as wireworms damage ratoon shoots, and the ratoon crops do not grow as well in damaged fields. At

the same time, labour costs increased, leading to lower profitability. Therefore, a strong demand existed for a new insecticide to prevent wireworm damage in ratoon fields.

In the present study, research was conducted on Minami-daitou Island, one of the areas of large-scale sugarcane production in Okinawa. Sugarcane growth and yield were found to be significantly increased in the fipronil bait-treated areas compared with the control area in each field studied. Early growth (6 months after planting) was increased substantially by the use of fipronil. Nagamine and Kinjo (1979) showed that wireworm damage was confirmed in the early growth stages until three months after planting, especially within the first month (personal communication). In our study, we did not detect any wireworm damage up to 6 months after planting. Future research will focus on sampling for damage earlier in the season.

Although ethylthiodemeton insecticide has been used for a long time to control wireworms in Minami-daitou Island, it was found to be not as effective in controlling wireworms as the fipronil bait. The results suggest that wireworms may have developed resistance to this insecticide on Minami-daitou Island. Future research will also focus on this resistance of wireworm.

From the present study, we speculate that the increase in growth and yield in the fipronil-treated sugarcane was caused by factors other than the reduction in wireworm-related damage. It is already known that 3-cyano-1-phenylpyrazoles, such as fipronil, regulate and promote plant growth. Therefore, we hypothesised that fipronil has a phytostimulatory effect that promoted sugarcane growth, and developed a simple test using sorghum, which has the same type of photosynthetic process as sugarcane. Although there were no significant differences, 10 ppm of fipronil led to a slight increase in the length of the coleoptile of sorghum seedlings. Stevens *et al.* (1999) showed that 2000 mg/L fipronil inhibited the early growth of rice. Though they examined higher fipronil concentrations (250–4000 mg/L), Garcia del Pino and Jove (2005) also showed that the optimum field rate for fipronil was 12–60 ppm, and suggested that the same concentration of fipronil should be used in sugarcane fields. These results show that fipronil at low concentrations of about 10 ppm can increase plant growth, whereas high concentrations of about 2000 ppm can inhibit plant growth. Moreover, Clive (2006) indicated that the application of fipronil as an incorporated soil treatment to cotton, maize, sugar beet or sunflower leads to limited uptake of fipronil into plants. However, it is not known about penetration of fipronil into sugarcane. Further study is required to clarify the uptake of fipronil into sugarcane and the phytostimulatory effect of fipronil on sugarcane.

The present study showed that fipronil bait treatment increases sugarcane growth and yield as compared with conventional insecticides on Minami-daitou Island; therefore, fipronil bait can be regarded as a promising insecticide for use in sugarcane production. However, further research is necessary to better quantify the systemic activity of fipronil in sugarcane as well as its phytostimulatory effect on growth.

Acknowledgments

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EFFETS D'UN APPAT À BASE DE FIPRONIL SUR LE RENDEMENT DE LA CANNE À SUCRE A OKINAWA, JAPON

Par

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**MOTSCLÉS: Appât au Fipronil, Phytostimulateurs,
Sorgho, Rendement De Canne, Taupin.**

Résumé

UN APPAT à base de fipronil (Prince[®] Bait, BASF Agro), nouvellement homologué comme insecticide pour la culture de la canne à sucre en 2006 au Japon, a réduit efficacement les dégâts du taupin, *Melanotus sakishimensis* et *M. okinawawesis* (Coleoptera: Elateridae), tout en augmentant le rendement. En janvier 2008, une croissance élevée de la canne à sucre a été observée dans les régions traitées à l'appât sur l'île Minami-daitou. Afin de confirmer cette forte croissance, nous avons étudié la pousse de la canne et le rendement dans deux champs sur l'île de mars à décembre 2008. De même, comme nous soupçonnions que le fipronil avait des propriétés phytostimulatrices permettant de promouvoir la croissance de la canne à sucre, nous avons effectué un test simple en utilisant le sorgho, une graminée à fort degré de parenté avec la canne à sucre et une plante C₄. La recherche sur l'île Minami-daitou a démontré que la croissance était significativement plus élevée dans les régions traitées à l'appât au fipronil comparée aux régions traitées à l'insecticide éthylthiodémeton (TD[®], Sankyou Agro) ou isoxathion (TD[®] Ace, Sankyou Agro) en formulation granulée. De plus, le rendement en canne dans les régions traitées au fipronil était significativement supérieur comparé aux traitements d'éthylthiodémeton et d'isoxathion. La longueur du coléoptile des graines de sorgho était supérieure avec un traitement de 0 à 10 ppm de fipronil par rapport à des concentrations de plus de 10 ppm où une diminution a été constatée. Il est conclu que l'appât à base de fipronil pourrait accroître le rendement ainsi que la croissance de la canne de manière significative. De plus, il semblerait qu'une faible concentration de fipronil ait un effet phytostimulateur sur les plantules de sorgho.

EFFECTOS DEL CEBO FIPRONIL EN LA PRODUCCIÓN DE CAÑA DE AZÚCAR EN OKINAWA, JAPÓN

Par

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**PALABRAS CLAVE: Cebo Fipronil, Fitoestimuladorio,
Sorgo, Rendimiento Caña de Azúcar, Gusano Alambre.**

Resumen

EL CEBO Fipronil (Prince Bait, BASF Agro), recientemente registrado en 2006 como un insecticida para la caña de azúcar en Japón, se encontró que reduce el daño causado por los gusanos alambres *Melanotus sakishimensis* y *M. okinawawesis* (Coleoptera: Elateridae) y a su vez puede aumentar la producción de la caña de azúcar. En enero de 2008, se observó una alta tasa de crecimiento de las cañas tratadas con cebo Fipronil en la isla de Minami-daitou. La confirmación del aumento en la tasa de crecimiento y su producción, se investigó en dos campos en la isla de Minami-diatou entre marzo y diciembre de 2008. Más aún, debido a que se sospechaba que el Fipronil, tiene un efecto fitoestimulador al promover el crecimiento de la caña de azúcar, se condujo una prueba sencilla usando sorgo, también un cultivo de una especie de gramínea C₄, especie que tiene un alto grado de colinearidad con la caña de azúcar. La investigación se realizó en la isla de Minami-daitou y demostró que el crecimiento de la caña de azúcar en las áreas tratadas con el cebo Fipronil fue significativamente más alto que en las áreas tratadas con el insecticida granulado etiltiodemeton (TD, Sankyou Agro) o el insecticida granulado isoxation (TD Ace, Sankyou Agro). Además, la producción de caña de las áreas tratadas con el cebo Fipronil también fue significativamente superior al de las áreas tratadas con los insecticidas granulados etiltiodemeton e isoxation. Las pruebas con sorgo mostraron que la longitud del coleóptilo de las semillas de sorgo aumentó cuando se trataron con 0 a 10 ppm de Fipronil y luego disminuyeron en las concentraciones superiores a 10 ppm. Se concluye que el tratamiento con el cebo Fipronil en campos de caña de azúcar pueden aumentar significativamente tanto el crecimiento como la producción de caña de azúcar. Más aún, se sugiere que el Fipronil a bajas concentraciones puede tener un efecto fitoestimulador en plántulas de sorgo.