

## ESTABLISHMENT, PERSISTENCE, AND GROWTH OF ASPEN (*POPULUS TREMULOIDES*) SEEDLINGS IN YELLOWSTONE NATIONAL PARK

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**Abstract.** Quaking aspen (*Populus tremuloides* Michx.) is a long-lived clonal species in which many genetically identical stems (ramets) arise from a common root system. Establishment by seed is extremely rare in the Rocky Mountain region, where most clones that exist today are thought to have established hundreds or thousands of years ago. However, recruitment of new genetic individuals through sexual reproduction has occurred periodically throughout the Holocene, and widespread establishment of seedling aspen occurred in Yellowstone National Park, USA, following the extensive 1988 fires. We combined extensive survey methods with manipulative experiments to investigate the patterns and mechanisms of new aspen genet establishment, growth, and survival during their first decade of development. Excavation and aging of 173 aspen stems in 1996 demonstrated that 65% had established within the first three years after the 1988 fires, and that none predated 1988. Random amplified polymorphic DNA (RAPD) genetic analyses revealed that 92% of the plants were genetically distinct individuals, and 8% were ramets. Annual surveys of 22 permanently marked aspen seedling plots revealed that 24% of 417 seedlings tallied in 1996 had died by 2000. However, mortality varied greatly among the 22 plots, from <10% to >40%, with greater mortality at lower elevations and where soil organic matter was low. To evaluate the mechanisms underlying seedling persistence or mortality, we constructed exclosures in three sites to protect aspen seedlings from ungulate browsing, and applied four experimental treatments from 1996 to 1998: (1) clipping of current year's growth to simulate browsing, (2) removal of potential competitors including lodgepole pine (*Pinus contorta* var. *latifolia*) saplings and herbaceous plants, (3) clipping and competitor removal, and (4) control, i.e., no treatment except protection from browsing. Clipping prevented stem elongation, but removing competitors had no significant effect on height growth of aspen seedlings. Even with protection from browsing, most aspen stems grew slowly (mean increment <25 cm from 1996 to 2000), and many died from causes unrelated to herbivory. Nevertheless, some individuals were >2 m tall in 2003 and appeared vigorous. The aspen cohort that germinated after the 1988 fires appears to be in the earliest stage of a long-term population process, a process that likely will entail a shift from many genetically distinct individuals but few ramets, to relatively few genets having numerous ramets. We predict that many or most of the post-1988 aspen seedlings will die within the next few decades, with little lasting effect on broad-scale vegetation patterns or postfire successional trajectories. However, some new genets appear likely to survive and to establish new aspen clones, with potentially important consequences for demographic and genetic structure of the Yellowstone aspen population.

**Key words:** browsing; *Cervus elaphus*; clonal plants; elk; experimental studies; herbivory; *Populus tremuloides*; quaking aspen; random amplified polymorphic DNA (RAPD) genetic analyses; Yellowstone.

### INTRODUCTION

Quaking aspen (*Populus tremuloides* Michx.) is the most widespread native tree species in North America (Fowells 1965) and is an important component of landscapes of the intermountain west. Aspen is a long-lived

clonal species in which many genetically identical stems (i.e., ramets) arise from a common root system (Day 1944, Barnes 1966). Although aspen can regenerate either by seed or through root sprouting, reestablishment by seed apparently has been extremely rare in the Rocky Mountains during the last century (see review by Kay 1993). Aspen seedlings require a high light environment plus consistent moisture conditions during the first several years, and frequently cannot survive the summers in the intermountain west under

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the current climate (Pearson 1914, Baker 1925, Moss 1938, Barnes 1966, Einspahr and Winton 1976, McDonough 1979, 1985). Most Rocky Mountain aspen clones that exist today are thought to have become established several hundreds or even thousands of years ago, and to have persisted via asexual root sprouting (Baker 1925, Barnes 1966, Mitton and Grant 1996). However, a variety of genetic and other evidence indicates that recruitment of new genets through sexual reproduction has occurred infrequently throughout the Holocene, during brief "windows of opportunity" when seed availability and suitable climatic and substrate conditions combined to provide the very exacting requirements for germination and survival of aspen seedlings (Jelinski and Cheliak 1992, Mitton and Grant 1996).

One such "window of opportunity" appears to have occurred following the extensive 1988 fires in Yellowstone National Park, Wyoming, USA. The fires affected approximately 45% (400 000 ha) of the park (Despain et al. 1989), most of which is covered by lodgepole pine (*Pinus contorta* var. *latifolia* Engelm.) forest. Fires of this scale have occurred in Yellowstone in the past, most recently in the early 1700s (Romme 1982, Romme and Despain 1989). Thousands of aspen seedlings were found in burned forests the year after the 1988 fires (Kay 1993, Romme et al. 1997, Turner et al. 2003a, b). Extensive surveys throughout the park demonstrated that the seedlings had a patchy distribution: local densities ranged from 0 to 46 000 stems/ha, with a median density of 2000 stems/ha on sites where they were present, and seedlings were entirely restricted to burned sites (Romme et al. 1997, Turner et al. 2003a). Before 1988, aspen forests occupied only ~1% of Yellowstone National Park (Despain 1991), and were restricted to lower elevations (<2100 m). However, many of the new seedling populations were found at higher elevations (up to 2600 m), and many were located several kilometers from pre-1988 aspen populations. Genetic analyses using random amplified polymorphic DNA (RAPD) techniques revealed higher genetic diversity among the new seedling populations than among long-established clonal populations in the park (Tuskan et al. 1996). Additional analyses of 23 seedling populations widely distributed throughout the park indicated that most of the genetic variation occurred within rather than between populations, and that geographic differentiation was minimal (Stevens et al. 1999).

If a large number of these seedlings survive to produce new aspen clones, the ecological ramifications of this rare recruitment event may be substantial. Aspen is highly valued as the only major deciduous forest type in Yellowstone Park and throughout much of the Rocky Mountain region (DeByle and Winokur 1985). Aspen forests support high numbers and diversity of breeding birds and other animals, and are visually striking in the fall when the foliage turns yellow in color. However, in Yellowstone and in many other parts of

western North America, aspen stands appear to be deteriorating, i.e., mature stems are gradually dying from disease and other natural causes (Hinds 1985), but are not being replaced by new stems (Schier 1975a). The lack of aspen understory in many stands is a result of chronic heavy browsing by native or domestic ungulates, notably elk (*Cervus elaphus*; e.g., Krebill 1972, Beetle 1979, Olmstead 1979, Kay 1990, Wagner and Kay 1993, Baker et al. 1997, Smith et al. 2000, Hart and Hart 2001). Almost no regeneration of tree-sized aspen has occurred in Yellowstone Park since about 1920, except in a few locations inaccessible to elk (Romme et al. 1995b, Ripple and Larsen 2001, Larsen and Ripple 2003).

However, there is strong and ongoing controversy about the extent and mechanisms of aspen "decline" in Yellowstone and elsewhere in the west (Hessl 2002, Hessl and Graumlich 2002). Aspen forests are thriving in some parts of the Rocky Mountains, despite browsing by native and domestic ungulates (Romme et al. 2001). Some have argued that drought or lack of fire are important reasons for the recent decrease in vigor and abundance of aspen (e.g., Loope and Gruell 1973, Houston 1982, Singer et al. 1998). Extirpation of wolves from most of the western USA in the early 20th century also may have produced changes in elk behavior that led to more intense browsing of aspen (White et al. 1998, Ripple and Larsen 2000, Ripple et al. 2001). A recent assessment of the status of aspen on the winter ungulate range of northern Yellowstone Park, conducted by a committee appointed by the National Research Council (National Research Council 2002), concluded that elk browsing was the primary reason for the lack of aspen regeneration in Yellowstone during the last 80 years, but also emphasized the need for continuing research on interactions among browsing, fire, climatic variability, production of secondary defensive compounds, and effects of wolf predation (now that wolves have been reestablished in Yellowstone Park). It is important to emphasize that all of the research reviewed here has dealt exclusively with mature aspen clones and asexual reproduction. The development of new clones, in new places and having new genetic makeups, would add a novel dimension to the aspen story in Yellowstone, and elsewhere in the west, that has not yet been addressed in any detail.

Investigation of this rare event also can provide more general insights into the influences of large, infrequent disturbances on demographic and genetic processes in long-lived clonal plants. Many clonal herbs, shrubs, and trees have life histories similar to that of aspen, i.e., long periods of almost exclusively asexual reproduction punctuated by rare episodes of sexual reproduction (seedling recruitment; Cook 1983, 1985, Eriksson 1992, 1993, Romme et al. 1995a, Eriksson and Froberg 1996). Although asexual reproduction is an effective mechanism for maintaining populations dur-

ing long periods of relatively stable environmental conditions, at least the occasional occurrence of sexual reproduction and seed dispersal is necessary if populations are to adapt to changing environmental conditions and to occupy new suitable habitat (Eriksson 1992, 1993). However, studying the demography and long-term population dynamics of these kinds of species is challenging because of the rarity and unpredictability of this critical phase of recruitment of new genetic individuals (Eriksson 1992, 1993). Thus, the widespread establishment of aspen seedling following the 1988 Yellowstone fires provides a rare opportunity to investigate the initial patterns and mechanisms of recruitment of new genets into the population of a long-lived clonal plant species, in response to a type of natural disturbance that occurs only infrequently.

In the present study, we combined extensive survey methods with manipulative experiments to investigate the patterns and mechanisms of new genet establishment, growth, and survival during the first decade of development of this new cohort of aspen individuals in the burned forests of Yellowstone National Park. We addressed four questions:

- 1) Did seedling establishment occur only during the first two years after the 1988 Yellowstone fires, as suggested by our earlier surveys (Romme et al. 1997), or did additional establishment occur in subsequent years?
- 2) Did the new genets begin to establish clonal structures within the first decade, as suggested by our earlier limited observations (Romme et al. 1997), or did new populations continue to consist predominantly of unique genetic individuals?
- 3) Where in the landscape did seedlings persist and grow throughout the first decade after the 1988 fires, and where did seedlings die or fail to increase in height or diameter? We hypothesized that aspen seedlings would persist primarily at lower elevations, close to the pre-1988 distribution of aspen, where browsing was light, and where competition from lodgepole pine saplings and herbs was not intense.
- 4) What were the proximate mechanisms of aspen seedling persistence and growth, i.e., how did local environmental conditions, browsing, and interspecific plant competition interact to determine aspen seedling growth and survival? We hypothesized that chronic heavy browsing would prevent height growth and could even kill seedlings. We also hypothesized that seedlings would grow faster in the absence of competing lodgepole pine saplings and dense herbaceous vegetation. Finally, given protection from browsing, we expected populations located near the pre-1988 range of aspen to grow faster than populations remote from the pre-1988 range because of more favorable microclimatic conditions in the former areas.

#### STUDY AREA AND METHODS

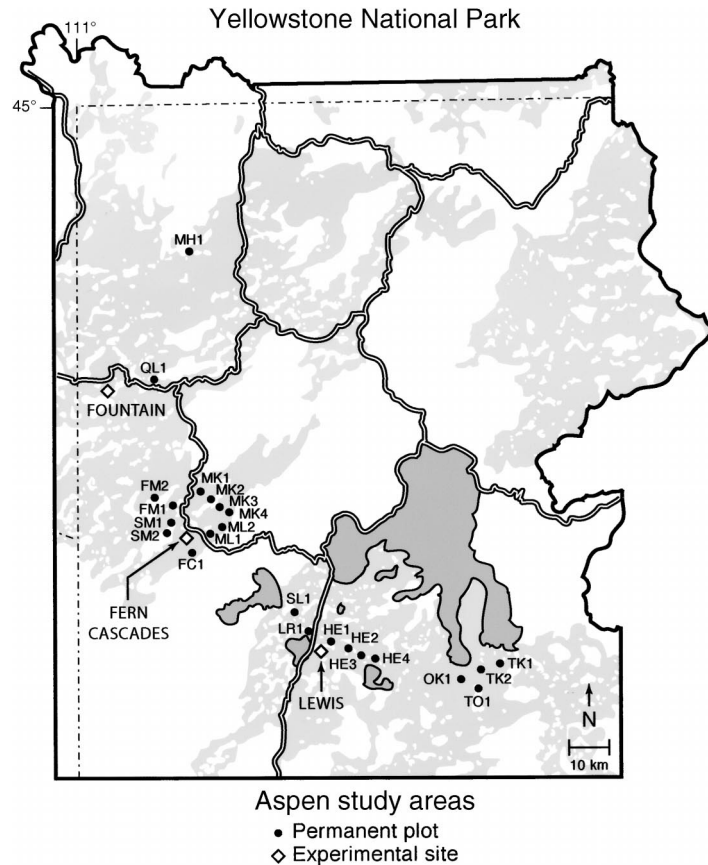
Yellowstone National Park (YNP) encompasses 9000 km<sup>2</sup> in the northwest corner of Wyoming, USA (Fig. 1). Our study focused on the forested subalpine plateaus that cover most of YNP and support similar vegetation throughout. The climate is generally cool, with moist winters and springs and dry summers and autumns (Martner 1986). The two most important environmental gradients controlling vegetation on the plateau relate to elevation and geological substrate (Despain 1991). Soil fertility and moisture-holding capacity are generally lower on rhyolite and tuff than on andesite and lake bottom substrates, although all four substrates are relatively infertile. Precipitation generally is greatest at higher elevations, and on the westerly and southerly portions of the subalpine plateau that receive orographic precipitation from winter storm systems arriving predominantly from the west. Approximately 80% of the park is covered with coniferous forests dominated by lodgepole pine (Despain 1991).

#### *Sampling methods (questions 1–3)*

*Timing of seedling establishment and development of clonal structure.*—To determine the timing of seedling establishment and the extent to which clonal structure has developed, we made a detailed analysis in 1996 (eight years after the fires) of three seedling populations located in three areas that were subjectively selected to represent contrasting environmental conditions (Table 1, Fig. 1). The Fountain site was at the lowest elevation (2060 m), in a lodgepole pine forest near the lower forest—steppe ecotone in western YNP, and near the upper elevational limit of mature aspen in 1988 (though no pre-1988 aspen were present within 0.5 km of the site). The Fern Cascades site was at an intermediate elevation (2270 m), within an extensive tract of lodgepole pine forest, nearly 7 km from the pre-1988 distribution of mature aspen. The Lewis site was at the highest elevation (2400 m), and also was located in extensive lodgepole pine forest nearly 11 km from pre-1988 aspen. All three sites were burned in 1988 at variable severity, ranging from stand-replacing crown fire to low-severity surface fire that killed only a few of the canopy trees (Turner et al. 1994).

One sampling plot was established at the Fountain and Lewis sites, and three plots were established at the Fern Cascades site. Within these five 5 × 10 m to 10 × 30 m rectangular plots (size varied to enclose 35–48 aspen stems), the position of each individual stem was recorded to the nearest decimeter on an *x*–*y* grid. Every aspen stem within the plot was partially excavated by hand trowel, and its age was determined from apical bud scale scars, which were very conspicuous on these plants, even in browsed individuals. Presence/absence of past browsing was determined from the occurrence of dominant lateral stems originating just below an obviously clipped and formerly dominant stem.

FIG. 1. Geographic locations of the 22 post-fire aspen seedling plots sampled within Yellowstone National Park, Wyoming, USA, and the three locations where experimental studies were conducted. Park roads are shown in double lines, state boundaries in dashed lines, and major lakes are shaded dark gray. The lightly shaded area depicts the extent of the 1988 fires. See Tables 1 and 2 for plot abbreviations and descriptions.



The major roots of each plant were then traced by means of further excavation, and the number of connections with other stems (potential ramets), maximum root length and depth, and presence/absence of lateral roots and descending roots (sinking roots; Pregitzer and Friend 1996) were recorded. A random sample of 20 stems was completely excavated and returned to the laboratory, where root and shoot sections were dried separately at 70°C for 24 h. Dry mass of roots and shoots was determined, and ratio of root mass:whole-plant mass was calculated for each plant.

The four youngest, fully expanded leaves (leaf plastochron index 8–12) also were collected from each stem within the mapped plots for genetic analyses to verify that aspen stems not physically connected to other stems were actually distinct genets. Samples were placed in a 1.5 mL microcentrifuge tube with 1 mL TE buffer, frozen under dry ice, and shipped to Oak Ridge National Laboratory for further sample preparation and analysis. Sample preparation and DNA extraction were identical to those described by Tuskan et al. (1996). Based on this previous study, four decamer primers (AC07, AD18, AN10, and Y18; Operon Technologies, Alameda, California, USA) were used to characterize the genetic makeup of each stem. A total of 32 polymorphic RAPD bands were used in the genetic analysis of band sharing among individuals.

*Landscape patterns of seedling growth and persistence.*—To determine where in the landscape the aspen seedlings were persisting and growing and where they were dying or failing to grow, we established 23 permanent sampling plots representing the range of elevation, substrate, browsing intensity, and potential plant competition that existed across the Yellowstone plateau in 1996. Plots were located along 13 foot trails (Table 2, Fig. 1) in areas where aspen seedlings were relatively abundant (Turner et al. 2003a), at a distance of 2.5–100 m (mean of 33 m) from the trail, and permanently marked with rock cairns. There was no evidence that aspen had been present in the vicinity of the plots prior to 1988, and genetic analyses conducted by Stevens et al. (1999) confirmed that the aspen seedlings were genetically distinct individuals rather than root sprouts from preexisting clones. Plot size was varied to encompass at least 10 aspen seedlings within an area of relatively homogeneous environmental conditions. The average plot size was 30 m<sup>2</sup>, but ranged from 4 m<sup>2</sup> to 180 m<sup>2</sup> depending on local density of aspen seedlings. Plot elevations ranged from 2145 m to 2505 m (Table 2). To provide broad spatial coverage, plots on the same trail were positioned at least 1.4 km apart. The largest geographic distance between plots was 74.3 km, but two plots, each at the head of separate trails, were only 0.2 km apart. The 23 aspen study plots clus-

TABLE 1. Environmental characteristics and aspen seedling characteristics in 1996 at three sites where seedlings were excavated to determine establishment time and clonal development (questions 1 and 2), and where experiments were conducted to determine mechanisms of aspen seedling growth and persistence.

Characteristics	Fountain site	Fern Cascades site	Lewis site
Elevation (m)	2060	2270	2400
Substrate	rhyolite (relatively infertile)	rhyolite (relatively infertile)	rhyolite + lake bottom sediments (relatively fertile)
Relative length of growing season	longest (lowest elevation)	intermediate	shortest (deep, late-melting snowpack)
Vegetation before the 1988 fire	130-yr-old lodgepole pine forest	280-yr-old forest of lodgepole pine, subalpine fir, and Engelmann spruce	220-yr-old forest of lodgepole pine, subalpine fir, and Engelmann spruce
Vegetation after the 1988 fire	lodgepole pine saplings, plus herbs <i>Carex geyeri</i> and <i>Epilobium angustifolium</i>	lodgepole pine saplings, plus low shrub <i>Vaccinium scoparium</i> and herbs <i>Carex geyeri</i> and <i>Epilobium angustifolium</i>	lodgepole pine saplings, plus low shrub <i>Vaccinium scoparium</i> and herbs <i>Carex geyeri</i> and <i>Epilobium angustifolium</i>
Distance to pre-1988 aspen clones (km)	0.5	6.9	10.7
UTM northing, UTM easting	4943050, 499500	4921800, 513800	4907000, 532750
Mean maximum root length (cm)	38.8	89.2	30.6
Mean maximum root depth (cm)	13.1	27.7	17.7
Plants having descending roots (%)	96	76	92
Plants having lateral roots (%)	65	40	43
Mean whole-plant biomass (g)	6.2	15.8	1.8
Mean ratio of root : whole-plant biomass	0.33	0.42	0.40
Browsed plants (%)	80	90	100

Note: See Fig. 1 for locations.

tered into four geographic regions: Northern, Heart Lake, Old Faithful, and Yellowstone Lake areas. The Northern region is only comprised of two plots because aspen seedlings were sparse in that area (Turner et al. 2003a).

Ten to 20 aspen seedlings occurring within each plot were measured in 1996, then again in 1997, 1998, and 2000. Measurements included height (length of the main stem), basal diameter, and whether the plant had been browsed by ungulates. Genetic samples were obtained in 1997 from each individual within the plots (Stevens et al. 1999). In 1998, a composite sample of mineral soil (0–10 cm depth) was obtained from each permanent plot to obtain general soil characteristics (percent total nitrogen, percent total carbon, C:N ratio, pH, percent organic matter, available phosphorus, and available potassium). Soils were air dried at the field station, and all analyses were conducted at the Wisconsin Soil and Plant Analysis Laboratory, Madison, Wisconsin, USA. We also estimated percent vegetative cover (all vascular plant species combined) in each plot in 1996, 1997, and 1998 using a line-intercept method along a representative 10-m transect along the central axis of the plot. (In plots that were less than 10 m in length, vegetative cover was determined along a 10-m transect centered on the plot.) The density of lodgepole pine seedlings was obtained by counting the number of such seedlings in two 1-m belts along the 10-m

transect. Elevation and Universal Transverse Mercator (UTM) coordinates of each plot were obtained from a global positioning system (GPS), and the geologic substrate was determined from detailed geologic maps of the Yellowstone area. The distance to the prefire distribution of aspen clones of trails along which permanent plots were located was computed using a geographic information system (GIS) (Turner et al. 2003a).

At the plot level, percent mortality of aspen seedlings at each plot was determined over a 2-yr interval (1996–1998) and a 4-yr interval (1996–2000). Variation in percent mortality among plots was evaluated using stepwise multiple regression; variables tested for inclusion in the model were lodgepole pine density, initial aspen seedling density, percent herbaceous cover, percent soil organic matter, soil C:N ratio, soil available potassium, soil available phosphorus, soil percent nitrogen, soil percent carbon, pH, elevation, and distance to prefire aspen clones. For analyses of aspen seedling height and diameter, all data were transformed prior to analysis (using square root and  $\log[\text{diameter} + 1]$ , respectively) to achieve normality. Repeated-measures ANOVA was conducted on height and diameter for individual aspen seedlings that survived the intervals 1996–1998 ( $n = 343$  seedlings) and 1996–2000 ( $n = 317$ ) to determine whether there were significant differences between years or among plots. Because there were no significant differences in height or diameter

TABLE 2. Site characteristics of 23 variable-sized plots in which postfire aspen seedlings were sampled in Yellowstone National Park in 1996, 1997, and 2000.

Plot name	Abbrev. on Fig. 1	Elevation (m)	Distance to prefire aspen clones (km)	Initial no. aspen sampled	Aspen seedling height (cm) in 2000		Mortality of marked aspen 1996–1998 (%)	Mortality 1996–2000 (%)
					Mean (SD)	Maximum		
Northern region								
Mount Holmes	MH1	2330	16.6	17	23.2 (11.0)	41.0	18	18
Harlequin Lake	QL1	2145	0.9	11	33.3 (15.3)	53.5	54	54
Old Faithful region								
Fern Cascades	FC1	2300	6.9	18	23.5 (10.1)	42.0	20	44
Firehole Meadow #1	FM1	2375	1.8	16	38.8 (10.1)	54.0	24	44
Firehole Meadow #2	FM2	2505	1.8	13	24.9 (7.7)	35.5	8	15
Mallard Creek #1	MK1	2160	7.5	21	42.9 (16.7)	81.0	18	24
Mallard Creek #2	MK2	2265	7.5	17	43.2 (11.1)	56.5	18	18
Mallard Creek #3	MK3	2370	7.5	15	16.4 (5.7)	29.0	20	20
Mallard Creek #4	MK4	2390	7.5	16	18.8 (14.0)	43.8	32	32
Mallard Lake #1	ML1	2290	8.8	18	25.3 (9.6)	39.7	32	50
Mallard Lake #2	ML2	2370	8.8	12	17.0 (6.1)	31.5	8	17
Summit Lake #1	SM1	2220	3.1	22	24.8 (12.2)	45.7	12	23
Summit Lake #2	SM2	2420	3.1	22	21.3 (13.1)	54.1	14	14
Heart Lake region								
Heart Lake #1	HE1	2390	10.7	17	22.47 (9.5)	34.8	25	29
Heart Lake #2	HE2	2455	10.7	14	24.2 (14.3)	50.5	†	7
Heart Lake #3	HE3	2390	10.7	39	32.1 (15.1)	62.0	13	20
Heart Lake #4	HE4	2335	10.7	30	35.9 (16.5)	67.7	15	43
Lewis River Channel	LR1	2374	10.7	17	22.5 (11.1)	43.9	9	12
Shoshone Lake	SL1	2417	6.7	17	16.8 (6.2)	24.5	41	41
Yellowstone Lake region								
Outlet Creek	OK1	2380	10.5	20	35.5 (10.8)	58.0	10	10
Trail Creek #1	TK1	2385	15.1	18	67.7 (14.3)	92.5	6	11
Trail Creek #2	TK2	2415	15.1	11	56.8 (21.1)	81.9	14	18
Two Ocean Plateau	TO1	2385	13.7	16	35.2 (12.4)	59.2	6	6

Note: Plots in relatively close proximity (Fig. 1) are grouped by geographic region.

† HE2 was not located in 1998 and thus was not sampled.

between years (see *Results: Question 3: landscape patterns of seedling growth and persistence*), subsequent analyses of aspen seedling height and diameter among plots used the overall plot means averaged across years. Stepwise multiple regression was used to identify factors that influenced variation among plots. Independent variables entered into the model at the plot level were the same variables as in the mortality regressions.

#### *Experimental methods: mechanisms of seedling persistence and growth (question 4)*

A factorial field experiment was established in 1996 to quantify the relative importance of and interactions among local site conditions, ungulate browsing, and interspecific competition. Experiments were conducted within an area of 2–4 ha at each of the three sites where aspen stems were excavated and analyzed for questions 1 and 2 (Table 1, Fig. 1). The design was a  $2 \times 2$  factorial with the following levels: (1) ungulate browsing (excluded or simulated) and (2) competitors (eliminated or undisturbed). Three replicates were established of each of the four treatments at each of the three geographic locations ( $n = 12$  plots per geographic location, 36 plots total).

Each plot was 100–500 m<sup>2</sup> in size and contained at least 20 aspen seedlings separated from other groups of seedlings and growing in habitats representative of the general study area. Elk-proof fencing was constructed around each plot using three or more rows of barbless wire attached to posts or standing dead trees; treatments were assigned randomly among the 12 plots. In every plot, the location of 20–25 aspen seedlings was recorded by its distance along and to the side of a meter tape stretched through the center of the plot. Each individual was numbered and tagged in 1996 to facilitate relocation in subsequent years.

For treatments in which interspecific competition was reduced, we eliminated aboveground vegetation within a 0.5-m radius of each aspen seedling. Lodgepole pine seedlings were eliminated by snipping them off at ground level using pruning clippers. Herbaceous vegetation was eliminated by cutting the plants to within 1–2 cm of ground level, then carefully applying a systemic herbicide (Roundup; Monsanto, St. Louis, Missouri, USA) with a small paint brush in wind-free conditions. Because the herbaceous vegetation continued to resprout, clipping and herbicide application were repeated during late summer and during subsequent

TABLE 3. Age structure of aspen stems in 1996 in three areas that burned in 1988 in Yellowstone National Park (Fountain, Fern, and Lewis sites; see Fig. 1, Table 1).

Measure	Year of origin (via germination or ramet formation)							
	1989	1990	1991	1992	1993	1994	1995	1996
Number (%) of all excavated stems; $n = 173$	63 (36)	36 (21)	32 (18)	14 (8)	11 (6)	7 (4)	7 (4)	3 (2)
Number (%) of excavated ramets only; $n = 24$	0	0	0	1 (4)	6 (25)	7 (29)	7 (29)	3 (12)

Notes: Aspen was not present in these areas prior to 1988. Data are aggregated over all three sites and are presented for all stems (ortets and ramets combined) and for ramets only (as determined from morphological attachment or genetic similarity).

field seasons. Despite the careful application, a few aspen seedlings showed indications of herbicide effects, particularly in places where they were growing in very close proximity to the treated herbs, and those individuals were excluded from all analyses. For treatments simulating ungulate browsing, all of the current year's growth was clipped during late summer in each year. Ungulates were excluded from all experimental plots, so browsing was either absent or simulated. We chose to simulate browsing because the occurrence and level of browsing within the study plots could be controlled. Reliance on natural browsing could lead to extremely high or no browsing in individual plots each year, and this would confound our ability to interpret potential effects of browsing.

Within each of the treatment plots, the following data were recorded near the end of each growing season (mid to late August) in each year of the study: location of all tagged aspen stems (to detect mortality or recruitment of new stems as well as to follow individual stems over the course of three years), height and diameter of the tallest leader, and number of main stems in groups that all appear to arise from the same point in the ground.

Experimental treatments (competitor removal and simulated browsing) were discontinued in 1998, but the exclosures surrounding the study plots are still being maintained. We remeasured aspen seedling height and diameter during the summer of 2000 in the control plots and the competitor removal plots to quantify recent aspen seedling growth in the absence of ungulate browsing and to continue our estimates of aspen seedling survival.

All data on aspen height and basal diameter were transformed prior to analysis (using square root and  $\log[\text{diameter} + 1]$ , respectively) to achieve normality. Repeated-measures ANOVA was used to test for effects of treatment and geographic location on aspen seedling height and diameter within the 36 experimental plots.

## RESULTS

### Questions 1 and 2: timing of seedling establishment and development of clonal structure

We determined the age of 173 aspen stems among all three sites, of which 36% had established in 1989

and comprised the largest yearly age class (Table 3). Stems also established in 1990–1996, but in progressively smaller numbers each year, with only 2% of the stems dating from 1996. Proportions by year were very similar in all three study sites. However, morphological characteristics of the stems varied substantially among sites (Table 1).

Clonal development in 1996 (eight years after the 1988 fires) was very limited. Of the 173 stems sampled over all sites, only 12 pairs appeared to be connected by a lateral root. The genetic analyses generally confirmed that the physically unconnected plants were all genetically distinct ortets, i.e., they each had a unique DNA makeup as determined by RAPD analysis, and that most of the physically connected plants were ramets, since they shared all scored bands (the probability of this occurring by chance was  $9.34 \times 10^{-21}$ ). Thus, our interpretations based on morphology were verified by genetic analysis of the 173 sampled stems in all but five cases: three pairs of physically unconnected plants were determined genetically to be ramets, and two pairs of plants that appeared to be physically connected were determined genetically to be separate genets. Thus, of the 173 plants sampled, 160 (92%) were genetically distinct ortets, and only 13 (8%) were ramets.

### Question 3: landscape patterns of seedling growth and persistence

Survival, height, and diameter were tracked from 1996 to 2000 on an initial cohort of 417 individual aspen seedlings distributed among the 23 permanent plots. (We refer to these plants hereafter as "seedlings," although our results above indicate that a very small proportion of them (<10%) actually may have been ramets.) Genetic analyses of 410 plants within these 23 plots (Stevens et al. 1999) revealed that all tested individuals were distinct genotypes. Initial (1996) density of aspen in the 23 plots averaged 1.4 stems/m<sup>2</sup> and ranged from 0.1 to 4.5 stems/m<sup>2</sup> (Table 4). The percent cover of herbaceous vegetation was generally high (averaging ~60%), and lodgepole pine densities averaged 2 stems/m<sup>2</sup> but ranged from 0.1 to 15 stems/m<sup>2</sup> (Table 4).

Between 1996 and 2000, living aspen seedlings declined from 417 in 1996, to 397 in 1997, to 343 in

TABLE 4. Mean characteristics (and range) among the permanent plots ( $n = 22$ ) in which aspen height and survival were monitored on an initial cohort of 417 seedlings.

Variable	1996	1997	1998	2000
Herbaceous cover (%)	58.7 (9.1–100)	61.8 (16.5–100)	59.8 (15.4–93.7)	... †
Lodgepole pine density (stems/m <sup>2</sup> )	2.1 (0.1–12.5)	2.3 (0.1–15.3)	2.1 (0.1–10.2)	... †
Initial aspen density in plot (stems/m <sup>2</sup> )	1.4 (0.1–4.5)			
Aspen seedling height (cm)	29.4 (15.1–51.5)	26.9 (10.4–49.7)	31.5 (16.6–68.2)	30.5 (16.4–68.8)
Aspen seedling diameter (mm)	7.0 (3.4–11.4)	10.4 (4.9–16.2)	10.4 (5.0–15.3)	8.8 (4.9–15.3)
No. leaders	2.2 (1.1–4.9)	1.9 (1.0–3.3)	1.9 (1.0–3.9)	1.7 (1.0–3.3)

† Not measured in 2000 due to time constraints.

1998, and to 317 in 2000. Thus, a total 24% of the original cohort of seedlings died during the four years. Among plots, aspen seedling mortality averaged 19% per plot but ranged from 6% to 56% between 1996 and 2000 (Table 2). Stepwise multiple regression revealed that mortality between 1996 and 1998 was greater in plots having lower soil pH, located at lower elevations, with lower initial aspen density, and with higher soil available potassium (Table 5). These variables together explained 75% of the variation in aspen seedling mortality. (Soil-available phosphorus and distance to pre-fire aspen also were significant in the model but explained only an additional 1% of the variation.) Multiple regression analysis of 1996–2000 mortality explained less of the variability in mortality ( $r^2 = 0.45$ ). As in 1996–1998, greater mortality occurred in plots having lower pH, lower elevation, and higher soil available K; however, initial aspen density was no longer significant, and greater mortality was associated with lower initial pine density (Table 5). Initial herbaceous cover and soil C:N ratio also were significant in the 1996–2000 model, but each explained only 1% of the variation. Additional variables—percent soil organic matter, percent N, and percent C—were tested but were

not significant in either the 1996–1998 or 1996–2000 models (Table 5).

Repeated-measures ANOVA on individual seedlings that survived from 1996 to 2000 revealed that individual aspen seedlings did not change significantly in height or diameter through time, but that height and diameter did vary among plots. Repeated measures ANOVA on the mean aspen seedling height and diameter per plot also indicated no significant change in mean height or diameter through time. Because differences among years were not significant, subsequent analyses of seedling height and diameter used plot means computed over all years.

Mean aspen seedling height among all plots averaged 30 cm and ranged from 16 to 68 cm in 2000 (Tables 2 and 4). As of 2000, even the tallest aspen in 22 permanent plots (one of the original 23 could not be relocated) was <100 cm in height, and in half of the plots, the tallest individual was <50 cm in height (Table 2). Annually, 85–88% of individual aspen stems showed evidence of past browsing. Multiple regression analysis revealed that mean aspen seedling height among plots was positively related to soil pH and available soil potassium (Table 6). These two variables to-

TABLE 5. Results of multiple regression analysis on the percentage mortality of aspen seedlings from 1996 to 1998 and from 1996 to 2000 in 22 permanent plots located in areas that burned in 1988 in Yellowstone National Park.

Variable	Coefficient	Partial $r^2$	Model $r^2$	$P$
Mortality from 1996 to 1998				
pH	-0.33	0.30	0.30	<0.0001
Elevation	-0.0011	0.23	0.53	<0.0001
1996 aspen seedling density	-0.036	0.14	0.67	<0.0001
Soil available K	0.0007	0.08	0.75	<0.0001
Soil available P	-0.0002	0.01	0.76	0.0002
Distance to prefire aspen	-0.0062	<0.01	0.76	0.0193
Mortality from 1996 to 2000				
pH	-0.30	0.13	0.12	<0.0001
Elevation	-0.0015	0.12	0.25	<0.0001
Soil available K	0.0012	0.11	0.36	<0.0001
1996 pine density	-0.018	0.09	0.45	<0.0001
1996 percent herbaceous cover	-0.0007	0.01	0.46	0.0016
Soil C:N ratio	-0.2122	0.01	0.47	0.0089

Notes: Herbaceous cover, 1996 pine density, and C:N ratio were not significant in the 1996–1998 model. Soil available P, 1996 aspen seedling density, and distance to prefire aspen were not significant in the 1996–2000 model. Percentages of soil organic matter, N, and C were also tested but were not significant in either model.



TABLE 6. Results of multiple regression analysis on aspen seedling height and diameter in 22 permanent plots located in areas that burned in 1988 in Yellowstone National Park.

Variable	Coefficient	Partial $r^2$	Model $r^2$	$P$
Aspen seedling height				
pH	1.458	0.44	0.44	0.0008
Soil available K	0.006	0.12	0.56	0.0333
Aspen seedling diameter				
pH	0.198	0.25	0.25	0.0183
Lodgepole pine sapling density	-0.020	0.19	0.44	0.0182
Distance to prefire aspen	-0.013	0.18	0.62	0.0094

Notes: Repeated-measures ANOVA found no significant change from 1996 to 2000 in either height or diameter; therefore, mean height and diameter were computed over all years in each plot;  $n = 22$  plots in this analysis. The other variables listed in Table 5 were also tested, but none was significant.

gether explained 56% of the variation in mean aspen seedling height among plots. Aspen seedling diameter averaged 7–10 mm (Table 4). Mean seedling diameter was greater in plots having higher soil pH, lower pine density, and in locations closer to prefire aspen clones (Table 6). These three variables explained 62% of the variation in mean aspen seedling diameter among the 22 plots.

*Question 4: mechanisms of seedling persistence and growth*

Repeated measures ANOVA (1996 to 1998) on the aspen in the experimental plots found significant main effects on aspen seedling height due to year, geographic location, and the browse treatment (Table 7). Interactions among main effects were occasionally significant, but they contributed little explanatory power and are not discussed further here. Among treatments, the simulated browsing treatment resulted in aspen that did not increase in height through time (Fig. 2a; note over-

lapping confidence intervals in 1996, 1997, and 1998), although stem diameters did increase (Fig. 2b). Seedling height was lowest (~26 cm) in plots that were subjected to both browsing and competitor removal (Fig. 2a). Removing competitors did not significantly increase aspen height or diameter relative to control plots (Fig. 2). Aspen seedlings in the control plots and the competitor-removal plots increased in height compared to other treatments, an average of 10 cm between 1996 and 1998, but did not differ from one another (Fig. 2a). By 1998, aspen in these two treatments averaged >40 cm in height, with the tallest individual reaching 153 cm. Increase in stem diameter was greater in control plots and competitor-removal plots and lower in plots with simulated browsing (Fig. 2b).

Geographic location had the strongest influence on both seedling height and diameter (Table 7, Fig. 3). Mean seedling height was lower in all years of measurement at the high-elevation Lewis site compared to the Fountain and Fern sites, and it did not increase

TABLE 7. Repeated-measures ANOVA results for aspen seedling height and diameter in the experimental plots.

Variable	df	MSE	$F$	$P$
a) Aspen height ( $r^2 = 0.30$ )				
Site	2	878.00	168.21	0.0001
Browse	1	84.2	16.13	0.0001
Competitor removal	1	7.51	1.44	0.2309
Replicate	2	1.66	0.32	0.7271
Site $\times$ browse	2	27.70	5.30	0.0052
Site $\times$ competitor removal	2	27.53	5.27	0.0054
Browse $\times$ competitor removal	1	0.005	0.00	0.9753
Error	595			
b) Aspen diameter ( $r^2 = 0.33$ )				
Site	2	78.02	135.54	0.0001
Browse	1	3.92	6.82	0.0092
Competitor removal	1	10.65	18.51	0.0001
Replicate	2	1.35	2.35	0.0962
Site $\times$ browse	2	1.52	2.64	0.0725
Site $\times$ competitor removal	2	1.41	2.45	0.0869
Browse $\times$ competitor removal	1	0.014	0.02	0.876
Error	595			

Notes: A  $2 \times 2$  factorial design (browsing, competitor removal) was conducted at each of three sites, with three replicates of each treatment ( $n = 12$  plots per site). The  $r^2$  values are for a model in which all factors were fully nested by year.

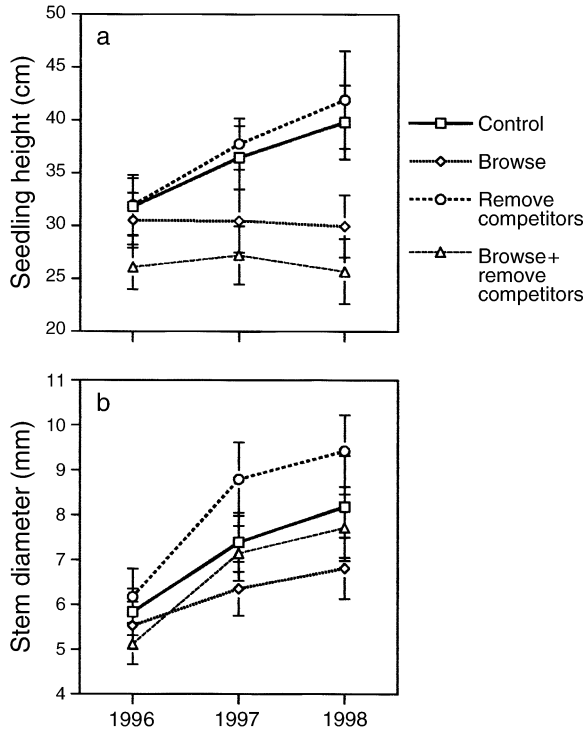


FIG. 2. Mean aspen seedling heights and diameter by experimental treatment across all three geographic locations, 1996-1998. Error bars are 95% confidence intervals ( $\pm 2$  SE).

between 1996 and 1998, although it did increase from 1998 to 2000 (Fig. 3a). Aspen seedlings elongated most at the low-elevation Fountain site. Stem diameters were significantly greater at the Fern site compared to either Lewis or Fountain, although increases were observed at all three sites (Fig. 3b). In the plots that were re-measured in 2000, the tallest aspen seedlings were observed at the Fountain site (mean = 61 cm, maximum = 194 cm). At Fern and Lewis, mean heights in 2000 were 48 cm (maximum, 102 cm) and 33 cm (maximum, 102 cm), respectively. Mortality was highest at the Lewis site, where 29% of 59 plants tagged in 1996 had died by 2000. At the Fountain site, 16% of 67 initially tagged plants had died, and at Fern only 7% of 66 initial plants had died.

DISCUSSION

We took advantage of a rare episode of seedling establishment following the 1988 Yellowstone fires to investigate the early patterns and mechanisms of genet recruitment in a long-lived clonal plant species, using a combination of survey and experimental methods. We first address the four questions that were raised in the *Introduction*, and then conclude with an overall assessment of the significance and likely outcome of this rare seedling event in quaking aspen.

*Questions 1 and 2: timing of seedling establishment and extent of clonal development*

Our intensive analysis of seedling age structure in 1996 did not entirely support our earlier impression, based on a limited survey (Romme et al. 1997), that nearly all of the aspen seedlings germinated in the first two years after the 1988 fires, because we found seedlings dating from every year between 1989 and 1996. Kay (1993) also reported seedling establishment in years subsequent to 1990. However, more seedlings dated from 1989 (36%) than from any other year in our study areas, and 65% of seedlings established within the first three years, with progressively fewer seedlings in each successive year (Table 3). In our subsequent field studies in Yellowstone, we have not observed new aspen seedlings in the areas burned in 1988 (though they have appeared in other areas that burned in 2000). Moreover, the data in Table 3 represent a static age distribution; some unknown fraction of the original 1989 cohort probably died before our survey in 1996. Thus, the overall picture of a relatively brief period of establishment (a “window of opportunity”) is supported by our analysis of seedling age structure. Studies with other clonal species have demonstrated similar brief periods of seedling establishment when both seeds

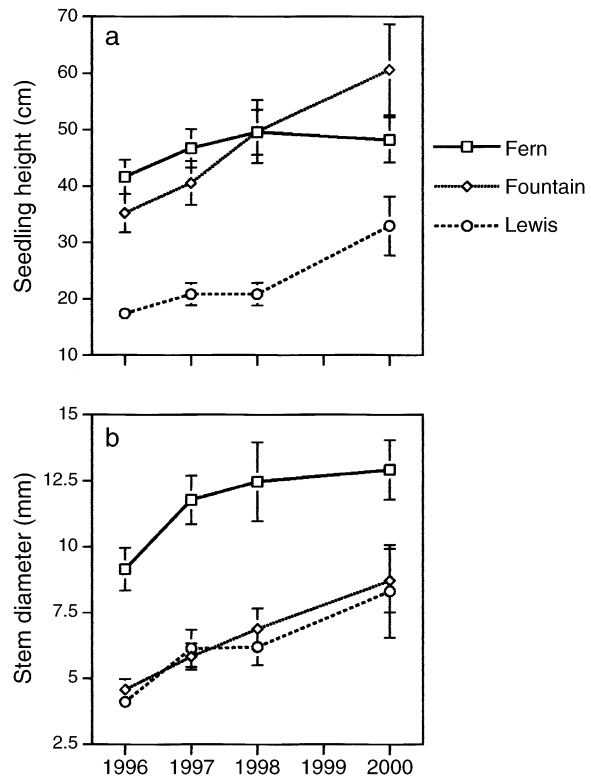


FIG. 3. Mean aspen seedling heights and diameters among geographic locations for control and competitor removal plots only (i.e., clipped treatments were excluded, and all plots were fenced to prevent browsing by native ungulates). Error bars are 95% confidence intervals ( $\pm 2$  SE).

and favorable substrates were available (see references cited in Eriksson [1993], also Eriksson and Froberg [1996]).

Our excavations and genetic analyses also did not confirm our earlier impression (Romme et al. 1997) of extensive clonal development in the new aspen populations. On the contrary, only 13 stems out of 173 sampled (8%) had formed ramets by 1996. Our earlier interpretations were based on a limited excavation of about 10 plants that happened by chance to include two pairs of physically connected stems (which we now know to be relatively rare in these populations) plus surface observations that stems dating from 1990 or 1991 were often in close proximity to stems dating from 1989. However, even if our earlier report suggested greater clonal development than we observed here, it is nevertheless noteworthy that at least some of the ortets began producing ramets while <7 yr old. Day (1944) also reported a 6-yr-old ramet on an 8-yr-old ortet in Michigan. Temporal patterns in formation of aspen root sprouts (ramets) and the morphological and physiological factors that control sprouting in mature aspen root systems are reviewed by Schier (1973a, b, 1975b), Schier and Zasada (1973), and Shepperd (1993). Presumably, similar patterns and controls apply to development of clonal structure in ortets, but it was beyond the scope of this study to address such questions. Future genetic analyses similar to what we did in 1996 will be warranted to determine the extent of clonal development as the ortets become older.

### *Question 3: landscape patterns of seedling growth and persistence*

Our surveys of 22 permanently marked aspen seedling plots from 1996 to 2000 revealed that these seedling populations persisted—but generally did not flourish. None of the 22 populations that we originally sampled in 1996 had been extirpated by 2000, but overall mortality was relatively high (~25% over a 4-yr period). Moreover, most individuals in the 22 plots did not grow appreciably in height or diameter during the period from 1996 to 2000 (Tables 2 and 4). Nevertheless, there was substantial variability among the 22 plots (Tables 2, 5, and 6). Survival, height, and diameter growth generally were greater in aspen seedlings growing in what appear to be better soils (i.e., less acidic), a pattern consistent with other studies of aspen that have demonstrated strong positive growth responses to soil nutrient availability (Kinney and Lindroth 1997, Hobbie and Chapin 1998, Lu and Sucoff 2001, Fraser et al. 2002, DesRochers et al. 2003). The significant predictors of mortality differed somewhat for the periods 1996–1998 and 1996–2000, and overall model  $r^2$  was lower for the latter period, but pH and elevation were positively associated with survival in both models (Table 5). Soil potassium was negatively associated with seedling survival, but positively associated with height (Tables 5 and 6). This seems con-

tradictory and may be spurious; at any rate, potassium contributed a relatively small partial  $r^2$  to both models. The apparent relationships of stem diameter with lodgepole pine sapling density and distance from pre-1988 aspen populations (Table 6) almost certainly are spurious: our experimental studies demonstrated no significant effect of removing lodgepole pine competitors, and several of the plots having higher soil pH also happen to be located far from pre-1988 aspen stands, e.g., near the southern portion of Yellowstone Lake (Fig. 1).

Wolves were reintroduced into Yellowstone in 1995, and Ripple and Larsen (2000), working with resprouting adult aspen in a lower-elevation landscape in northern YNP, reported that aspen stems located near wolf dens grew more rapidly than stems remote from wolf dens. The apparent mechanism was a reduction in elk browsing pressure because of the presence of the wolves. Wolves established territories near some of our aspen plots by 2001 (Smith et al. 2003), so we tested for an effect of distance to the nearest wolf den on mean and maximum aspen height in 2000 within our 22 plots. A simple regression revealed a negative relationship between mean seedling height and distance from a wolf den ( $r^2 = 0.23$ ). However, distance from wolf dens was also positively correlated with soil pH. In a multiple regression utilizing all variables, only pH and available potassium were significant, and distance to wolf dens was no longer significant. Therefore, we find only equivocal evidence that wolves may have enhanced aspen seedling growth and survival by reducing ungulate browsing pressure, but acknowledge that such an effect may become more important in the future.

Overall, the environmental variables that we measured explained only 56% and 62% of the variability in stem height and diameter, respectively (Table 6). Some of the additional variability may be related to genetic differences that we have not yet investigated (Jelinski 1993). For example, carbon allocations to root growth, shoot growth, and disease resistance appear to be under strong genetic control (Pregitzer and Friend 1996), and a positive correlation between plant growth rate and amount of enzyme heterozygosity has been documented in two previous aspen studies (summarized by Mitton and Grant 1996). Considerable genotypic variation also has been observed in general aspen phytochemistry and resistance to herbivory (Osier and Lindroth 2001, Lindroth et al. 2002).

Finally, although we documented some interesting environmental patterns in seedling survival and growth rates, it is noteworthy that even the tallest stems in our 22 plots were still <1 m tall in 2000. This height is well within the reach of an elk or moose (*Alces alces*), and indeed, most of the stems showed evidence of previous browsing in every year that they were sampled. The combination of slow height growth, heavy browsing, and mortality that we observed from 1996 through

2000 throughout the study area, suggests that few of the aspen seedlings are likely to ever achieve canopy stature, at least not under current environmental conditions. Ungulate browsing undoubtedly is a major contributing factor to the poor growth and survival of the aspen seedlings, because average heights were consistently greater in the exclosures where plants were protected from browsing than in unprotected locations throughout the park. However, browsing is not the whole story, as revealed by our experimental study which we now discuss.

*Question 4: mechanisms controlling aspen seedling persistence and growth*

Our two experimental treatments that simulated browsing (clipping and clipping plus removal of competitors) conclusively demonstrated that continual removal of annual growth prevented stem height growth (although some diameter increase did occur). This result was hardly surprising. However, two unexpected findings did emerge from our experimental study. First, removal of competitors, both lodgepole pine saplings and herbaceous plants, had no significant effect on height growth of aspen seedlings, although it did increase diameter growth (Table 7, Fig. 2). Evidently aspen seedling height growth was not being inhibited by interspecific competition at this stage of development, although competition may have been important when the seedlings were younger (Barnes 1966).

The second surprising result was that most aspen stems grew very slowly even when protected from browsing. Average height increment in unclipped stems within the exclosures (the control plus competitor-removal plots combined) during the 4-yr period from 1996 to 2000 was <10 cm at the Fern Cascades site, ~15 cm at the Lewis site, and ~25 cm at the Fountain site (Fig. 3). We observed die-back of the dominant leaders of many unclipped plants (from ~25% of leaders at Fountain to ~90% at Fern and Lewis), as the terminal bud and distal portion of the stem turned black and withered away. A lateral stem from lower on the main stem then assumed dominance for a year or two, before it too died back. The cause of this die-back phenomenon is unknown, but it contributed to the very slow increase in height of un-clipped plants. We also observed considerable mortality from 1996 to 2000 of the stems that we initially marked for measurements in the unclipped exclosures (6% at Fern, 16% at Fountain, and 29% at Lewis).

The slow growth and substantial mortality of aspen seedlings protected from ungulate browsing may be simply the result of adverse local climatic conditions at the exclosure sites. The Fern Cascades and Lewis sites are ~200 and 350 m higher in elevation than the pre-1988 range of aspen in YNP, so the slow rates of growth and high mortality may be a function of low temperatures and short growing seasons. Barnes (1966) reported high mortality of seedlings in locations subject

to subfreezing summer temperatures in Michigan. At our Lewis site in particular, exceptionally deep snow-pack and late snowmelt in 1996 and 1997 resulted in pools of standing water within some of the exclosures and probably also maintained low soil temperatures and hypoxia within the root zone of many of the aspen seedlings (Neuman et al. 1996). Low soil temperatures have been shown to reduce hydraulic conductance and sucker formation in aspen roots (Wan et al. 2001, Fraser et al. 2002). In contrast, the Fountain site lies within the pre-1988 elevational range of aspen (though just barely), and it was in this site that we measured the greatest height growth of protected stems.

*Summary and conclusions*

What is the likely long-term fate and significance of the new quaking aspen genets that appeared in the first few years after the 1988 Yellowstone fires? A commonly used model for development of clonal plant populations (e.g., Fig. 1 in Eriksson 1993) predicts an initially large cohort of new genets, followed quickly by a substantial loss of genets as selection removes genetic individuals that are poorly adapted to current conditions or those that initially established in what is unsuitable habitat over longer time periods. While the number of genets decreases, the number of ramets increases, as successful genets begin to develop clonal structure. Eventually, local populations may become dominated by many ramets of only a few genetic individuals.

A long-term population process of this kind may now be occurring with the new aspen genets in YNP. The 1988 fires created a large area of bare substrate, which, combined with favorable climatic conditions and adequate seed production during the next few years, resulted in a rare "window of opportunity" for aspen seedling germination and establishment across a large portion of the park (Jelinski and Cheliak 1992, Eriksson and Froborg 1996, Romme et al. 1997). Seedlings became established soon after the 1988 fires in many places, including locations where aspen was formerly not present, and possibly including places where local soils, micro-climate, and browsing pressure were not suitable for long-term persistence. We do not have mortality data for the period 1989–1996, nor have we explicitly sought evidence for differential mortality among genotypes which would demonstrate selection. Nevertheless, the present study found a relatively high mortality rate (~25%) between 1996 and 2000 within a spatially extensive sample of seedlings. Our exclosure experiments also demonstrate that many individual genets have been unable to grow or persist even when protected from browsing, presumably because they cannot tolerate local climate and soils conditions. These observations are consistent with Eriksson's (1993) model of high early mortality in a new cohort. On the other hand, many individuals have survived and continue to increase in height and diameter (albeit

slowly), even in the high-elevation Lewis site which appears to be the least favorable of our three experimental sites because of its short growing season and propensity for excessive soil moisture. The rate of mortality also appears to have slowed between 1996–1998 and 1998–2000: 22 of our widely distributed permanent plots show a loss of seedlings from 1996 to 1998, but nine of those plots (41%) show no further loss from 1998 to 2000 (Table 2). These observations are consistent with the idea that mortality rates may have begun to decline and that many of the extant individuals will persist. It obviously is too soon to make confident interpretations or predictions about the long-term population processes now occurring, but future re-sampling of these permanent plots will enable us to clarify long-term trends in demographic and genetic structure of these seedling populations, and will allow a fuller test of the Eriksson (1993) model.

As of 1996 (when the oldest aspen ortets were 7 yr old), only a small fraction (<10%) had produced ramets, as shown by our intensive excavations and genetic analyses of three different seedling populations. It is interesting, however, that a few individuals did in fact begin to develop clonal structure at such an early age. We predict that ramet production will become more common as the populations age and as the number of genets continues to decrease. Genetic diversity, initially high because of the large number of unique genets, probably will gradually decrease at a local scale (Tuskan et al. 1996), but will remain high at broad spatial scales because of different selection pressures in different areas plus occasional recruitment events as new “windows of opportunity” occur (Jelinski and Cheliak 1992, Ellstrand and Roose 1987).

If mortality continues at a rate similar to that documented between 1996 and 2000, and especially if a very high proportion of seedlings continue to show little substantial height growth, then many or most of the aspen seedlings that established after 1988 probably will disappear, perhaps over the next one to two decades. It is even possible that none of the post-1988 seedlings will survive under current climatic and browsing conditions. However, we predict that at least some of the new genets will survive and will produce new aspen clones. These new clones could potentially survive for many centuries or millennia, regenerating by asexual root sprouting, in a manner similar to the clones that established hundreds or thousands of years ago (Mitton and Grant 1996). Thus, the “window of opportunity” that followed the 1988 Yellowstone fires may have a lasting legacy in the demographic and genetic structure of quaking aspen populations in the Yellowstone region. However, because the number of genets likely to persist appears to be relatively small, broad-scale vegetation patterns and postfire successional trends in the Yellowstone landscape probably will not be significantly altered.

Where in the landscape are new aspen genets most likely to persist? We suggest at least two kinds of places. The first is on relatively fertile soils at higher elevations, where our analyses indicated the greatest survival and growth (Table 2). For example, in plots located in the Yellowstone Lake region (Fig. 1, Table 2), mortality was relatively low from 1996 to 2000 (<20% in all four of the plots located in this region) and many individual stems were approaching 1 m in height by 2000 (Table 2). This area is at relatively high elevation (~2400 m), where winter snows are deep (thus discouraging ungulate browsing), but soils are well drained in summer and are derived from andesite and lake bottom substrates that are somewhat more fertile than soils derived from rhyolite and tuff (Despain 1991). The second kind of place where new aspen genets appear likely to persist is at lower elevations (<2100 m), near or within the pre-1988 distribution of aspen, where a dense stand of lodgepole pine forest burned in 1988 and the dead trees have since fallen to produce a tangle of logs up to 1.5 m deep. This mass of fallen trees may discourage ungulates from browsing the aspen growing within the tangle, and the relatively long growing season at lower elevations may promote relatively rapid height growth. Ripple and Larsen (2001), working with aspen root sprouts in northern YNP, documented greater height growth within dense tangles of fallen fire-killed trees than in more exposed locations, and a similar pattern may be expected for the aspen seedlings. None of our 22 extensive plots was located in a such a mass of fallen trees, but this situation is well represented at our Fountain enclosure site (Fig. 1, Table 1). Mean height of unclipped aspen stems in the enclosures at this site was 60 cm in 2000 (the tallest of any site), with many stems >1 m. We discontinued maintenance of the enclosures at this site in 2000, because of the damage from falling trees, but browsing has been minimal even without the fences. Indeed, we have seen little evidence of browsing since the mid-1990s throughout this area of dense fallen trees, and the height and growth form of the post-1988 aspen genets within a several-hectare area surrounding the Fountain site are similar to those inside the former enclosures. One individual aspen stem had reached 2.6 m in height by 2003, with a straight growth form and a single dominant leader, and many others were ~2 m tall.

In sum, we have used a combination of survey and experimental methods to document the early patterns and mechanisms of establishment, growth, and persistence of an unexpected cohort of new quaking aspen genets resulting from a rare “window of opportunity” created by the 1988 Yellowstone fires. Throughout the first decade after the fires, the population was still in an early stage of what we anticipate will be a long-term developmental process, a stage characterized by relatively high mortality of genets and only the beginnings of ramet production. This rare seedling recruit-

ment event in the aftermath of the 1988 Yellowstone fires provides an unusual opportunity for long-term study and understanding of the effects of large, infrequent natural disturbances on demographic and genetic processes in long-lived clonal plant species.

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