BIOLOGY AND ECOLOGY OF *CORYNEBACTERIUM SEPEDONICUM*

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Corynebacterium sepedonicum causes the bacterial ring rot (BRR) disease of potatoes. This disease is feared in geographic areas where it is known to occur because direct and indirect losses attributed to its detection in certified seed lots can be substantial. The primary control procedure for over four decades has been regulatory, *i.e.*, rejection of seed lots from certification upon disease detection (24). This approach to control has been effective in limiting BRR to low frequencies in and among potato operations; but, alone, it has not been sufficient for disease eradication. In fact, one of the most disturbing aspects of BRR is that it may be detected on seed operations with a BRR-free production history and no clear indication of probable inoculum sources. It is the purpose of this contribution to review our current state of knowledge on the biology and ecology of *C. sepedonicum* as a portion of the "North American Task Force for the Eradication of Bacterial Ring Rot".

Description of the Organism

The causal organism, described by Spieckermann and Kotthoff in 1914, is the bacterium *Corynebacterium sepedonicum* (Spieck. & Kotth.) Skapt. & Burkh (25, 28). The species name sepedonicum literally means "leading to decay". It is morphologically and biochemically similar to other gram-positive plant-pathogenic bacteria such as *C. michiganense* and *C. insidiosum*, the causal agents of bacterial canker of tomato and alfalfa bacterial wilt, respectively. These plant pathogenic bacteria all possess a cell wall peptidoglycan based on diaminobutyric acid, group B. Other names proposed recently include (6):

i) *C. michiganense* pv. *sepedonicum*
ii) *Clavibacter sepedonicum*
iii) *Clavibacter michiganense* pv. *sepedonicum*
iv) use of subspecies (subsp.) rather than pathovar (pv) in (i) and (iii).

For the purposes of this treatment, the name *C. sepedonicum* will be used. The bacterium is gram-positive with a size approximately 0.5 x 1.0 μm. Cells are generally slightly club-shaped, appear in L or V formations, and exhibit bending division (4, 30). Under some conditions, coccoid forms have been observed in culture, but reversion to the short rod form has been observed upon subculture. Cultures usually develop mucoid colonies, but non-mucoid mutants can be found in culture and, in one instance, a non-mucoid type has been reported in nature (1).

Unfortunately, *C. sepedonicum* has not been well characterized with regard to biochemical and physiological characteristics. A list of useful characteristics that are fairly consistent is shown in Table 1.

TABLE 1. — *Some biochemical and physiological characteristics of* C. sepedonicum.

Acid production from carbohydrates		Organic acid utilization	
Arabinose	+	Acetate	+
Fructose	+	Citrate	+
Inulin	-	Formate	-
Lactose	-	Fumarate	+
Mannitol	+	Lactate	-
Melibiose	-	Malate	+
Rhamnose	-	Propionate	-
Ribose	-	Succinate	+
Sucrose	+		
Trehalose	-		
Colony pigmentation: - (some yellow) Strict aerobe			
Motility—		Gelatin liquefaction—	
Max. growth temp. 30-32 C		Potato starch hydrolysis +	
NaCl tolerance 3%		Major menaquinone MK-9	
G+C 69.8-74.9			
Peptidoglycan: diaminobutyric acid (DAB), group B (B2)			

Identification and Detection

Although ring rot diagnosis is based primarily on the characteristic symptoms, diagnosis is normally confirmed by a laboratory test. Until recently, the usual confirmatory test was to gram stain smears prepared from stem exudates or tuber vascular tissue (12). Positive bacterial ring rot preparations contain many gram-positive bacteria (0.5 x 1.0 μm). When symptoms are typical and gram stains are prepared and interpreted carefully, these diagnostic procedures appear to work satisfactorily. However, when symptoms are atypical or lacking or are masked by advanced decay from secondary microorganisms as happens frequently, diagnosis is difficult. Since secondary microorganisms can include other gram-positive bacteria (*i.e.*, *Clostridium* spp., *Bacillus* spp., and saprophytic coryneforms), results of the confirmatory gram stain test may be ambiguous (5, 18).

Serological tests (indirect fluorescent antibody stain, latex agglutination, enzyme-linked immunosorbent assay, and Ouchterlony gel diffusion tests have been utilized) have also been used to confirm diagnoses (8, 9, 19, 26, 27). Although serological tests have significantly enhanced the reliability of diagnostic efforts, they are not absolute. Cross-reactions and user error are notable problems (5). Monoclonal antibodies (10) may further enhance test

specificity and, certainly, they provide the possibility that tiered tests (*e.g.*, a primary polyclonal assay followed by selected monoclonal assays) can be devised to handle difficult samples. Methods for sampling and testing seed lots (protocols for composite sampling) need attention as well. In some cases, pathogenicity tests are desirable. Test plants are generally eggplant or tomato because symptoms will develop in 1-3 weeks (15, 16). However, these tests require more time and space, and attention to plant growth condition is important for good symptom development. The bacterium can also be isolated in pure culture when secondary organisms are not present at high populations. Since it grows slowly on agar and is a poor competitor with other bacteria, this approach is usually not used to confirm diagnoses. Unfortunately, a good selective medium is not available for this organism. As with all prior tests described, care in the execution and interpretation of tests is paramount.

Sources of Inoculum

The primary source of the inoculum is considered to be infected tubers, either in storage or in the field (2, 14, 29). Infection occurs through wounds, particularly wounds occurring during handling (harvesting, grading, seed-cutting, and pick-type planters). Infected seed tubers and stored commercial tubers are a source of contamination and spread each season. Volunteers from tubers left in the field can be inoculum sources, especially when recommended crop rotations are not followed. Recent work in North Dakota has demonstrated that sugar beet (*Beta vulgaris* L.) is a symptomless host for *C. sepedonicum* (3). However, the relationship of sugar beet infection to the over-all epidemiology of ring rot is not clear at present. The role of other crop and/or weed species as potential hosts needs to be reassessed as well.

Host Range

Potato has been considered to be the only natural host for *C. sepedonicum* (13). As noted, this perception may change as the role of sugar beet as an inoculum reservoir is elucidated. Although botanical seed can be contaminated with the bacterium, there is no direct evidence that botanical seed has played an epidemiological role. Other susceptible species have played a role primarily as diagnostic indicator hosts (*e.g.*, tomato and eggplant) (15). Susceptible species include: *Althenaea* sp., *Lycopersicon esculentum* (tomato), *L. pimpinellifolium, L. racemgerum, Solanum melongena* (eggplant), *S. antipoviczii, S. balleii, S. cardiophyllum, S. chacoense, S. citrullifolium, S. commersonii, S. corymbosum, S. demissum, S. endlicheri, S. fendleri, S. integrifolium, S. jujuyense, S. mammosum, S. pampasense, S. parodii, S. radicans, S. tequilense, S. thaxcalense, S. vavilovii, S. verrucosum,* and *S. warscewiczii.*

Survival in Nature

Perpetuation by infected potato tubers and, potentially, by infected plants of other species grown in rotation with potato or even found as weeds or volunteers has been discussed. Surfaces in contact with infected plants or plant parts (debris) can be contaminated. Nelson (20, 23) has shown that infectious bacteria can be recovered from dried potato stems from infected plants for 26 months and, in the case of cv. Russet Burbank, this period has been expanded to 63 months (over 5 years!). The bacterium is not a soil inhabitant, however, and there is no evidence for long-term persistence of cells in soil (20).

Long-term survival on clothing, machinery and production and storage surfaces has been demonstrated by Nelson (21). The bacterium can persist for at least 24 months on contaminated surfaces of burlap, kraft paper and polyethylene plastic at 12% relative humidity and 5 C or 20 C. This period is reduced to <14 months if the relative humidity is raised to 94%. Dried slime from symptomatic tubers especially favors long-term maintenance of the bacterium on surfaces. Freezing temperatures, rather than being deleterious to the bacterium, actually appear to enhance persistence.

Methods of Dissemination

The activities of man are pre-eminent in ring rot dissemination. The cultural practices of cutting seed tubers and using pick-type planters have long been condemned for the rapid spread of ring rot within seed lots and the contamination of new seed lots (2, 11, 14, 29). The low incidence and/or elimination of ring rot, especially in Europe, have been held as the example of the effect of these cultural practices. This spread may occur in seed as well as in commercial plantings. Long-distance dissemination is by vehicular transport. Further, redistribution can occur in community seed-cutting and seed-distribution systems. Inadequate sanitation at any point in the production cycle assures continued contamination of future crops.

Some mechanisms of dissemination are not well established. For example, it is known that latent infections do occur in potatoes, but the absolute role in maintenance and spread of the bacterium is not clear (*i.e.*, symptomatic plants or tubers are not present, not just undetected, in a seed lot). Infections resulting from low bacterial numbers have been shown to result in a latency condition with no disease development (22). There is no question that late-season infections and environmental conditions unsuitable for disease development (*e.g.*, early frosts) contribute to maintenance in seed lots from one season to the next season (11). The role of varietal differences in contributing to this phenomenon is not well documented, but tolerant (resistant) cultivars have been suggested as "carriers" (17). Further, the role of insects, birds and animals other than man in the spread of ring rot has not been fully explored. Colorado potato beetles, leafhoppers, the ternate bug

and aphids have all been reported to transmit the bacterium to healthy plants (7).

Conclusion

Recent studies on the ecology and biology of *C. sepedonicum* have altered our perception of BRR. As additional research contributions broaden our knowledge base, this perception will continue to change. Disease eradication is not easy under ideal circumstances and an incomplete description of the organism and the ecological niche it occupies only makes the task more difficult. It is imperative that we continue to explore the ecology and biology of BRR and to share the fruits of those explorations. Realistic cost-benefit assessments for BRR eradication will be no better than the information utilized in making those assessments.

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