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Morphology and Nutrient of Leaf in *Quercus castaneifolia* Seedling as Affected by Blackberry (*Rubus fruticosus* L.)

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Abstract: Morphology and nutrient of leaf in *Quercus castaneifolia* (C.A.Meyer) seedlings in competition with blackberry (*Rubus fruticosus* L.) was investigated in an intervened lowland forest of Noor city, north of Iran. The research was conducted as factorial experiment in three replications. For this purpose, seedlings of small (3 mm) and large (4 mm) in collar diameter (CD) were sown in two treatments (i) covered with *Rubus* and (ii) moved from *Rubus* in 0.5 m radius around the *Quercus* seedling, replicated three times. In the end of the first growing season, leaf samples were randomly taken from 9 seedlings in each treatment in order to determine leaf number, leaf dry matter, leaf area, growth flush number, apical shoot growth and concentrations of leaf nutrient (N, P, Mg and Ca). Leaf dry matter did not differ among the treatments. Leaf number and leaf area were greatest with large CD seedlings growing in moved *Rubus* area. The greatest apical shoot growth was observed with large CD seedlings growing in full *Rubus* area. The same as growth flush number, concentration of P, N and Mg was greatest with small CD seedlings in moved *Rubus* area. From this research it can be deduced that the seedlings growing in moved *Rubus* area produce greater plant biomass and mineral nutrient matter.

Key words: Competition, nutrient, *Quercus castaneifolia* (C.A.Meyer), *Rubus fruticosus* L., seedling morphology

INTRODUCTION

Quercus castaneifolia (C.A. Meyer) is one of the most prevalent species growing in the Caspian forests, north of Iran. Reduction of standing volume as well as defect of natural regeneration in recent decades in *Q. castaneifolia* stands has made concerned Iranian silviculturists. To this reason, its natural regeneration problem has led forest managers to use the artificial regeneration, where regeneration establishment is difficult (Tabari *et al.*, 2007). Researchers have identified several factors that may reduce oak seedling survival, including damage by deer and cattle (Zachary *et al.*, 2007; William and Beschta, 2008), acorn predation by rodents (Wang and Gao, 2006) and competitive suppression by exotic annual grasses and shrubs (Gordon and Rice, 2000; Dickie *et al.*, 2007). In literature has been referred that blackberry (*Rubus* spp.) directly compete with seedlings for soil moisture and nutrients and has negatively effect on their survival and growth. Blackberry often dominates successional stages in areas that are suitable for the establishment of forests (Torres, 2003). Dense blackberry layers may hamper the establishment of forest species (Sans *et al.*, 1998; Chambers *et al.*, 1999; Castro *et al.*, 2002). The surface areas covered by oak forest have declined in Asia, Europe and North America (Kelly, 2002; Standiford, 2002). A concern on the future of oak forests has fostered vast restoration programs in recent years. In addition to the large-scale reduction in surface area, oaks frequently show poor recruitment (Torres, 2003). The reasons for this may be numerous, including lower dispersion, higher rodent predation, interference with extant vegetation and unfavorable climatic conditions

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for seedling establishment (Pons and Pausas, 2006). Whether *Q. castaneifolia* establishment may be favored by blackberry clearing, has been the subject of debate. The positive effect of shading can be strong enough to compensate the negative effects of competition with neighboring vegetation. To our knowledge, no study has evaluated the effect of different intensities of blackberry clearing on the establishment of *Q. castaneifolia*. We determine the effect of blackberry clearing and collar diameter of planted seedlings on characteristics of morphology and nutrient of leaf in *Q. castaneifolia* seedlings and discuss the results on the basis of their interactions on these characteristics. At a management level, results provide relevant information to generate recommendations on the best practices to favor *Q. castaneifolia* establishment in these areas.

MATERIALS AND METHODS

A field experiment was carried out at Tarbiat Modares University, north of Iran (51° 46' E, 36° 47' N, -15 m a.s.l.) (Fig. 1). The experiment was conducted in a flat, deep soil and homogenous area that was formerly a mixed oak stand. In the experimentation year total rainfall was 875.6 mm, mean annual temperature was 15.2°C and mean relative humidity 79%. The climatology census of this year is given in Table 1. In this research, in March 2007, 162 *Quercus* seedlings of small (3±0.3 mm)

Table 1: The climatology census of study area based on meteorological data of Nowshahr (51° 45' E, 36° 47' N, -20 m a.s.l.)

| Months | Temperature (°C) | Precipitation (mm) | Relative humidity (%) |
|-----------|------------------|--------------------|-----------------------|
| December | 10.3 | 146.2 | 82 |
| January | -5.0 | 34.3 | 74 |
| February | 5.2 | 72.4 | 77 |
| March | 9.5 | 33.6 | 74 |
| April | 11.8 | 100.9 | 84 |
| May | 16.2 | 57.4 | 84 |
| June | 23.6 | 18.1 | 75 |
| July | 23.8 | 61.4 | 84 |
| August | 26.5 | 1.9 | 80 |
| September | 25.2 | 141.0 | 77 |
| October | 19.4 | 78.9 | 75 |
| November | 15.6 | 129.5 | 78 |
| Mean | 15.2 | 875.6 | 79 |

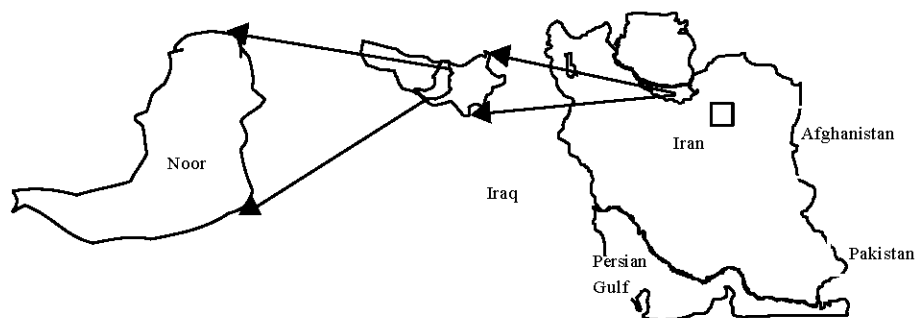


Fig. 1: Position of study area in North of Iran

and large (4 ± 0.3 mm) size in collar diameter (CD) were selected. The height of seedlings was averaged 28.9 (± 5.5) cm. Seedlings were sown in (i) full blackberry cover (30-40 cm in height) and (ii) removed blackberry cover (in radius 50 cm around the seedling). The experiment was set up as factorial with completely randomized block design in three replications. Totally 12 plots 2×2 m were set up for this purpose. Some nine one- year old seedlings were planted with a regular distribution in each plot. Seedlings were produced in a neighboring public nursery from locally collected acorns grown on equal proportions of peat, coco-peat and mineral soil. We determined seedling morphology and nutrient (N, Mg, Ca and P) content of leaf in the end of the first growing season to evaluate seedling response to the experimental treatments. Sampled seedling were reordered for leaf number, growth flush number apical shoot growth, leaf area and leaf dry weight. Leaf area was registered by measuring a Leaf Area Meter. Leaf samples were dried to a constant weight in an oven at 70°C and ground and powdered in laboratory to determine nutrient elements. Data were tested with Kolmogorov-Smirnov for normality of data. The resulting data were subjected to ANOVA. Means were compared for significant differences using Multivariate Analysis of Variance and Duncan's multiple range tests.

RESULTS AND DISCUSSION

The Multivariate Analysis of Variance revealed that leaf number was affected by collar diameter (CD) and interaction $\text{CD} \times \text{Blackberry}$. Leaf dry weight was not influenced by any treatment but leaf area, growth flush number and apical shoot growth was affected only by blackberry (Table 2).

The mean values of seedling morphology as affected by treatments are given in Table 3. In reality, contrary to leaf dry matter, leaf number and leaf area differed among the treatments. Both were greatest with large CD seedlings grown in moved *Rubus* area. Apical shoot growth was different among treatments; it was greatest in full *Rubus* area where large CD seedlings growing in. Growth flush number was greatest in moved *Rubus* area with small CD seedlings.

The results also indicated that N concentration was affected by blackberry and interaction $\text{CD} \times \text{blackberry}$. No treatment influenced on Ca concentration but blackberry did on P concentration. Both CD and blackberry and also their interaction affect Mg concentration (Table 4). Concentration of P, N and Mg were greatest in moved *Rubus* area with small CD seedlings (Table 5).

The results of present study show that introduction of *Q. castaneifolia* seedlings in dense *Rubus* cover can be favored by clearing. Seedlings with large collar diameter could have higher volumes exhibiting higher nutrient concentrations. Thus, probably increased total biomass in seedling large collar diameter may have resulted in greater nutrient use, supply, uptake and storage.

Table 2: Multivariate analysis of variance for seedling morphology

| Source | Collar diameter (CD) | | | Blackberry cover | | | CD \times Blackberry cover | | |
|---------------------|----------------------|---------|---------|------------------|---------|---------|------------------------------|---------|---------|
| | df | F-value | p-value | df | F-value | p-value | df | F-value | p-value |
| Leaf No. | 1 | 10.222 | 0.002 | 2 | 2.638 | 0.076 | 2 | 4.947 | 0.028 |
| Leaf dry weight | 1 | 2.059 | 0.182 | 2 | 1.330 | 0.308 | 2 | 1.119 | 0.315 |
| Leaf area | 1 | 4.122 | 0.049 | 2 | 3.654 | 0.035 | 2 | 0.799 | 0.377 |
| Growth flush No. | 1 | 0.777 | 0.462 | 2 | 17.118 | 0.000 | 2 | 0.657 | 0.419 |
| Apical shoot growth | 1 | 1.209 | 0.303 | 2 | 7.513 | 0.007 | 2 | 0.279 | 0.598 |

Table 3: Characteristics of leaf morphology in areas moved blackberry and control (full blackberry)

| Morphology characteristics | Moved | Moved | Full | Full |
|------------------------------|---------------------|---------------------|---------------------|---------------------|
| | blackberry-large CD | blackberry-small CD | blackberry-large CD | blackberry-small CD |
| Leaf number | 13.44 \pm 0.95a | 8.95 \pm 1.98c | 10.03 \pm 0.81ab | 9.24 \pm 0.62c |
| Leaf dry weight (g) | 0.19 \pm 0.07 | 0.40 \pm 0.18 | 0.30 \pm 0.14 | 0.32 \pm 0.14 |
| Leaf area (cm ²) | 19.66 \pm 3.5a | 11.44 \pm 1.20ab | 17.66 \pm 1.30ab | 14.44 \pm 3.39ab |
| Growth flush No. | 1.52 \pm 0.90b | 2.05 \pm 0.23a | 1.30 \pm 0.54 b | 2.06 \pm 0.94ab |
| Apical shoot growth (cm) | 8.39 \pm 1.49ab | 5.54 \pm 0.68b | 11.07 \pm 1.28a | 6.86 \pm 1.33b |

CD: Collar diameter, Means followed by different letter(s) in row are significantly different

Table 4: Multivariate analysis of variance for nutrient of leaf

| Source | Collar diameter (CD) | | | Blackberry cover | | | CD × Blackberry cover | | |
|--------|----------------------|---------|---------|------------------|---------|---------|-----------------------|---------|---------|
| | df | F-value | p-value | df | F-value | p-value | df | F-value | p-value |
| N | 1 | 4.810 | 0.053 | 2 | 8.926 | 0.006 | 2 | 5.627 | 0.039 |
| P | 1 | 1.121 | 0.315 | 2 | 46.355 | 0.000 | 2 | 1.121 | 0.315 |
| Ca | 1 | 0.665 | 0.434 | 2 | 2.091 | 0.174 | 2 | 0.635 | 0.444 |
| Mg | 1 | 184.305 | 0.000 | 2 | 9.921 | 0.004 | 2 | 98.537 | 0.000 |

Table 5: Characteristics of leaf nutrient in areas moved blackberry and control (full blackberry)

| Nutrient characteristics | Moved | Moved | Full | Full |
|--------------------------|---------------------|---------------------|---------------------|---------------------|
| | blackberry-large CD | blackberry-small CD | blackberry-large CD | blackberry-small CD |
| N (ppm) | 1.82±0.03b | 2.16±0.22a | 2.06±0.12ab | 2.04±0.14ab |
| P (ppm) | 1320.00±52b | 1400.00±60a | 1200.00±50c | 1200.00±60c |
| Ca (ppm) | 0.40±0.50 | 0.10±0.00 | 0.10±0.00 | 0.10±0.00 |
| Mg (ppm) | 5.60±0.25d | 9.60±0.36a | 6.60±0.30c | 7.20±25b |

CD: Collar diameter, Means followed by different letter(s) in row are significantly different

Annuals grow rapidly in the winter and early spring, depleting surface soil layers of water quickly; these annuals flower and complete their life cycle by mid-summer. In contrast, native perennials have slower growth rates and consume soil water at a slower rate so that the plants maintain photosynthesis well into the summer months while water is still available (Holmes and Rice, 1996). Increased content p in seedling leaf with and clearing blackberry probably implies decreased water available for *Quercus* seedling. Present results show that content N (nitrogen) and Mg (magnesium) in moved blackberry area can control aboveground competition, decrease below ground competition, improve seedling performance while avoiding the intense disturbance associated with blackberry removal.

Shading particularly resulted from crown cover has proven beneficial for the establishment of several *Quercus* species (Rey-Benayas, 1998; Gómez-Aparicio *et al.*, 2005; Puerta-Piñero *et al.*, 2007). However, drought under shade may be more stressful than in open areas (Valladares and Percy, 2002; Valladares *et al.*, 2004). Experiments with shading seedling *Q. castaneifolia* suggest that shade does not improve the short-term survival of *Q. castaneifolia* seedlings under gentle conditions (Mirzaei *et al.*, 2007). Thus, it can be stated that in our experiment, shade probably played a minor or even negative role, in the balance between positive and negative interactions between *Q. castaneifolia* seedlings and neighboring vegetation in the undisturbed blackberry cover. Nitrogen hardening reduced shoot size, root collar diameter, leaf area, specific leaf area and root growth potential. Generally, short-term field survival is highly dependent on the nutritional conditions (Trubat *et al.*, 2008).

CONCLUSION

This research suggests that blackberry has a net facilitative effect on the early stages of tree seedling establishment compared with areas of moved blackberry and the most suitable conditions for growth and establishment of *Quercus* seedlings is removal of black berry (Holl, 2002). Seedlings with large collar diameter create growing attributes. Likewise, the seedlings growing in moved *Rubus* area produce greater plant biomass and mineral nutrient matter. From this research it can be concluded that for plantation of *Quercus castaneifolia* seedling, using large collar diameter seedlings and for seedling establishment, cleaning herbaceous vegetation around the seedling, can be a best technique.

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