

MATERIAL FACT SHEET

BACILLUS SUBTILIS

MATERIAL NAME: *Bacillus subtilis*

MATERIAL TYPE: Microbial

U.S. EPA TOXICITY CATEGORY: III, "Caution"

USDA-NOP: considered nonsynthetic, allowed. Preventive, cultural, mechanical and physical methods must be first choice for pest control, and conditions for use of a biological material must be documented in the organic system plan (NOP 2000).

MATERIAL DESCRIPTION: *Bacillus subtilis* is a ubiquitous naturally occurring saprophytic bacterium that is commonly recovered from soil, water, air, and decomposing plant material. Under most conditions, however, it is not biologically active and is present in the spore form. Different strains of *B. subtilis* can be used as biological control agents under different situations. There are two general categories of *B. subtilis* strains; those that are applied to the foliage of a plant, and those applied to the soil or transplant mix when seeding.

The *B. subtilis* strain QST713 is a naturally occurring strain that was isolated in 1995 by AgraQuest Inc. from soil in a California peach orchard. This product is applied to foliage (NYDEC 2001). In contrast, *B. subtilis* strain GB03 (Kodiak®) was discovered in Australia in the 1930's and is applied either as a seed treatment or directly to soil. Neither strain is considered a genetically modified organism.

HOW IT WORKS: *B. subtilis* bacteria produce a class of lipopeptide antibiotics including iturins. Iturins help *B. subtilis* bacteria out-compete other microorganisms by either killing them or reducing their growth rate (CPL 2002). Iturins can also have direct fungicidal activity on pathogens.

B. subtilis products are made for many uses. For plant disease control, these include foliar application and products applied to the root zone, compost, or seed. When applied directly to seeds, the bacteria colonize the developing root system, competing with disease organisms that attack root systems (CPL 2002).

According to the manufacturer, *B. subtilis* inhibits plant pathogen spore germination, disrupts germ tube growth, and interferes with the attachment of the pathogen to the plant. It is also reported to induce systemic acquired resistance (SAR) against bacterial pathogens (NY DEC 2001). According to an Agraquest sales representative, in the Serenade® product it is the lipopeptide activity that gives disease control. Any living *B. subtilis* cells in this product have only a minor effect (Cline 2004).

According to the manufacturer, the GB03 strain (Kodiak®) delivers extended protection against disease pathogens through three distinct modes of action:

1. Colonies of *B. subtilis* take up space on the roots, leaving less area or source for occupation by disease pathogens.
2. Kodiak® feeds off plant exudates, which also serve as a food source for disease pathogens. Because it consumes exudates, Kodiak® deprives disease pathogens of a major food source, thereby inhibiting their ability to thrive and reproduce.
3. Kodiak® combats pathogenic fungi through the production of a chemical (an iturin) that inhibits the pathogen's growth (Gustafson 2004). Backman et al. (1997) reported that 60-75% of the seed used for the US cotton crop was treated with Kodiak® for suppression of *Fusarium* and *Rhizoctonia* pathogens.

TYPES OF PESTS IT CONTROLS:

Iturins are reportedly active against the fungus *Sclerotinia fruticola*, which causes rots of harvested stone fruit. *B. subtilis* has also been tested for control of the pathogenic fungus *Verticillium*. *B. subtilis* has been used in conjunction with *Streptomyces gramicifaciens* for control of root rot in cucumber, corky rot of tomato and carnation wilt. According to manufacturers, Norway maple inoculated with *B. subtilis* also shows increased resistance to fungal diseases. It is also claimed to suppress diseases caused by *Fusarium spp.* and *Rhizoctonia spp* (CPL 2002).

FORMULATIONS AND APPLICATION GUIDELINES:

The labels require use of personal protective equipment (long-sleeved shirt and long pants, gloves, shoes plus socks, dust/mist filtering respirator) to mitigate the risk of dermal sensitivity and possible allergic reactions.

AVAILABLE STRAINS:

QST 713 is a foliar application product predominantly used against powdery mildew.

GB03 is a soil application or seed treatment product predominantly used to control root-infecting fungi.

MBI 600 is used for soil application or seed treatment.

B. subtilis var. *amyloliquefaciens* strain FZB 24 is also used for soil application.

OMRI LISTED PRODUCTS:

Bacillus subtilis strain QST 713
 Rhapsody® ASO (Agraquest Inc)
 Serenade® (Agraquest Inc.)
 Serenade® ASO (Agraquest Inc)
 Serenade® Garden Disease Control Ready-To-Use (Agraquest Inc.)
 Serenade® Garden Disease Control Concentrate (Agraquest Inc.)
Bacillus subtilis strain GB03
 Kodiak® Concentrate Biological Fungicide (Gustafson LLC)

NON OMRI-LISTED PRODUCTS:

Bacillus subtilis GB03
 Kodiak Flowable Biological Fungicide (Gustafson)
 Kodiak Hb Biological Fungicide (Gustafson)
 Companion (Growth Products, Ltd.) - liquid for turf, greenhouse, nursery plus other microbes

Bacillus subtilis MBI 600
 Histick N/T (Becker Underwood Inc.) - seed treatment also includes
Bradyrhizobium japonicum
 Subtilex Biological Fungicide (Becker Underwood)
 Subtilex HB (Becker Underwood)
 Pro-Mix With Biofungicide (Premier Horticulture, Inc) - potting mix

Bacillus subtilis var. *amyloliquefaciens* strain FZB 24
 Taegro® (Earth Biosciences Inc.)

REENTRY INTERVAL (REI) AND PRE-HARVEST INTERVAL (PHI): The EPA Workers Protection Standard requires a minimum of 4 hours before reentering a treated field. PHI (days to harvest) is zero.

AVAILABILITY AND SOURCES:

B. subtilis products are available in a number of strains and formulations. The OMRI approved sources are generally available from pesticide dealers.

EFFECT ON THE ENVIRONMENT:

According to information submitted by the manufacturer and supported by reviewers at NY DEC, when used as a seed inoculant both *B. subtilis* and *Bradyrhizobium japonicum* (the naturally occurring nitrogen fixing bacteria used in Histick) are ubiquitous in the environment. The microbes used in this combination product are not genetically modified, and this combination has been used in the past by farmers with no reports of negative impacts on crops. The USEPA required limited toxicological data and established an exemption from tolerance for residues of *Bacillus subtilis* in or on all raw agricultural commodities when applied as a seed treatment for growing agricultural crops (NY DEC 2000).

The US EPA Biopesticides and Pollution Prevention Division documents indicate that *B. subtilis* MBI 600 is non-toxic/pathogenic to birds or insects and

no aquatic exposures are anticipated. No adverse effects to fish or wildlife resources are likely through labeled use of this product and no impact to groundwater is anticipated.

The Department's evaluation of non-target organisms found the Serenade® Biofungicide product to be practically non-toxic to mammals and birds. Label instructions are adequate to protect aquatic organisms and bees. Modeling of maximum use rate applications did not exceed toxicity thresholds for birds, mammals, or aquatic organisms (NY DEC 2000).

EFFECT ON HUMAN HEALTH:

In terms of human health, reviewers found the *B. subtilis* bacteria to be relatively benign. It is not a known human pathogen or disease causing agent. *B. subtilis* produces the enzyme subtilisin, which has been reported to cause dermal allergic or hypersensitivity reactions in individuals repeatedly exposed to this enzyme in industrial settings. The oral, dermal and pulmonary acute toxicity data, as well as eye and skin irritation data on the active ingredient and/or the formulated product, indicate that neither the *B. subtilis* strain QST713 nor the Serenade® Biofungicide product was very toxic, irritating, pathogenic, or infective to laboratory animals by the above noted routes of exposure. The Serenade® Biofungicide product elicited a mild contact hypersensitivity response (tested on guinea pigs) indicating that it is a potential skin sensitizer (NY DEC 2001).

No toxicological effects were reported for *B. subtilis* MBI 600 following oral or dermal inhalation studies and no infectivity or pathogenicity was observed. HiStick® N/T (based on data for the formerly labeled product Epic®) may be somewhat irritating to eyes and skin and may cause skin reactions from direct contact (NY DEC 2000).

EFFICACY

Serenade® –In the many tests done with Serenade®, it gave good results against onion diseases, downey mildew in grapes, and powdery mildew on greenhouse tomatoes. Other trials showed poor to fair efficacy. It should be noted that trials against fireblight in apples showed some efficacy, though low. This disease is notoriously difficult to control, and even low levels are promising.

Serenade is often used and trialed in rotation or combination with other fungicides such as copper products. Results from such trials are not included here, because it is impossible to attribute efficacy data from them directly to Serenade®. However, there is some indication that this use of Serenade® may allow for reduced rates or frequency of the companion fungicides.

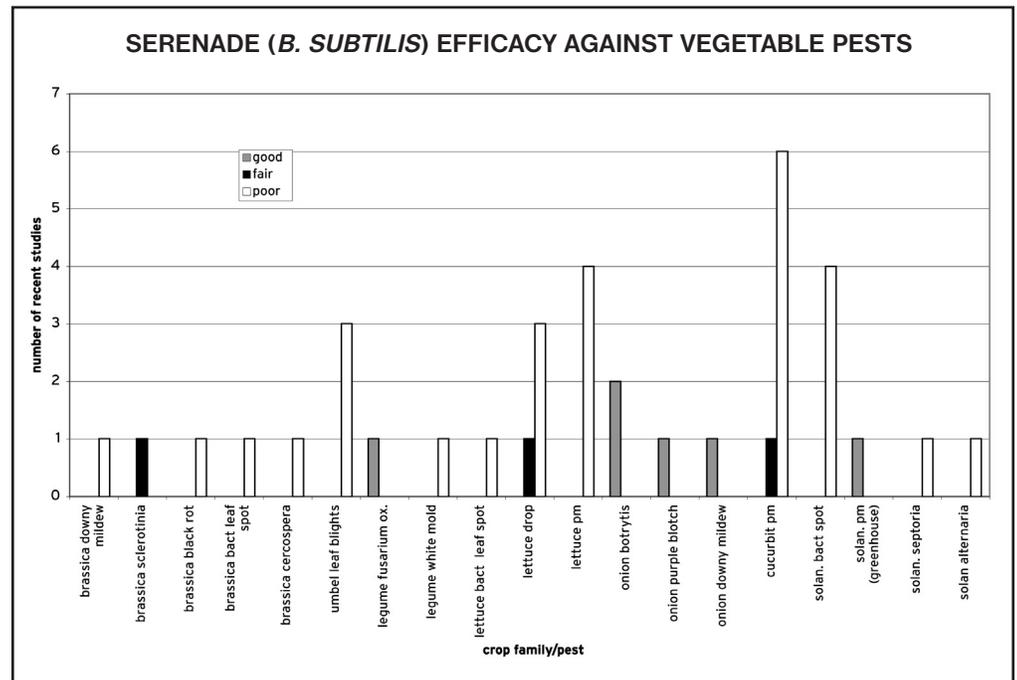
Kodiak®–Kodiak seed treatment needs to be evaluated differently from spray products directed against pathogens. In several of the trials in our database, yield was used as a measurement of the efficacy of the treatment. Kodiak showed significant yield increase of 11 and 15% in two trials, and a non-sig-

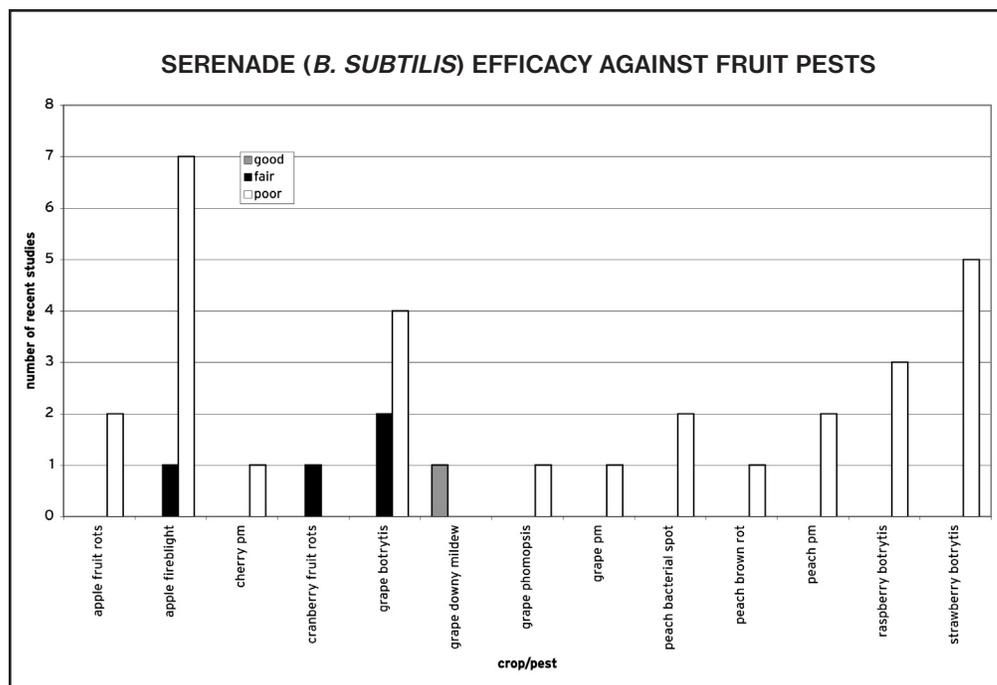
nificant increase of 28% in another. Since the cost of treatment is small, such increases can be very worthwhile to the farmer, even though the numbers may not be impressive.

In four studies, Kodiak® gave little or no visible control of root rot pests, but in one it gave a significant 22% control of Fusarium root rot in beans, and in another its use resulted in an 81% stand increase in chickpeas. A summary of recent university field trials of Serenade® and Kodiak® on fruit and vegetable crops commonly grown in the Northeast was compiled for this fact sheet. These university-based trials typically test products with untreated buffer rows and other conditions that create unusually severe pest pressure.

In the table below, “good control” means statistically significant reductions in pest numbers or damage of 75% or more, compared to an untreated control. “Fair control” includes those with significant reductions of 50-74%, and any non-significant reductions of over 50%. The “poor control” group includes any results with less than 50% reduction.

The level of pest control obtainable is likely to be higher than shown, on completely treated fields in which a good program of cultural controls has also been implemented.





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