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Sugarcane Ratoon Stunting¹

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Ratoon stunting, also known as ratoon stunting disease (RSD), is considered by many sugarcane pathologists to be the most important disease affecting sugarcane production worldwide, because it can cause 5% to 15% crop yield losses without growers even realizing their fields are infected (Comstock 2002; Davis and Bailey 2000). The disease is caused by a bacterial pathogen and has no easily recognized external symptoms, only stunting of growth that may not always be apparent in the field. Furthermore, even when stunting of growth is noticeable, other factors could be the cause, including poor cultural practices, inadequate moisture, or nutrient deficiencies. During dry weather, the diseased cane will often show signs of drought stress earlier than healthy cane, but with adequate moisture, visual detection of differences may be difficult or impossible.

Partial resistance to RSD occurs in several sugarcane cultivars, but no cultivar is completely immune to infection. In Florida, CP96-1252 and CP00-1101 are resistant; CP78-1628, CP80-1743, CP88-1762, CP89-2143, and CP01-1372 are moderately resistant; however, CL88-4730 and CP84-1198 are moderately susceptible to the disease. A clean seed-cane program is therefore recommended for the cultivars that are moderately resistant or susceptible. Yield losses in a 1999–2002 trial in Florida were estimated at 5%–7% loss in raw sugar (Flynn et al. 2005). Recent experiments have indicated that most cultivars would suffer yield losses if RSD incidence was high.

Symptoms

Although there may be no externally conspicuous symptoms of the disease, internally there is usually an orange-red discoloration of the vascular bundles that contain the water-conducting tissues (xylem) at the basal nodes of the stalk (Figure 1). Similar discoloration is also associated with other sugarcane diseases or insect damage; therefore, it is not a totally reliable indicator of RSD infection. However, in contrast to some other sugarcane diseases, the discoloration of vascular bundles caused by the RSD pathogen does not extend into the internodes. Adjacent nodes in mature stalks will usually show similar discoloration if these stalks are infected by the bacteria.

In some sugarcane cultivars, very young shoots may have a pink discoloration in the immature nodes near the apical meristem. Again, this symptom is not a reliable indicator of the bacterial infection but may serve as an aid in detection of the disease at an early stage.

In diseased fields where stunting is apparent, the shortening of stalks is not usually uniform from stool to stool. Such

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fields may show an irregular or "up and down" growth appearance across the canopy. The effects of the disease are usually more severe in ratoon crops than in plant-cane crops. This is especially true following a drought or other stressful crop conditions, which usually increase losses due to RSD.



Figure 1. Reddish discoloration of vascular bundles at the sugarcane node due to the causal agent of ratoon stunting. Credits: Sushma Sood, USDA

Causal Agent

The organism that causes RSD is a small aerobic bacterium named *Leifsonia xyli* subsp. *xyli*. The genus of this pathogen was previously called *Clavibacter*. Although it can be isolated from diseased cane, pathogen isolation is very difficult since it is very slow-growing (2–3 weeks to obtain bacterial colonies) and must be grown on specialized culture media.

Historically, diagnosis of RSD has been difficult because there are no definitive external symptoms, and internal symptoms do not develop adequately in all varieties. Reliable diagnosis of the disease can be performed using microscopic, serological, and/or molecular techniques. Phase contrast microscopic techniques have proven to be rapid but are not as sensitive as serology and PCR assays for detection of the RSD pathogen. Serological techniques include direct fluorescent-antibody staining, dot blot immunoassay, and the Enzyme-Linked Immunoassay technique was also developed to diagnose RSD and to assess its severity. This technique, whose application results in the identification of infected vascular bundles in the sugarcane stalk, is called the Tissue Blot Immunoassay (TBIA), and it can be used to rapidly test large numbers of sugarcane samples for RSD. TBIA has been used to screen clones for RSD resistance in the USDA Canal Point (CP) sugarcane cultivar development program and is applied to determine disease incidence within seed cane fields. The advantage of TBIA over PCR is that it is less costly and more productive (hundreds of stalks can be assayed quickly).

Spread of the Disease

The RSD pathogen is primarily transmitted through seed cane taken from diseased plants. Because symptoms of the disease are not readily visible, the bacterium may be spread unknowingly from one area to another. Stalks in potential seed fields can be randomly sampled and serologically assayed to determine RSD incidence. The RSD pathogen can be also readily transmitted by knives and mechanical harvesting machines that become contaminated with the bacteria that are contained in the juice from diseased stalks. Transmission by harvesting machinery is very significant.

Cane-chewing animals such as rodents may be capable of transmitting the pathogen when they gnaw on a diseased stalk and then on a healthy one. Not much is known about this means of transmission or its significance. According to reports, the pathogen survives in the soil after harvest to re-infect healthy plants (Bailey and Tough 1992). However, the extent of infection by *Leifsonia xyli* subsp. *xyli* surviving in the soil is not known.

Prevention and Control

The use of healthy, disease-free seed cane is an important control measure (Figure 2). This planting material can be obtained by tissue culture (meristem tip tissue culture), or after hot-water treatment of sugarcane, to eliminate bacteria prior to the establishment of seed cane nurseries. The hot-water treatment consists of an immersion of sugarcane cuttings in running water at ambient temperature for 24-48 hours followed by an immersion in water at 50°C (122°F) for 2-3 hours. This method is most commonly used to control RSD in quarantine facilities or in seed cane nurseries and in numerous sugarcane growing locations in the world. A hot-water treatment program has not been established in Florida or Louisiana so far, because commercial clean-seed nursery stocks derived from tissue culture are available in both locations (Hoy 2017). Serological or molecular assays can be used to monitor the effectiveness of the heat therapy and tissue culture procedures. RSD incidence in 10 Florida Sugar Cane League (FSCL) cultivar increases was checked in 2014 using a tissue blot immunoassay. Only one cultivar at one location was found infected by the RSD pathogen

with a disease incidence of 6.7%. RSD incidence of this cultivar was nil at eight other geographical locations where it was grown. The RSD pathogen was not detected in the nine other cultivars. It was recommended to growers to not use seed cane from the infected field.



Figure 2. Sugarcane plants issued from disease-free seed cane (left) versus sugarcane plants with reduced growth that are issued from seed cane infected by the RSD pathogen (right). Credits: Diego Luzuriaga, Florida Crystals Corporation

Furthermore, since the RSD bacteria are easily transmitted mechanically, sanitation is important in preventing healthy cane from becoming infected. All cane-cutting implements should be protected from contamination from diseased cane or should be disinfected before use on healthy cane. Disinfection can be achieved by heat or chemical treatments. Chemical disinfectants that may be used on cane-cutting knives include Lysol, Dettol, ethanol, Mirrol, and Roccal. At least five minutes of contact with the cutting surface is needed to assure disinfection, but a shorter disinfection time will be partially helpful. Due to the likelihood of reinfection, both heat therapy and/or disease-free plants produced from tissue culture must continually be produced to ensure disease-free seed cane for planting (Hoy 2017; Young 2018).

The use of resistant sugarcane clones has proven to be an efficient method to control RSD. Former commercial cultivar CP 72-2086 has been grown in Florida in the absence of both hot-water treatment and sanitation measures with less than 2% disease incidence in sampled fields. However, CP 72-2086 is an exception, since no other commercial varieties have been found with this level of resistance to RSD. The USDA-ARS Sugarcane Field Station at Canal Point, Florida, screens for RSD resistance; the level of resistance available slows the spread of RSD. However, use of disease-free seed cane is still necessary to completely control the disease.

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