EFFECTS OF ELEVATED CO_2 AND SHADE ON THE DECOMPOSITION OF SENESCED TREE FOLIAGE: IMPACTS ON THE GROWTH AND SURVIVAL OF TREEHOLE MOSQUITOES

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Abstract: We tested the hypothesis that growth, survival, and reproductive capacity of treehole mosquitoes can be affected by alterations of forest sunlight and CO_2 levels. Larval *Aedes triseriatus* were fed naturally senesced, abscised foliage from red oak (*Quercus rubra*) and paper birch (*Betula papyrifera*) seedlings grown in ambient and elevated CO_2 atmospheres. Oak seedlings were grown in full sunlight. Birch were grown in full sun and partial shade. Females fed birch leaves grown in elevated CO_2 were larger than those fed birch grown in ambient CO_2. However, fewer females emerged from microcosms containing birch foliage grown in elevated CO_2, which significantly lowered estimates of microcosm total egg production. Elevated CO_2 did not affect the performance of mosquitoes reared on leaves of either species grown in full sunlight. Shading birch foliage led to the production of more mosquitoes of both sexes. Males fed shaded foliage were larger and survivorship was higher for males and females, which led to a significantly higher estimate of microcosm egg potential. Birch diets produced larger females and a larger quantity of smaller males than did diets of oak. Mosquitoes of both sexes took significantly longer to develop on birch. Our results indicate that *A. triseriatus* larvae are more sensitive to forest light intensity and tree species composition than they would be to a doubling of atmospheric CO_2 elevation.

INTRODUCTION

Aedes triseriatus is a common inhabitant of water-filled treeholes in eastern North America wherein larvae graze on allochthonous foliage and the microbes that decompose it. In the Great Lakes region, the *A. triseriatus* life cycle begins in early spring as eggs break winter diapause to hatch. Mosquitoes that complete development pass through four larval instars and a pupal stage prior to adult emergence. Development from egg to adult typically requires two weeks to two months depending on growing conditions. Adult females live only long enough to mate, obtain a blood meal, and oviposit in cavities. Males are similarly ephemeral. Cohorts are univoltine in the northern part of the range and bivoltine elsewhere when habitat requirements are met (Walker et al. 1991).

Water-filled treeholes are distinct ecosystems fueled almost exclusively by organic carbon derived from allochthonous foliage and inorganic nutrients that enter the system via stemflow (Carpenter 1983, Walker et al. 1991). Treehole communities are predominantly comprised of decomposer microbes and larval insects that consume decomposing foliage. Like other aquatic insect detritivores (Cummins and Klug 1979), *A. triseriatus* larvae are known to be sensitive to quantitative and qualitative changes in microbially conditioned, senesced-leaf diets

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(Carpenter 1983, Walker et al. 1991). Therefore, alteration of any environmental variable that affects the quantity or quality of foliage inputs could affect all biological processes occurring in a treehole.

Qualitative changes in leaf chemical composition and quantitative changes in total leaf production can result from forest CO_2 and sunlight alteration (Kubiske and Pregitzer 1995). Such changes in leaf chemistry can affect the performance of decomposer microbes (Kaufman et al. this issue) and the insects that feed on leaves (Herms et al. this issue). In this study, we examine the effects of forest sunlight and CO_2 alteration on the development time, adult mass, survivorship, and reproductive capacity of a detritivorous insect common to the Great Lakes region.

MATERIALS AND METHODS

Leaves

Senesced leaves were collected following autumn abscission from one-year-old paper birch and red oak seedlings grown in 350 ppm and 750 ppm CO_2 atmospheres in full sunlight and from birch seedlings grown in both CO_2 levels under a simulated forest canopy (26 percent full sunlight) (see Kubiske and Pregitzer 1995).

Leaf material was sufficient to establish 42 microcosms: eight oak-sun-ambient CO_2, five oak-sun-elevated CO_2, nine birch-sun-ambient CO_2, eight birch-sun-elevated CO_2, seven birch-shade-ambient CO_2, and five birch-shade-elevated CO_2 microcosms.

Mosquitoes

Eggs were collected from automobile tire oviposition traps that were placed in woodlots on the Michigan State University campus (47°N, 84° W, 244-274 m altitude). Eggs were hatched in deoxygenated water and immediately added to microcosms. Each 500 ml plastic microcosm contained 20 larvae and 1 g of leaves conditioned for seven days in 300 ml of deionized water and a 5 ml inoculum of microbes in an inorganic ion solution derived from field-collected stemflow (see Kaufman et al. this issue). The experiment was conducted in a walk-in growth chamber. Temperature was held constant at 23 °C. Microcosms were shaded. Upon emergence, adults were aspirated from microcosms and immediately frozen at -20 °C. At the end of the experiment, they were dried for 12 hours at 65 °C and weighed on a Cahn model 27 microbalance.

Fecundity was estimated with Livdahl's (1982) regression equation for *A. triseriatus* egg production potential (fecundity = 7.13 + 45.85 mg dry mass). Total microcosm egg production was calculated by summing fecundity estimates for each surviving female, thereby providing an estimate of the potential of treatment effects to influence *A. triseriatus* population dynamics.

Statistical Analysis

Effects of the six treatment combinations on development time, adult mass, and estimated fecundity were determined with the use of two ANOVA models. The first tested the effects of CO_2, sunlight, and their interaction on the development time, adult mass, and estimated egg production potential of mosquitoes reared on birch leaves. The second model tested the effect of CO_2, tree species, and their interaction on the development time, adult mass, and estimated potential egg production of mosquitoes reared on oak and birch grown in full sunlight. All data are reported as untransformed means except microcosm fecundity estimates which were square-root transformed to reduce variance heteroscedasticy.

RESULTS AND DISCUSSION

Effects of Elevated CO_2

Females fed birch leaves grown in elevated CO_2 were larger, and therefore more fecund, than those fed birch grown in ambient CO_2 (Table 1, Figure 1). This positive response by a detritivore to elevated CO_2 is in direct contrast to previously observed and predicted effects of CO_2 elevation on foliage decomposition (Lambers 1993, Cotrufo et al 1994).

Elevated CO_2 is predicted to decrease decomposition rate because it increases leaf C:N ratios and the concentration of lignin and other decomposition-inhibiting secondary metabolites (Lambers 1993). However, it is generally thought that the key component of the *A. triseriatus* larval diet is not leaves, but the microorganisms that decompose leaves (Walker and Merritt 1988, Walker 1991, Kaufman et al. this issue). Kaufman et al. (this issue) observed that elevated CO_2 slows decomposition by microbes. Therefore, females, which take longer to develop than males, may achieve greater mass due to increased substrate permanence as would result from delayed nutrient availability for microbes.

Table 1. F values from ANOVA of mosquito performance on senesced birch leaves exposed to two levels of CO_2 and sunlight (* denotes $P \leq 0.10$, ** ≤ 0.05, *** ≤ 0.01).

	SOURCE		
	CO_2	Light	CO_2 * Light
females per microcosm	6.9 ***	4.2 **	1.7
males per microcosm	0.1	6.5 ***	0.2
female development time	0.3	17.9 ***	2.2
male development time	0.7	94.4 ***	0.0
female mass	3.3 *	2.6	1.6
male mass	0.1	43.5 ***	1.5
microcosm fecundity	3.4 *	2.9 *	0.3

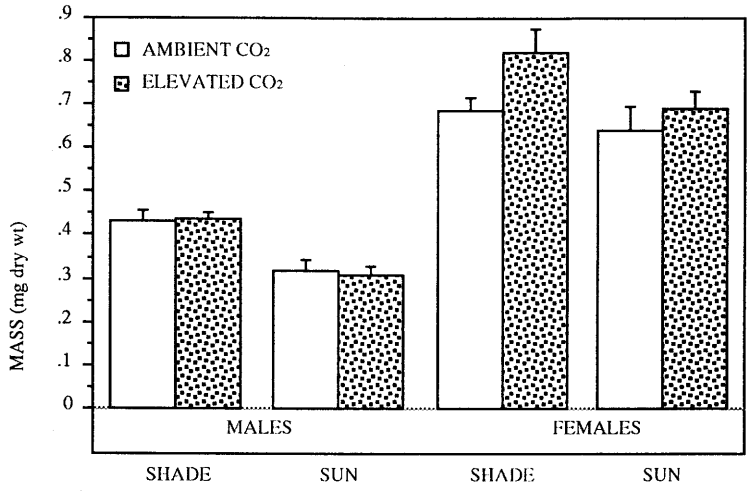

Figure 1. Effects of sunlight intensity and CO_2 level on *A. triseriatus* mass (± 1 S.E.).

Females that survived to adulthood on diets of elevated CO_2-treated leaves were more fecund, but fewer survived per microcosm, which led to lower estimates of potential microcosm egg production (Figures 2 and 3). Therefore, we observed no potential positive population-level responses to elevated CO_2.

Male development time and adult mass were unaffected by CO_2-elevation effects on birch leaves grown at either light level. Elevated CO_2 had no discernible effect on the performance of males or females reared on leaves of either species grown in full sunlight (Figures 6-10).

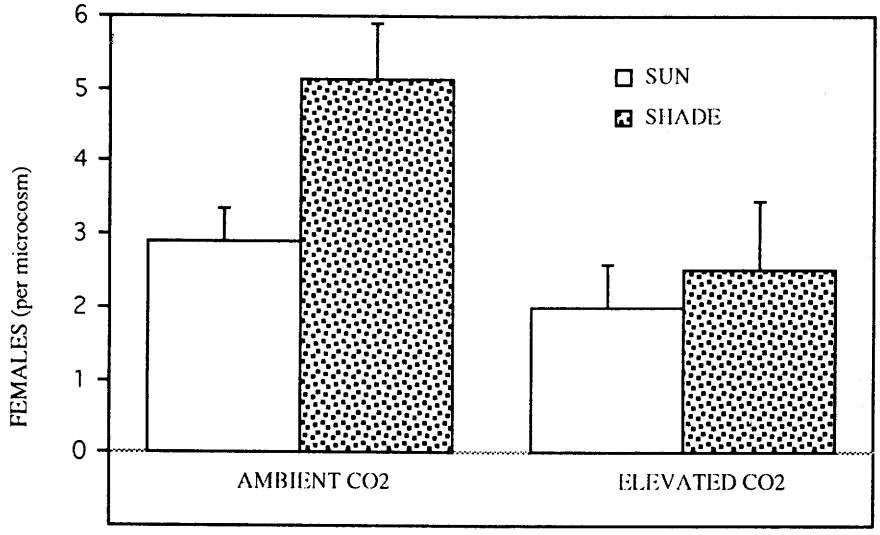

Figure 2. Effects of sunlight intensity and CO_2 level on *A. triseriatus* female survival (± 1 S.E.).

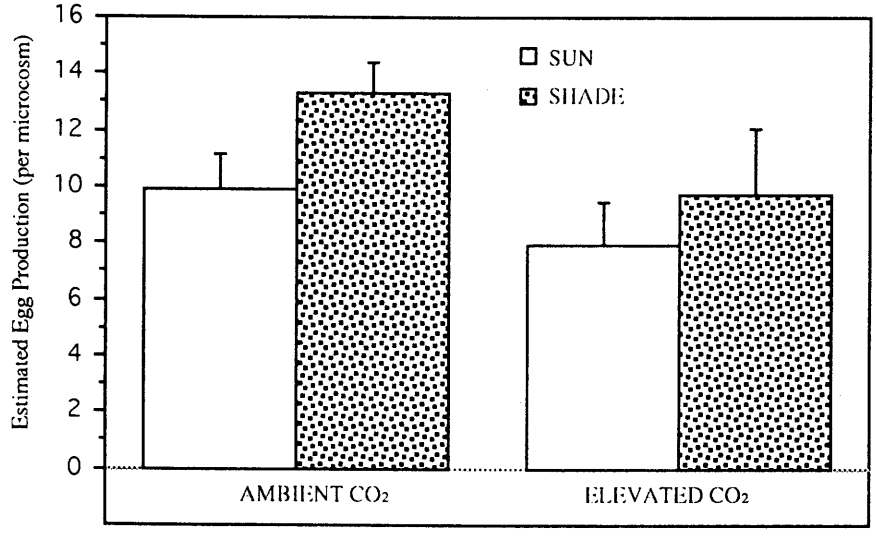

Figure 3. Effects of sunlight intensity and CO_2 level on *A. triseriatus* fecundity (square root mean ± 1 S.E.).

Effects of Shade

Mosquito performance was greatly enhanced by the effects of shade on birch (Table 1). Shading led to the production of a larger number of more-rapidly developing mosquitoes of both sexes (Figures 2, 4, and 5). Also, microcosms containing shaded foliage had a higher potential egg production than did those containing foliage grown in full sunlight. This was attributable to enhanced female survival (Figure 3).

Like the effects of elevated CO_2 on mosquito performance, those of shading may be caused by alterations of leaf chemistry and structural integrity. Typical plant responses to shade include decreased C:N ratios and lowered concentrations of carbon-based allelochemicals such as lignin, tannins, and phenolics (Herms and Mattson 1992). These reductions would be expected to increase decomposition by microbes, as observed by Kaufman et al. (this issue). Decomposition of shaded foliage may, therefore, progress too rapidly to allow for the complete development of some female mosquitoes. Males were able to gain more mass on leaves grown in shade (Figure 1), presumably due to their relatively rapid development time (Figure 5).

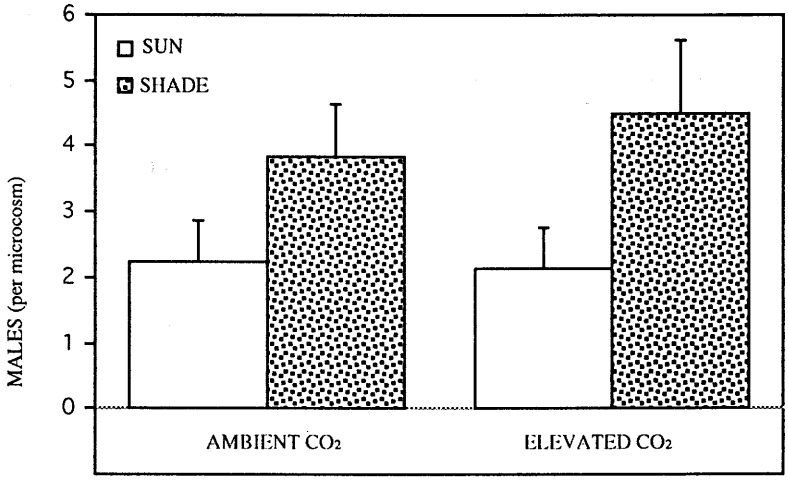

Figure 4. Effects of sunlight intensity and CO_2 level on *A. triseriatus* male survival (± 1 S.E.)

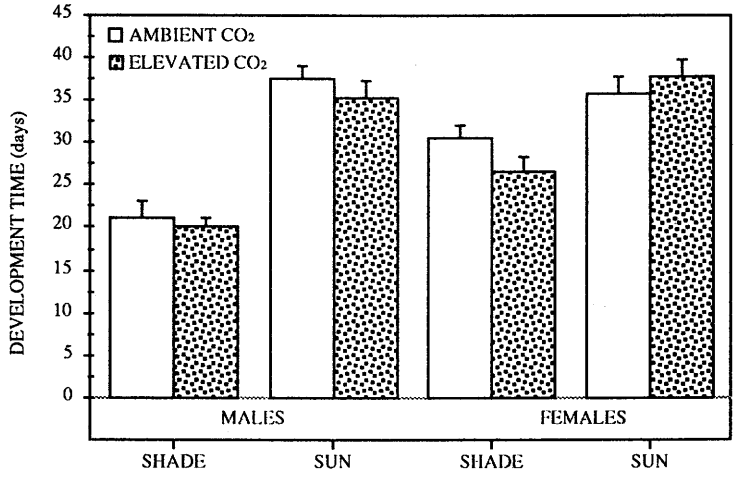

Figure 5. Effects of sunlight intensity and CO_2 level on *A. triseriatus* development time (± 1 S.E.).

Birch responses to shade apparently caused the observed effects on mosquito performance. Light intensity reduction can affect tree species differentially, which could alter forest vegetation composition. Therefore, changes in light intensity may be predicted to generate effects that cascade through an ecosystem from producers to detritivores.

Effects of Tree Species

Females fed birch leaves were larger than those fed oak, but they required much more time to complete development (Table 2, Figures 6 and 7). Males fed birch also took longer to develop and they were more numerous, but smaller than those fed oak (Table 2, Figures 8, 6, and 7). Any positive population-level effects caused by increased female mass and male survivorship would likely be counteracted in natural systems by the increased rates of death by predation, disease, and habitat drying that would result from prolonged development time.

Females reared on birch were larger, and therefore more fecund, than those fed oak. However, this increase did not lead to differential estimated total egg production per microcosm, due, in part, to slightly lower female survivorship on birch relative to oak (Table 2, Figures 9 and 10).

Table 2. F values from ANOVA of mosquito performance on senesced oak and birch leaves exposed to two levels of CO_2 (* denotes P ≤ 0.10, ** ≤ 0.05, *** ≤ 0.01).

	SOURCE		
	CO_2	Tree Species	CO_2*Species
females per microcosm	0.8	0.3	0.2
males per microcosm	0.6	10.4 ***	0.7
female development time	0.0	65.4 ***	0.7
male development time	1.9	294.9 ***	0.0
female mass	0.2	11.0 ***	0.9
male mass	0.1	2.9 ***	1.9
microcosm fecundity	0.1	0.1	1.2

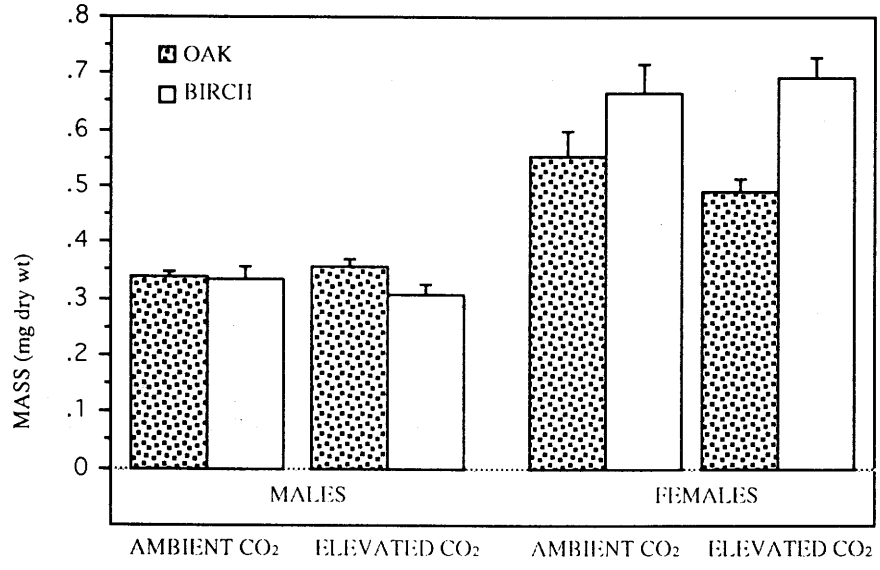

Figure 6. Effects of tree species and two levels CO2 on *A. triseriatus* mass (± 1 S.E.).

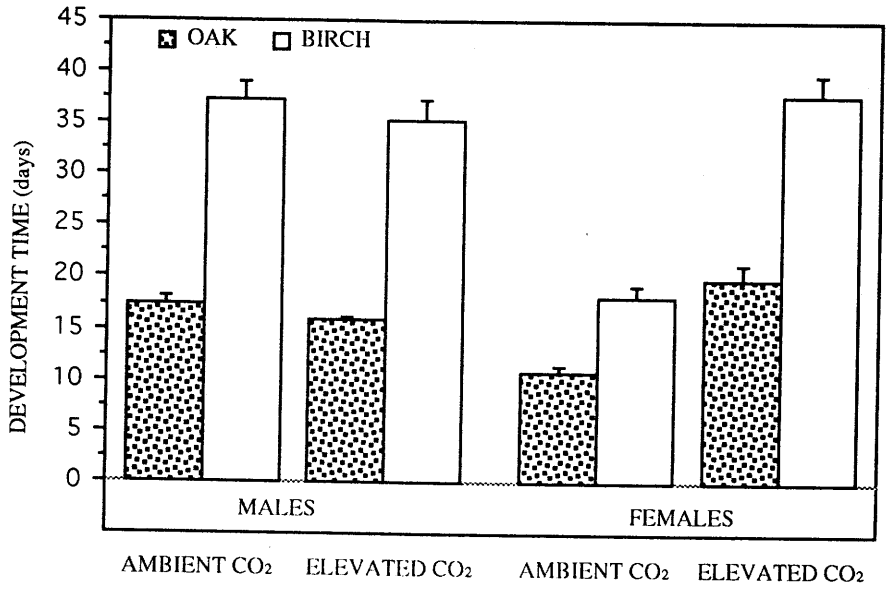

Figure 7. Effects of tree species and two levels CO_2 on *A. triseriatus* development time (± 1 S.E.).

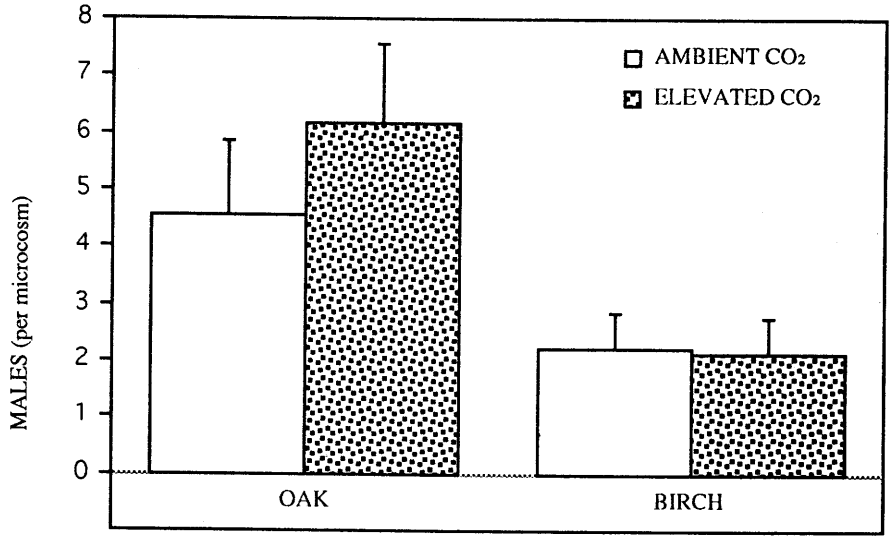

Figure 8. Effects of tree species and two levels CO_2 on *A. triseriatus* male survival (± 1 S.E.).

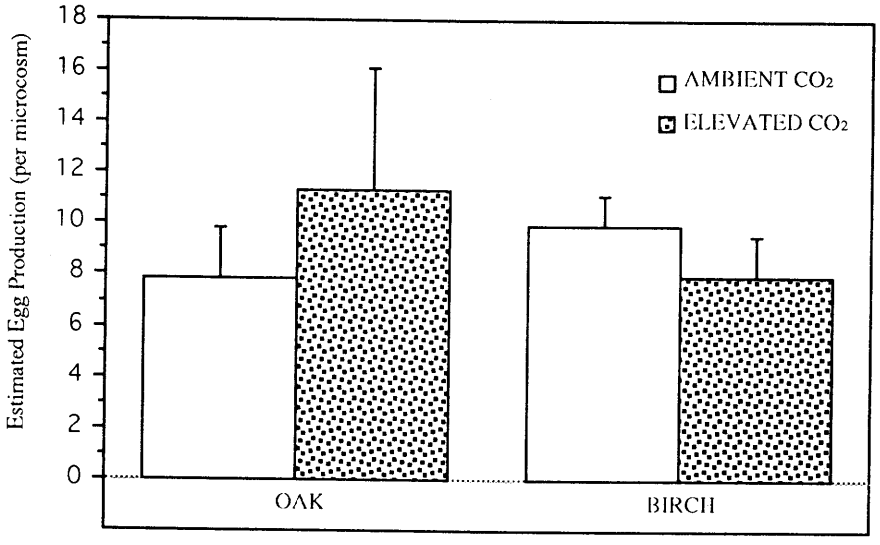

Figure 9. Effects of tree species and two levels CO_2 on *A. triseriatus* fecundity (square root mean ± 1 S.E.).

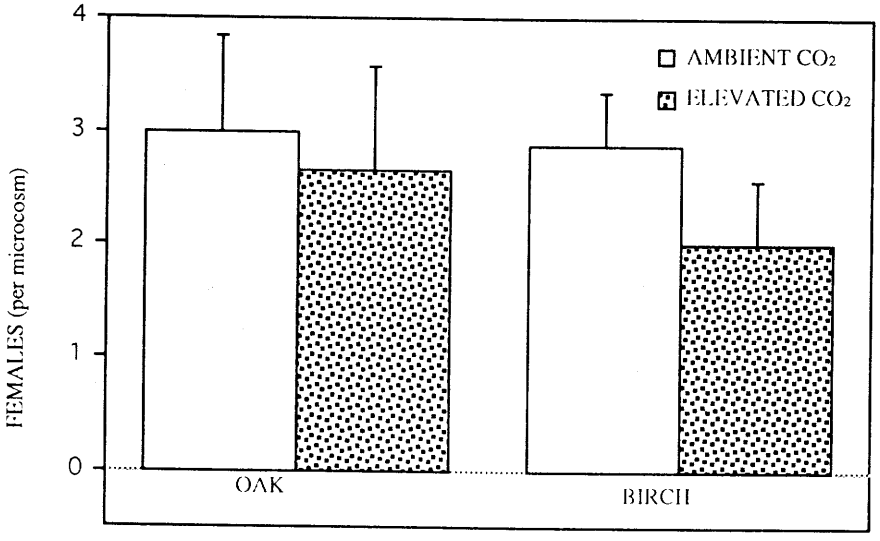

Figure 10. Effects of tree species and two levels CO_2 on *A. triseriatus* female survival (± 1 S.E.).

The relative dominance of any tree species is determined by a variety of abiotic conditions including ambient light and CO_2 levels (Herms and Mattson 1992). Trees respond differentially, both inter- and intraspecifically, to changes in these parameters (Kubiske and Pregitzer 1995). Therefore, ambient light intensity and CO_2 level could independently and in concert, shape forest vegetation composition. Our results suggest that such an effect would ultimately affect detritivorous insect populations.

SUMMARY

Differences in light intensity and atmospheric CO_2 level can generate effects that cascade through an ecosystem from producer to detritivore. The effects of elevated CO_2 observed here (increased female mass and decreased female survival and potential egg production per microcosm) were generally small compared to those of sunlight (increased male and female survival and estimated potential egg production per microcosm and decreased male and female development time) and tree species (increased size and development time of females fed birch, prolonged development time and decreased mass of males fed birch, and decreased estimated egg production for microcosms containing birch). These results suggest that elevated CO_2 impacts on *A. triseriatus* may be difficult to detect against the backdrop of environmental variation already experienced by this insect in the forests of the Great Lakes region.

Further study is required to establish whether or not the patterns observed here for treehole mosquitoes also apply to other detritivores in heterotrophic aquatic systems or elsewhere. However, the reliance of detritivores on abscised foliage does lend support to the supposition that change of any environmental variable that alters leaves qualitatively, will likely impact ecosystem detritivory processes.

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