

Final Report

SURVIVAL AND RECOVERY OF AMERICAN CHESTNUT TREES HISTORICALLY TREATED WITH HYPOVIRUS

SUBMITTED TO

The American Chestnut Foundation

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This project supported our efforts to initiate and document the recovery of the American chestnuts in the West Salem stand by evaluating the status of trees that were originally treated with hypovirus in the mid-1990s, but were not subsequently treated and do not fall in the current system of permanent plots established in 2001. Specifically, we evaluated: (1) whether the trees were living or dead; (2) the general health of their crowns; (3) the morphology of cankers; and, (4) the level of hypovirus infection among cankers on those trees.

Background and Overview

When chestnut blight was first detected at the West Salem stand in the late 1980s, a clonal population of *C. parasitica* was responsible for all detectable infections. When this study was initiated in 1992, we believed that if we introduced a hypovirus as a biological control agent it would spread rapidly throughout this clonal population. In 1995, we began the introduction of a hypovirus of Italian origin that we believed had good potential to spread from canker-to-canker on the same tree and to other trees within the stand.

By 1997, the detectable spread of the Italian hypovirus was promising enough that, in 1998, we decided to forego additional treatments with the assumption that the released hypoviruses would persist, spread and initiate the process of biological control beyond the area where trees had been treated. However, as we continued to follow hypovirus dissemination, the virulent epidemic continued unabated. In spite of the chestnut blight epidemic, we successfully demonstrated hypovirus dissemination, hypovirus persistence in cankers on trees that received treatment and hypovirus spread to new infections that developed on treated trees. Remarkably, the survival of treated trees in the area of the stand with the longest history of infection has been high even though many trees have been infected for 10-25 years. Some of the treated trees that initially suffered dieback have begun to produce new crowns. This is in contrast to many non-treated trees in the same area that were infected more recently where survival has been poorer.

After the 2000 season, the size of the stand and the increasing number of cankered trees necessitated the installation of permanent plots since it was physically impossible to assess disease among all infected trees. In 2001, twelve permanent plots were established in areas of the stand representing differing incidences of disease [Disease Center (high incidence); Disease Front (moderate incidence); and, Beyond the Disease Front (low incidence)].

When data from 2001-2002 was assessed, it became clear that dissemination of hypoviruses to trees that received no treatment was much lower than expected. Therefore, hypovirus treatment was reinstated in 2003. We have continued to treat trees and assess disease development within these plots. Additional funding provided by a 2008 TACF grant enabled us to focus on non-treated trees immediately outside the twelve permanent plots. We were interested in the fate of trees and infections outside the plots that had not received further treatment. While conducting the 2008 survey and in subsequent field seasons, we observed that many of the originally diseased trees, outside our current system of research plots remained alive and showed significant signs of recovery (Fig 1). Many of these 60 trees had cankers that exhibited morphology typical of hypovirulent infections and some trees were beginning to regenerate new crowns via a system of epicormic branches. Since these trees were not incorporated in the permanent plots, collection of bark samples and data from these trees had not been taken since 2001 when the plot system was installed. Essentially, we had a significant group of recovering trees that were treated over a six-year period 10-20 years ago about which we knew very little.



Figure 1. Two trees in the oldest infected area of the stand that were treated in the mid 1990s that show drastically different responses to treatment. Neither tree had leaves on the main stem in 2004 but the tree on the left is regenerating its crown by a proliferation of new epicormic branches; this "recovery" is becoming more common even in areas that have not received treatment for more than 10 years.

Summary of Findings: Trees that received hypovirus treatment in 1996-1997 but were not incorporated into the plot system in 2001 were located and sampled in June 2009. Approximately 50 living trees that were treated with hypoviruses 12-13 years before were located (based on GPS data). Many had no visible cankers and some had died. Bark samples (12 per canker) were removed from 50 cankers on 29 historically treated trees (if cankers were visible) or from cankers on their non-treated sprouts. Thirty-nine of fifty cankers (78%) yielded hypovirulent isolates and 80-90% of the isolates from some cankers were hypovirulent. Thus, many of these historically treated trees not only have harbored hypovirus for more than a decade, but the hypovirus has proliferated in many other infections.

Two general data tables from the sampled cankers are listed below. Morphology of cankers (based on a subjective 1-4 rating system [1=callus (best) and no stroma; 2=callus and some stroma; 3=callus and moderate stroma; 4=no callus (worse) and heavy stroma] was correlated to the amount of hypovirus detected in a canker. Of the 50 cankers sampled, 74% of the cankers were rated as #1 or #2 (callus with little or no visible stroma) providing evidence that a good level of biological control had been established.

The general tables below show that most cankers fell into category #1 and #2 and the majority of isolates recovered from those cankers were hypovirulent, both measures of successful hypovirus establishment. However, the effectiveness of the biological control at West Salem can only be measured by the long-term survival of many of these trees, a statistic we continue to monitor.

Number of Cankers by Canker Rating			
#1	#2	#3	#4
24	13	8	5

Categories of Isolates Recovered from Cankers			
	Virulent	Euro 7	Non <i>C. parasitica</i>
#1 Rated Cankers	1.5	7.1	3.3
#2 Rated Cankers	0.8	6.3	4.8
#3 Rated Cankers	7.3	2.3	2.5
#4 Rated Cankers	10.2	0	1.8