The Variation in Leaf-Edge Serration

by
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Abstract

Populus tremuloides have leaves that have a quantitatively measurable average serration, or rugosity. This project aims to calculate and determine whether or not there is a relationship that describes the rugosity with respect to genetic differences. We hypothesize that there will be a distinct relationship between leaf-margin rugosity and genetic differences. Looking at a specific population of aspen trees in Big Cottonwood Canyon, Utah, we will collect data and run necessary tests to determine said relationship. If our hypothesis is correct, it would help to determine that, along with environmental factors, genetic tendencies are a player in the development of leaf margin formation.

Introduction

Variation in leaf serration affects the ecology of any tree in several ways. Spatial patterning of serrations is shown to depend on the formation of auxin maxima, which are regions that have cells with higher concentrations of auxin, a plant hormone that controls growth (Kawamura *et. al.*), therefore auxin may have a role in regulating the serration of the leaf. Auxin, however, also affects other processes in the leaf, including venation patterns (Pahari *et. al.*). Venation patterns affect the distribution of water through the leaf, which in turn affects the amount of water that can be transpired from the leaf. Transpiration affects the thermodynamics of the leaves, showing how they expel excess heat absorbed throughout the day, and also how much water will be lost. Lobed leaves are more hydraulically efficient because they have fewer minor veins that have a lower leaf hydraulic resistance, which is a property that measures the difficulty of transporting water from the roots to the edges of the leaves (Nicotra *et. al.*). However, lobes are a larger-leaf quality, while serration is more prevalent on small leaves. The relationship between serration and thermodynamics in the leaf has not been explored, but may have a similar function to the lobes in larger-leaved trees. Serration and venation, since they are both controlled by auxin, may be related; therefore serration may be a phenotypic indicator of important thermodynamic processes in the leaf.

Quaking aspens (*Populus tremuloides*) are a unique species of tree because they have the ability to reproduce sexually and asexually. Asexual reproduction in *Populus tremuloides* creates nearly genetically identical groups, called ramets, of a single genet, or genotype. Within these clones, much of the biomass is in the leaves, and thus much of the phenotypic variation between the trees can be found in the qualities of the leaves, for example variation in leaf area, leaf color, venation, petiole length, and serration of edges. In this paper, we will be focusing specifically on diversity of leaf margin serration for leaves gathered from a group of trees in Big Cottonwood Canyon, Utah.

We studied the extent to which the leaf margin serration in *Populus tremuloides* can be attributed to genetic variation. A program in Python will determine the average rugosity of leaf edges, and it will be applied to different genets. We predict that variation is most likely to be more significant between different clones and less significant within a single ramet or genet, and that variation in serration may be dependent on leaf size as well as other factors in the genotype.

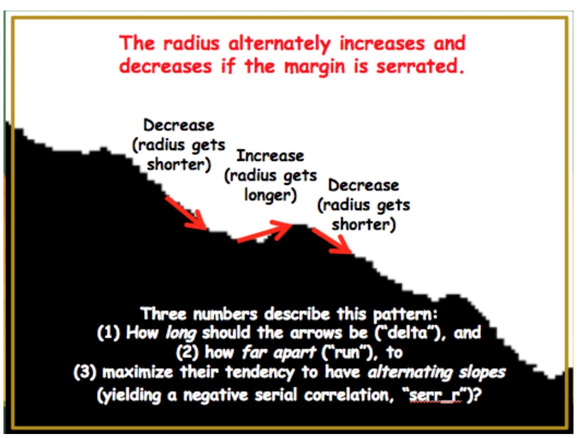
Methodology

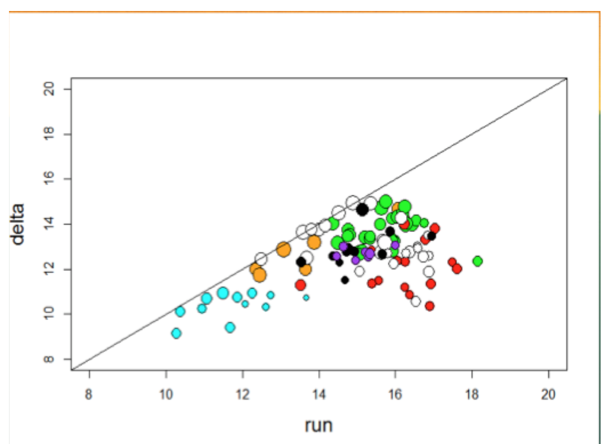
Leaf samples were collected randomly from about 140 trees located within a blank range in Big Cottonwood Canyon, Utah, with an average of 30 leaves per tree. DNA was extracted from leaf samples from all of the trees and run using a microsatellite analysis procedure. Trees with identical microsatellites were determined to be clones.

Using ImageJ, we converted all of the pixels to black or white on the leaves to make running the R program easier. A script was written that gave three numbers to describe a leaf radius and margin pattern: (1) How long should the arrows be ("delta"), and (2) how far apart ("run"), to (3) maximize their tendency to have alternating slopes (yielding a negative serial correlation, "serr_r")? These can be visually shown and understood based on "Figure 1" below. Least-square regression lines were created to fit a plot built on the data outputs in R.

Rugosity is traditionally defined as the measurement of small-scale height variations on a surface; however we measured the rugosity as the change in slope of the tooth edges. This works because it shows the relative length of the tooth edges over a distance, so a larger magnitude serr_r value indicates that there is more serration.

Using the database of leaves from clones that had been genotyped and named c12, c14, c4;6, c5a, c8, and c9, we analyzed the relationship between serration (serr_r) and genet, ploidy, ramet, and leaf area using R to determine the source of phenotypic variation of leaf edge serration.

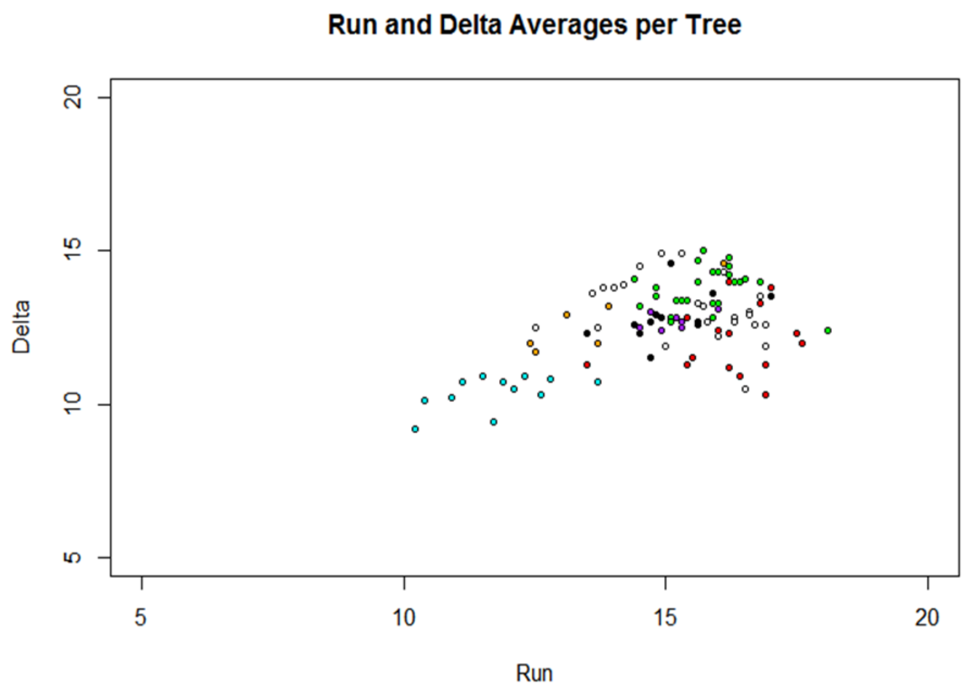


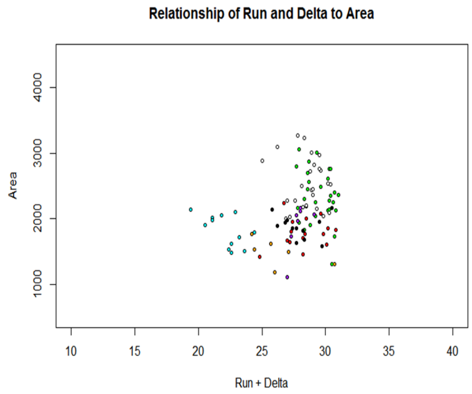


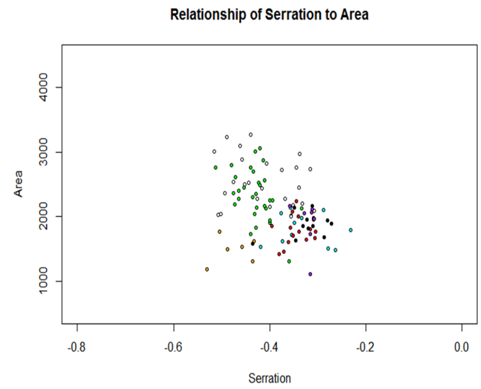
Results

There were significant p-values for nearly every analysis we performed, so numerous conclusions could have been made about the relation of rugosity to various aspects of the trees. This indicated that we should look for the most influential trait. An original thought, or a focus for our hypothesis, had been that area of the leaf seemed to have an influence on the serration of the leaves, because larger leaves appeared to be more serrated than magnified versions of smaller leaves. However, as seen by the graphic indicating the relationship between run and delta and area, there appeared to be no correlation between rugosity and leaf area. The same goes for the relationship between the serration score and area. Running an analysis of both serration to area, and run and delta to area, we found significant result, but as stated earlier, significance is easy to have when the sample size is as large as this. Area only described about %14 of both serration and run/delta.

The next test performed was the relationship between run and delta averages per tree. Trees from the same genet tended to have similar relationships between run and delta, showing that serration did vary between different genotypes. The linear slope of run and delta for clone fourteen (in cyan) was especially striking, as shown in the graphic below. An analysis of variance table was run to determine the correlation between individual leaves and rugosity or different clones and rugosity. The correlation between clones and rugosity was very significant; about 70% of the variation in run plus delta could be explained by clone, and about 50% of the variation in serration score could be explained by clone. In contrast, individual leaves accounted for only 14% of variance in run plus delta and 13% of variance in serration score. Thus it can be concluded that clone is the primary source of variance in rugosity.







Discussion

This study frames several other important questions. Ploidy plays a small role in determining run and delta of a leaf, but overall was insignificant in determining the variance of rugosity in *Populus tremuloides*. If rugosity is a genetically determined trait, why doesn't ploidy play a bigger role? In addition, both the correlation between run and delta and serration score were used to determine rugosity, but when serration and run plus delta were put through a linear regression, they had little to no relationship. Could rugosity be affected by several unlinked genes? Finally, it is important to question what relationship rugosity may have to the ecology of the tree.

While we had predicted that leaf size may affect rugosity, the results of the experiment suggested otherwise. Auxin determines the amount of serration in a leaf, and since auxin production is genetically determined, leaves from the same trees should have the same patterns of auxin production. So leaves from the same tree should have similar run and delta and serration scores regardless of their area. Thus rugosity may not be affected by leaf area because leaves from the same tree carry the same genetic material and will thus have similar rugosity values.

There may have been some minor issues with the validity of our study. The leaves were sampled as randomly as possible, but generally were taken from the lower branches in groups. There might have been some small effect on results because of this sampling methods. Human error always plays a role in using large data tables. An incorrectly entered data point, or a forgotten decimal could change the results.

Despite these issues, rugosity is clearly affected by clonality. Trees from the same genet would have the similar run and delta correlations and similar serration scores, and would vary considerably from trees of a separate genotype. A study of this size and focus has never been done on aspens, and despite the possible sampling errors, our data indicates that results can be inferred from this in that variation in rugosity is genetically determined, indicating that the gradient of different serrations exist either as part of natural selection--whichever type of serration is more suited to the environment will eventually become the most dominant--or that serration, while perhaps not directly affecting a tree's ecology, is tied to other traits that vary on a genetic basis.

References

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