

## SEXUAL DISTRIBUTION OF ASPEN IN THE SIERRA NEVADA

DAVID BURTON, Aspen Delineation Project, 2070 Orange Drive, Penryn, CA, USA

**ABSTRACT:** A study to establish a more complete understanding of the sexual diversity of aspen in the Sierra Nevada was undertaken in 2001 in order to test the hypothesis that there are only male aspen clones on the Western Slope of the range and thus only vegetative reproduction of aspen would be possible. Transects identifying the presence of both male and female clones were completed in the Eldorado and Tahoe National Forests in the spring of 2001. These transects produced nearly identical male to female clone ratios of 2:1.

**Key words:** Aspen, *Populus tremuloides*, sexual identification, sexual distribution, flowers, catkin, Sierra Nevada, California

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A study to establish a more complete understanding of the sexual diversity of aspen, *Populus tremuloides*, in the Sierra Nevada was undertaken in 2001. This study was part of a larger project which included a study of the genetic diversity of aspen in the Eldorado National Forest (Hipkins and Kitzmiller 2004). This sexual diversity study was designed to address the hypothesis set forth in some Sierra Nevada reference sources that there are only male aspen trees (clones) found on the western slope of the Sierra Nevada (Johnson 1994). The earliest known published reference to this hypothesis is in Judy Hutchinson and G. Ledyard Stebbins' *A Flora of the Wrights Lake Area* (1986). If this hypothesis were confirmed by this study, the results would vary from previous gender distribution studies of aspen conducted in North America. In those studies female catkins have always been found (Grant and Mitton 1979). If it were confirmed that only males exist, it would make aspen on the western slope of the Sierra Nevada biologically different from any other studied aspen habitat in North America. Additionally, if this hypothesis were proven true, it would demonstrate that sexual reproduction of aspen would not be possible on the western slope of the Sierra Nevada since female clones are a necessary element for sexual reproduction.

### OVERVIEW

Because seed bed conditions and soil water demands during initial germination and growing season are so critical to successful aspen seedling establishment (McDonough 1979), it is believed that the principal means of aspen reproduction in the West is vegetative (Jelinski and Cheliak 1992). In the vegetative process, adventitious shoots (ramets) grow from lateral roots of a parent tree producing suckers as far as 75 feet away from the parent (Shepperd 2001). After a disturbance in a stand of aspen, as from fire, clearcutting, or avalanche, as many as 100,000 ramets per acre can occur (Bartos et al. 1991). Because it is believed that vegetative reproduction has continued over an extended period, it is hypothesized

that many clones in the Intermountain West may be thousands of years old (Einspahr and Winton 1976, Jelinski and Cheliak 1992), having reproduced over and over again asexually. However, although rare, seeding events have been recorded in the Rocky Mountains (Romme et al. 1997, Kay 1993). This study looks at the possibility of one of these rare events being able to occur in the Sierra Nevada.

It is known that aspen is dioecious (Jones and DeByle 1985): individual trees are either male or female with male (staminate) and female (pistillate) flowers found in catkins on separate stems (Fig. 1 and Fig. 2). Trees flower several weeks before the leaves appear (Jones and DeByle 1985), with both male and female clones producing catkins with flowers of their own sex. Only on rare occasions have hermaphroditic flowers been reported in aspen catkins (Einspahr 1960, Maini and Coupland 1964). Studies have shown that aspen produce an abundance of seeds generally every two to five years after trees reach an average age of fourteen (Einspahr and Winton 1976). Both sexual or vegetative reproduction repeated over time will produce stands that consist either of a single multi-stemmed clone of one sex or of multiple clones, each having its specific sex (Hipkins and Kitzmiller 2004).

### METHODS

To establish the sexual distribution of clones, two transects across the western slope of the Central Sierra Nevada were conducted during the spring of 2001. The first twenty-five mile long transect ran east to west in the Echo Summit/Carson Pass region of the Eldorado National Forest—ranging in elevation from 5300 feet to 8200 feet. The second transect ran for 18 miles—east to west—along the Interstate 80 corridor in the Tahoe National



Forest— stands ranging in elevation from 5100 feet to 7200 feet.

Observations of stands began on April 1, 2001, and the last observation was completed on May 12, 2001. In nearly all cases, leaf (vegetative) buds had not yet flushed and there was still snow under much of the aspen canopy. Branches with catkins were cut with an extended pole clip when necessary, and the catkins were observed by hand lens. When possible, additional identifications were made with a spotting scope for catkins that were unreachable with the pole clip. When catkins could not be differentiated with the spotting scope, they were not included in the findings. Additionally, some branches whose buds had not yet opened, were collected, tagged, and placed in water in a greenhouse to force bud flushing, and the flower's sex was recorded once the catkins appeared.

Because this study was designed to discover whether female clones exist on the Western slope, only one male clone and one female clone were recorded per stand if catkins of both sexes appeared. While this protocol might fail to identify some clones, it did prevent recording flowers from the same clones more than once. A separate genetic study started in the Eldorado National Forest in 2001, while not addressing the sexual distribution of clones, accurately established the actual number of clones in each stand studied. In that genetic study, 663 leaf samples were taken from individual stems in 82

stands. Electrophoresis analysis showed that there were 2.6 clones per stand with a range of one clone to eleven clones per stand (Hipkins and Kitzmiller 2004).

#### FINDINGS AND DISCUSSION

Nearly identical 2:1 male to female ratios were observed in both transects. The Eldorado transect was found to have both staminate (male flowers) and pistillate (female flowers) catkins. Flowers were recorded in twenty-four (24) identifiable clones in this transect. Fifteen (15) stands had male clones, seven (7) stands had female clones, and one stand was identified as having two clones, one of each sex. The Tahoe transect had twenty-three (23) stands with identifiable clones. Fourteen (14) stands had a male clone present, seven (7) stands had a female clone, and one stand was found to have clones of each sex. Additionally, one clone in the Tahoe transect was found to contain both male and female flowers. This hermaphroditic flowering contained catkins with both pistillate and staminate flowers.

Since the protocol of this study identified only flowering stems and identified only one possible clone of a given sex in a stand, it is understood that this study only established the presence or absence of female or male clones on the western slope of the Sierra Nevada and did not attempt to address the question of the actual sexual ratio of clones. Thus, given the limitation of the protocol, there is no way of knowing for certain if the ratio of male to female clones is the actual sexual distribution of clones in the study area. However, it is interesting to see how these transects compare to similar studies. This 2:1 male to female ratio deviates from the expected 1:1 ratio found



Figure 1. Male (staminate) flower distinguishable by its four-lobed anther. The image was taken by Andrew Groover, Institute of Forest Genetics, USDA Forest Service. Tissue for scanning electron microscopy was fixed with FAA, dehydrated, subjected to critical point drying using a Tousimis Samdri 780A, and coated with gold using a Denton Desk II sputter coater. Scanning electron microscopy was performed with a Hitachi S3500N under high vacuum in-depth mode with accelerating voltage of 5kV.



Figure 2. Female (pistillate) flower distinguishable initially by its wing like reddish stigmas and then by its green, conical ovaries. The image was taken by Andrew Groover, Institute of Forest Genetics, USDA Forest Service. See Fig.1 for details on how the image was captured.

in some research (Einspahr 1962), but is in keeping with the result of other studies (Grant and Mitton 1979). Besides the restrictions of the study protocol, the deviation from 1:1 could be explained as not the true ratio of sex found in the sampled stands but rather the ratio of flowering for the year of the 2001 study. Additionally, it has been found in Colorado that although a 1:1 ratio was found in stands on the western slope of the front range of the Rocky Mountains, a significant change in the sexual ratio of clones occurred when stand elevation was taken into consideration (Grant and Mitton 1979). That study found that there was a decreasing female proportion as a function of elevation. This variable could account for the ratio found in this study, but because of the sample size there was no attempt to find a correlation between sexual distribution and elevation. The observation of the bisexual flowers, while rare, has been reported in the past (Maini and Coupland 1964). After review of existing literature, it was hypothesized by Maini and Coupland that as many as five percent of all clones may be bisexual.

#### CONCLUSIONS

The hypothesis that there are only male aspen trees (clones) on the western slopes of the Sierra Nevada is not accurate—at least in the Central Sierra. Further research could be conducted to see if the prevalence for males over females as discovered in the two transects in this study is consistent, or whether there is a ratio closer to 1:1. This can be accomplished in the Eldorado National Forest by identifying sexual distribution in stands containing catkins in which clonal distribution has been established during the 2001 genetic analysis study. Variable attributes like seasonal flowering variations, stand elevation, or seed viability could also be examined.

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