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BIGGER IS NOT ALWAYS BETTER: CONFLICTING SELECTIVE PRESSURES ON SEED SIZE IN *QUERCUS ILEX*

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Abstract.—Most theoretical treatments of the evolutionary ecology of offspring size assume a simple and direct effect of investment per offspring on offspring fitness. In this paper I experimentally determine the relationship between seed mass and several main fitness components of the oak *Quercus ilex*, to estimate phenotypic selection acting on seed mass during the early life cycle and to discover any potential selective conflicts occurring between different stages from dispersal to establishment. I found a positive effect of acorn size on most fitness components related to seedling establishment. Large size increased germination rate and seedling survival, accelerated germination timing, and enhanced seedling growth. Nevertheless, there was also a direct negative effect of acorn size on survival to predation, because large acorns were highly preferred by the main postdispersal seed predators at the study site, wild boars and wood mice. Because of the low probability of escape from predation, the fitness of large acorns estimated on this component was significantly lower than the fitness of smaller acorns. Therefore, seed size affected fitness in two different ways, yielding opposing and conflicting selective forces. These findings suggest that the general assumption that offspring fitness is a fixed positive function of seed size needs to be reconsidered for some systems. The existence of conflicting selection might explain the occurrence of an optimal seed size in some plant species without invoking a seed number-size trade-off.

Key words.—Evolutionary conflicts, logistic selection gradients, offspring size, seed-mass evolution, seed predators, selection episodes.

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The study of the evolution of propagule size has engaged the attention of many evolutionary biologists (Lack 1954; Williams 1966; Clutton-Brock 1991; Roff 1992, 2002; Stearns 1992). In a seminal paper, Smith and Fretwell (1974) proposed a model that predicted the evolution of an optimal propagule size that maximizes the fitness of the mother. This model was based on two main assumptions: (1) the existence of a trade-off between offspring number and size; and (2) the occurrence of a simple and direct effect of investment per offspring and offspring quality, which results in a positive association between offspring mass and fitness (Smith and Fretwell 1974). Most theoretical treatments of the evolutionary ecology of offspring size developed later have included these preconditions (Lloyd 1987; Clutton-Brock 1991; Rees 1997; Rees and Westoby 1997; Sakai and Harada 2001; Roff 2002 and references therein). For plants, the dependence of offspring fitness on seed size has been extensively supported by the empirical evidence; many studies have found that, for both intra- and interspecific comparisons, larger seeds benefit seedlings against competition (Rees 1995; Jakobsson and Eriksson 2000), herbivory (Bonfil 1998), shading (Hewitt 1998; Bond et al. 1999), drought (Leishman and Westoby 1994; Seiwa et al. 2002), and nutrient limitation (Jurado and Westoby 1992; Seiwa 2000). Consequently, directional phenotypic selection on seed mass acting through offspring fitness is now widely accepted (Rees 1997; Westoby et al. 1997; Sakai et al. 1998; Leishman et al. 2000; Messina and Fox 2001 and references therein).

An accurate estimation of the fitness of an organism requires the consideration of multiple stages of the life cycle of the individuals (Campbell 1991; Charlesworth 1994; van Tienderen 1995, 2000; Kozlowski 1999). Plant life cycles comprise several to many sequential stages connected by many transitional processes such as dispersal, germination, survival, and fecundity, each process constituting a fitness component (van Tienderen 2000; Crone 2001). However, phenotypic selection on a continuous character, expressed as a significant covariance between that phenotypic trait and the individual fitness, has often been estimated exclusively for one component (Bell 1997; Fairbairn and Reeve 2001). This approach is appropriate only when the covariance between the focal trait and the fitness does not vary among fitness components and persists throughout the entire life cycle (Arnold and Wade 1984a,b; Wade and Kalisz 1989; Campbell 1991; Fairbairn and Reeve 2001). Unfortunately, for traits affecting multiple components, the fulfillment of this prerequisite is not always possible. First, trade-offs between fitness components are ubiquitous in most life cycles (Stearns 1992; Roff 2002). Second, a phenotypic trait can covary in a different way with different fitness components (Preziosi and Fairbairn 2000; Roff 2002). All of this would yield conflicting selection pressures, in which the effect of a given trait on a fitness component can be altered or even nullified by an opposite effect on another fitness component, resulting in an unexpected net relationship between the trait and the overall fitness (van Tienderen 1995; Preziosi and Fairbairn 2000).

Growing evidence is demonstrating that seed size in many plant species can affect other fitness components in addition to seedling establishment. For example, size can alter the probability of seeds escaping pre- and postdispersal predation (Cipollini and Stiles 1991; Gómez and Zamora 1994; Moegenburg 1996) or being dispersed far from mother plants (Ganeshaiah and Uma Shaanker 1991; Hegde et al. 1991). The relationship appearing between these fitness components and seed size is in many cases negative rather than positive, the smaller seeds being favored against the larger ones (Ganeshaiah and Uma Shaanker 1991; Hegde et al. 1991; Moegenburg 1996). This suggests that the relationship between seed size and fitness seems to be much more complex than previously accepted (Geritz 1995, 1998; Sakai et al. 1998; Eriksson and Jakobsson 1999). Consequently, it is necessary to quantify the phenotypic selection acting on seed size during different fitness components to reach an accurate understanding of the association between seed size and offspring fitness.

The main objective of the present study is to experimentally determine the magnitude and direction of phenotypic selection acting on *Quercus ilex* seed mass for the following fitness components of this plant species: probability of seeds escaping postdispersal seed predation, seed-germination rate, and seedling emergence and survival. The integration of these data will be used to estimate the net effect of seed size on fitness resulting from these components and to test for selective conflicts occurring among different selection episodes from dispersal to establishment.

MATERIALS AND METHODS

Study System

Holm oak, *Quercus ilex* L. (Fagaceae), is a Mediterranean sclerophyllous evergreen tree abundant in the western Mediterranean region. Holm-oak acorns are dispersed during autumn mainly by rodents and European jays, *Garrulus glandarius* (Gómez et al. 2001a; Gómez 2003a). These animals usually transport acorns far from adult oaks and bury them under shrubs or under other tree species (Gómez et al. 2001a; Gómez 2003a). Nondispersed acorns fall to the ground directly beneath adult oaks, remaining in those locations until being removed by rodents or covered by litter.

The study site, located at 1650 m elevation in the Sierra Nevada (southeastern Spain), is a mosaic of patchy oak-pine woodlands coexisting with afforestation stands. Climate in the zone is continental Mediterranean, with cold winters and hot, dry summers. Rainfall is concentrated mainly in autumn and spring, with an annual precipitation of 825 mm (mean for 1990–1998).

Analysis of Phenotypic Variation in Acorn Size

I examined phenotypic variation in acorn size in 1999 and 2000. To do this, I collected 2000 and 1000 acorns each year, respectively, from 50 permanently marked adult oaks at the study site. In the first year, acorns were collected from all marked oaks and pooled before analysis. In the second year, I sought to determine the maternal and offspring (acorn) contribution to size variation, and thus I collected 200 acorns from each of five oaks selected from the adult oak population. The acorns were collected both years when completely ripe but before dispersal (November in both years) and were immediately transported to the laboratory, to be stored at 2°C until weighed with a digital precision balance (± 0.1 mg, Mettler-Toledo, Griefensee, Switzerland).

The between-tree and within-tree contribution to acorn size was statistically analyzed using the model II ANOVA in SAS software (Proc VARCOMP, SAS Institute 1997), introducing oak identity as a random effect. I fitted the model using the recommended restricted maximum likelihood (REML), because it generates a more accurate estimate of the variance component of each source of variation than does the standard least-square methodology (Littell et al. 1999).

Estimation of the Sequential Fitness Components

Total fitness W_t (z) of an individual with character z can be estimated as the product of several fitness components corresponding to sequential transition rates among life-cycle stages, as: $W_t(z) = \Pi W_k(z)$ (Koenig and Albano 1987; Wade and Kalisz 1989; Campbell 1991). For *Q. ilex*, fitness during its early life can be expressed as: $W_t(z) = W_p(z) \times W_g(z) \times W_{em}(z) \times W_s(z)$, where $W_t(z)$ is survival from dispersal to the sapling stage, $W_p(z)$ is probability of escape from postdispersal seed predation, $W_g(z)$ is germination probability, $W_{em}(z)$ is seed emergence probability, and $W_s(z)$ is seedling survival during the first years. The last three components can be grouped into a single component $W_e(z)$, establishment probability. Seed predation usually occurs after acorns have dropped from the adult oak or have been cached by jays, between November and January (Gómez 2003b). The main seed predators at the study site are wild boars (*Sus scrofa*) and wood mice (*Apodemus sylvaticus*,) which some years consume up to 100% of the total crop (Gómez et al. 2001a; Gómez 2003b). Although *A. sylvaticus* disperses acorns in other locations, at the study site it seems to behave as seed predator, consuming more than 96% of the transported acorns (J. M. Gómez, unpubli. data). Germination occurs at the study site from January to March and emergence from May to June, and thus acorns can spend a long time germinated under soil without producing aerial biomass. Most seedling death occurs during the summer of the first season (Gómez et al. 2001b). Germination is affected mainly by water content in soil, but also depends on several allelopathic compounds (Gómez 2003b; Puerta Piñero et al. 2003). The main selective agent affecting emergence and seedling survival at the study site is summer drought (Gómez et al. 2001a,b; Gómez 2003b). These three processes are affected by environmental variables such as microhabitat type and seed burial (Gómez 2003b).

Relationship between Acorn Size and Predation

The relationship between acorn size and probability of escaping postdispersal seed predation was determined by sowing 1200 individually weighed (to 0.1 g) acorns in December 1999. Acorns were collected from five oaks located at the study site and stored for several weeks in a cold room at 2-4°C. Nonviable acorns (empty or preyed upon by insects) were detected by floating them in water (Bonfil 1998) and excluded. Acorns were placed at the study site according to a stratified random design to assess the spatial variation in predation probability (Gómez 2003b). Thus, 300 acorns (60 per adult oak) were placed in each of the following microhabitats: (1) Holm-oak, under the canopy of adult and juvenile holm oaks; (2) pines, under the canopy of adult trees belonging to any of the three species of pines growing in the zone (*Pinus sylvestris*, *P. nigra*, and *P. pinaster*); (3) tall shrubs, under the canopy of shrub species $>$ 1.50 m tall, specifically *Prunus ramburii* and *Crataegus granatensis*; and (4) open, which includes both bare soil with small gravel and cobble and with sparse herbaceous cover, mainly annuals and short-lived perennials, as well as deep soil with a thick cover of herbs. These microhabitats are the most abundant at the study site, covering more than 80% of the surface, and are the most frequently used by dispersers for hoarding acorns (Gómez et al. 2001a; Gómez 2003a). Acorns from each adult oak were randomly distributed within each microhabitat, separated from each other by more than 50 cm, and buried horizontally in the soil at 1–3 cm, simulating dispersal by European jays (Bossema 1979; Kollman and Schill 1996; Gómez et al. 2001a; Gómez 2003a).

The experiment was checked after 7, 15, and 90 days, noting the number of acorns depredated in situ, the number removed, and, when possible, the identity of the predators. Wild boars find the acorns by rooting and break them longitudinally. By contrast, wood mice remove the acorn from the soil without any sign of rooting activity, gnaw a hole in the pericarp, and feed on the cotyledons (Gómez et al. 2001a; Gómez 2003b).

Relationship between Acorn Size and Seedling Establishment

The relationship between acorn size and seedling establishment was studied by planting 800 individually weighed (to 0.1 g) fresh acorns in December 1999. Microhabitat was expected to have a large effect on establishment (Gómez 2003b); therefore, I sowed the acorns according to a random stratified design across four microhabitats as described above. Acorns were planted in 30 × 30–cm planting stations (20 per microhabitat), each containing 10 acorns. Each planting station was completely covered by wire cages to exclude predators. Acorns were randomly assigned to each planting station and to the position within each station and were buried in a hole 1–3 cm deep, simulating dispersal by jays (see above). After sowing, the planting stations were visited once monthly for the first six months and once every three months for the next year, noting on each visit the acorns that had germinated, the timing of germination, the probability of seedling emergence, and the survival of the emerged seedlings. Any acorn with the radicle longer than 1 cm was listed as germinated, and any acorn with shoots growing beyond ground surface was listed as emerged. On 20 July 2002, shoot height was measured on all surviving seedlings.

Statistical Analysis of the Effect of Acorn Size on Acorn Fate

All binomial or multinomial dependent variables (predation, germination, emergence, and survival) were analyzed by generalized logit models (Proc CATMOD, SAS Institute 1997) using maximum-likelihood estimations (Stokes et al. 1995). Seedling height was analyzed by general linear model (Proc GLM, SAS Institute 1997). In the predation experiment, the microhabitat and acorn mass were introduced as main factors. In the analysis of the germination experiment, the microhabitat, the acorn mass and the planting plots nested into microhabitat were introduced as main factors. In all models, the individual acorns or seedlings were introduced as the error term. To determine the effect of acorn size on seedling height, while removing the confounding effects of the other factors, I used the partial regression leverage plots of each trait on fitness residuals (Rawling et al. 1998). Leverage plots were made by regressing the residuals of each variable against the residuals obtained from the regression of the dependent variable on all the remaining independent variables excluding the target variable. The slope of the linear regression is the partial regression coefficient for that independent variable in the full model (Rawling et al. 1998). This technique also calculates a confidence function with respect to each variable, from which one can give the sign and percentage of the variability in fitness explained by each variable.

Estimation of Phenotypic Selection

I estimated phenotypic selection on seed size separately for each fitness component considered in this study (Arnold and Wade 1984a,b; Wade and Kalisz 1989; Fairbairn and Reeve 2001). I used the independent method proposed by Koenig and Albano (1987), in which the selection estimates for each fitness component are calculated by the use of only the individuals with nonzero fitness at the start of the life-cycle episode. These independent gradients are insensitive to the ordering of the life-cycle episodes and are not additive (Fairbairn and Reeve 2001).

The fitness components used in this study are all dichotomous, with $W_{ki} = 1$ indicating survival of the ith individual during the kth fitness component and $W_{ki} = 0$ indicating death (W_{ki} represents absolute fitness). Standard least-squares regressions are not appropriate to analyze these types of data (Brodie et al. 1995). Thus, as suggested by Janzen and Stern (1998), I used logistic regressions to estimate the phenotypic selection acting on seed size through each sequential fitness component. According to this methodology, $W_k(z) = \mathrm{P}(W = 1|z)$ denotes the survival probability (or expected fitness) for an individual during the kth component depending on the value of the phenotypic trait z, and it is found as:

$$W_k(z) = \frac{e^{\alpha_0+\alpha_1 z}}{1 + e^{\alpha_0+\alpha_1 z}}, \tag{1}$$

where z is in this case seed mass, α_1 is the logistic regression coefficient and α_0 is the intercept (Janzen and Stern 1998). The likelihood function L_k, assuming a sample of n independent individuals, is used to fit the logistic regression model to data and to infer the regression coefficient α_1 for each fitness component, and it is expressed as:

$$L_k = \prod \left(\frac{e^{\alpha_0+\alpha_1 z_i}}{1 + e^{\alpha_0+\alpha_1 z_i}}\right)^{w_{ki}} \left(\frac{1}{1 + e^{\alpha_0+\alpha_1 z_i}}\right)^{1-w_{ki}}, \tag{2}$$

where W_{ki} is the absolute fitness of the ith individual for the kth fitness component (Allison 1999). I used SAS software, which fits this function with maximum likelihood using a modified Newton-Raphson iterative algorithm with normal approximation (Proc LOGISTIC, SAS Institute 1997).

The logistic regression coefficients cannot be applied to microevolutionary equations (Janzen and Stern 1998). For a numerical approximation of the selection gradient for acorn size on each fitness component, I transformed the logistic regression coefficients using the technique proposed by Janzen and Stern (1998) as:

$$\beta_{\mathrm{avggrad}} = [\mathrm{W}_k z(1 - \mathrm{W}_k z)]_{\mathrm{avg}}\alpha_1. \tag{3}$$

By using equations (1) and (2), I estimated the expected survival probabilities of the acorns with different masses during each fitness component. Afterward, I estimated the fitness functions for each fitness component by fitting a function relating the $W_k(z)$ values to acorn mass. Because it is not possible to quantify all the fitness components to every acorn, I was not able to obtain an overall selection gradient for the entire life-cycle period considered in this study, $W_t(z)$. However, I did estimate the predicted overall probability for every acorn to survive the entire period, $W_t(z)$, by using the equation $W_t(z) = \Pi\ W_k(z)$. This provides a visualization of the fitness function for all the life-cycle period studied here, from dispersal to establishment.

RESULTS

Variation in Acorn Mass

The average (SE) acorn mass was 2.25 (0.02) g (CV = 31.03, N = 2000 acorns, range = 0.75–4.86 g) in 1999 and 2.64 (0.03) (CV = 36.96, N = 1000, range = 0.79–6.95 g) in 2000. Acorn size distribution fitted a log-normal curve both years ($P > 0.05$ in both cases, KSL test), indicating that there were more smaller acorns than larger acorns (Fig. 1). There were among-oak differences in 2000 with respect to acorn size ($F_{4,\,995} = 184.58$, $P = 0.0001$, model II ANOVA, Fig. 1), the oak identity explaining 38.3% of the variability in acorn size and the within-oak factor explaining 61.7% (REML variance component estimates).

Postdispersal Seed Predation

Only 2% of the experimental acorns escaped predators. The factors significantly affecting the probability of postdispersal seed predation were acorn mass and oak identity (Table 1). Acorns surviving to postdispersal predation were significantly smaller (mean ± SE: 1.77 ± 0.11 g) than those that died (2.14 ± 0.02 g, Fig. 2). Wild boars consumed the heaviest acorns (2.51 ± 0.09 g), whereas wood mice consumed the next-heaviest acorns (2.12 ± 0.03 g). There was a marginally significant relationship between average acorn size per maternal oak and predation rate (log[predation] = 1.91 + 0.13 log[acorn mass], $F = 7.57$, $P = 0.07$, $R^2 = 0.71$, df = 4, P-value found by 2000 iterations with bootstrapping).

Seedling establishment

Only 9% of acorns survived to two-year seedlings. The statistical models testing the effect of acorn size on seedling establishment proved significant for all fitness components, explaining between 37% and 50% of the variability of the dependent variables (Table 1). Acorn mass benefited almost every process occurring during seedling establishment (Table 1). Thus, acorn mass was positively related to germination; the mass of germinated acorns was 2.71 (0.05) g, whereas the nongerminated acorns averaged 2.59 (0.04) g. Acorn mass was also a main factor affecting germination timing (Table 1), with large acorns germinating much earlier than the small acorns. In fact, as shown in Figure 2, the acorns germinating during February and March were significantly larger than those germinating during April and May. Seedling survival was again positively related to acorn mass; seedlings surviving the two first years came from a significantly heavier acorn (2.79 ± 0.15 g) than did seedlings that died (2.51 ± 0.17 g). Finally, seedling height growth was affected by acorn mass (Table 1); bigger acorns produced taller seedlings (seedling height growth = 5.33 + 0.82[acorn mass]; $F_{1,\,109} = 11.03$, $P = 0.004$, $R^2 = 0.14$, partial regression leverage plot). The only fitness component not affected by acorn mass was emergence probability (Table 1).

Microhabitat had a significant effect only on germination and seedling survival and growth (Table 1). The nonsignificant interactions between acorn mass and microhabitat occurring for most fitness components (Table 1) suggest that the effect of acorn mass on fitness was consistent across microhabitats. In fact, there was significant interaction between acorn mass and microhabitat only for germination rate (Table 1).

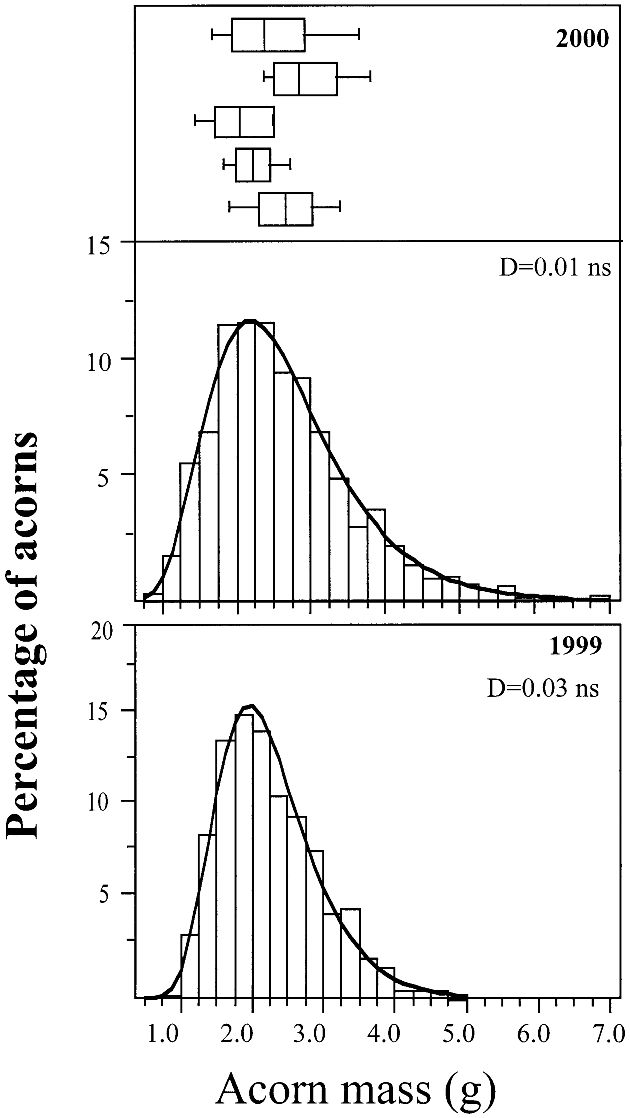

FIG. 1. Frequency distribution of seed mass for the two years of study. The fit to a log-normal curve (KSL test, ns, nonsignificant) is also shown, together with the box-plot of seed mass for each adult oak studied in 2000.

TABLE 1. Analysis of the effects of acorn mass on predation and seedling establishment using generalized logit models (χ^2) or general linear models (*F*). Planting stations were not introduced into the model for seedling height growth because there were many empty stations. Nonsignificant interactions were pooled with the error term.

		Predation rate (N = 1200)		Germination rate (N = 800)		Germination timing (N = 374)		Emergence rate (N = 374)		Seedling survival (N = 111)		Seedling height growth (N = 111)	
	df	χ^2	*P*	χ^2	*P*	χ^2	*P*	χ^2	*P*	χ^2	*P*	*F*	*P*
Microhabitat	3	0.02	0.99	25.59	0.0001	11.80	0.22	4.13	0.25	15.73	0.003	2.71	0.04
Acorn mass	1	5.16	0.02	5.09	0.02	12.74	0.005	0.97	0.32	6.53	0.01	10.33	0.002
Mother/plot[1]	4/1	10.56	0.03	6.97	0.008	5.04	0.17	0.15	0.69	0.01	0.90	0.02	0.87
Planting station (Plot)	18			22.61	0.21	21.02	0.99	17.89	0.46	11.50	0.78		
Acorn mass × microhabitat	3	0.01	0.99	9.90	0.02	5.53	0.79	3.35	0.34	8.43	0.08	1.45	0.23
Overall model		46.33	0.0001	194.38	0.0001	194.38	0.0001	79.18	0.0001	46.78	0.004	3.45	0.005

[1] Mother was introduced in the model testing postdispersal seed predation, and plot was introduced in the remaining ones.

Phenotypic Selection on Acorn Mass during Each Fitness Component

The phenotypic selection action on acorn size was significantly negative when the fitness component considered was the probability of surviving to postdispersal seed predation (Table 2). The logistic coefficient was higher than one, although the average selection gradient decreased due to the extremely low value of $W_p(z)_{\text{avg}}$. The fitness function found for this fitness component suggests that there was intense selection against big acorns, according to the negative exponential relationship between $W_p(z)$ and acorn mass (Fig. 3).

By contrast, there was significant positive selection on acorn size for fitness components related to seedling establishment, such as germination, seedling survival, and the entire establishment probability (Table 2). In this case, larger acorns had a much higher probability of becoming established than did smaller ones. This is reflected in the fitness function for the entire establishment process, the relationship between $W_e(z)$, and acorn mass in this case being a positive exponential (Fig. 3).

As a consequence of the severe selection acting through postdispersal predation, the function relating $W_t(z)$ and acorn size was also a negative exponential (Fig. 3), suggesting that smaller acorns had a higher probability of surviving during their early life cycle than did larger acorns. In fact, the magnitude of the positive selection gradient on seed size through establishment was lower than the magnitude of the negative selection gradient through seed predation. Because that $W_t(z) = W_p(z) \times W_e(z) = 0.0018$, I can estimate a $\beta_{\text{avggrad}} = -0.0019$ for the total life-cycle period considered in this study. The positive effect of size on establishment is apparently cancelled out by the effect that this trait has on survival to seed predation.

DISCUSSION

This study suggests that there is opposing and conflicting selective pressures acting on *Q. ilex* acorn size in the study site. I have found significant positive phenotypic selection on seed size during some part of the life cycle but negative selection during another part.

Acorn size positively affected most fitness components related to seedling establishment. Specifically, large acorns had increased germination rates and seedling survival, accelerated germination timing and enhanced seedling height growth. Oak seedlings receive most of the energy from the acorn in the first growing season (Long and Jones 1996), and therefore it is not surprising a positive effect of acorn size on seedling fitness (Tripathi and Khan 1990; Sonensson 1994; Bonfil 1998). The observed effect of acorn size on seedling survival and growth might be a direct result, because large seeds had more reserves in their cotyledons, but also an indirect result because large acorns germinated earlier, prolonging the growing season and thereby enhancing both below- and aboveground biomass gain, as well as the vertical growth of the resulting seedling (Tripathi and Khan 1990; Ke and Werger Seiwa 1999; 1998, 2000). Timing of germination and emergence plays a critical role in seedling establishment in many plant species, with earlier germinators usually showing an

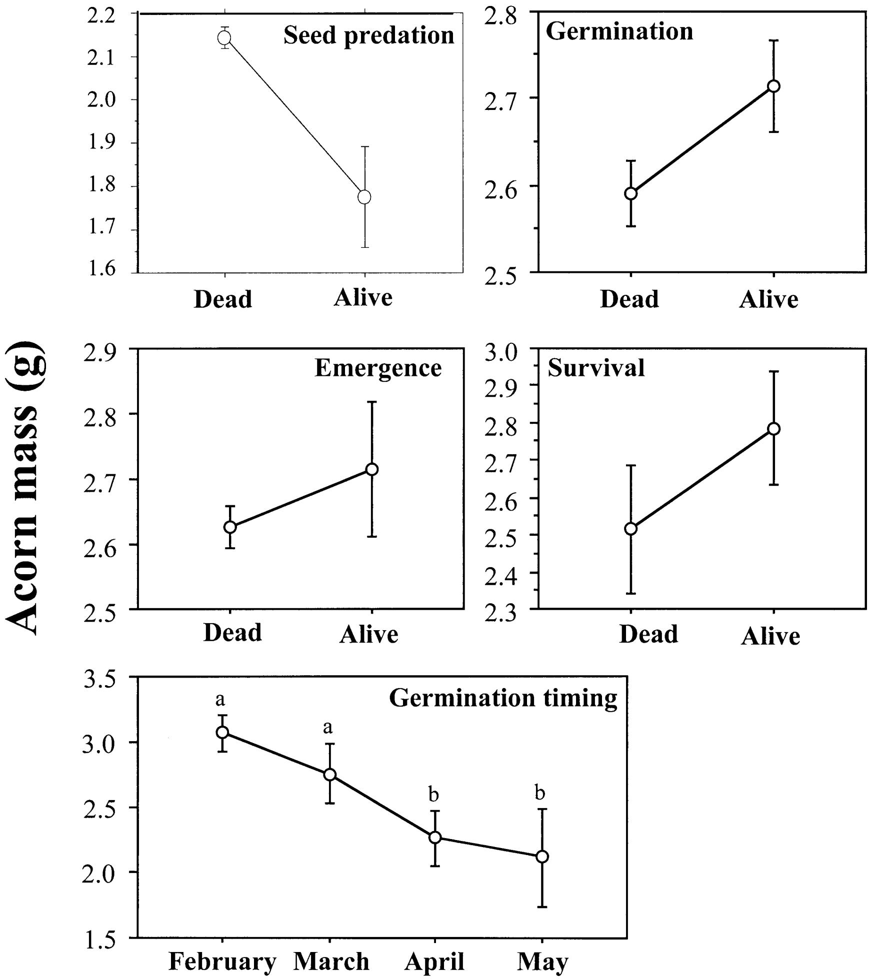

FIG. 2. Difference in seed mass between acorns surviving and not surviving for each fitness component. Means with different letters in the germination timing panel are statistically based on a Bonferroni-Dunn post hoc test.

advantage over later ones (Seiwa Seiwa 1998; and Kikuzawa 1996). The nonsignificant interaction between seed size and microhabitat for any fitness component suggests that the benefits of large seed size for *Q. ilex* recruitment are realized regardless of the environment at the study site. Furthermore, the advantage of acorn size held in sunny (open sites) as well as in shaded sites, despite the fact that *Q. ilex* is a shade-tolerant species that fails to recruit when exposed to full sunlight (Espelta et al. 1995; Retana et al. 1999).

Nevertheless, this study has also shown a direct effect of acorn size on predation probability; large acorns were strongly preferred by the main postdispersal seed predators of Holm-oaks at the study site, the wild boars and the wood mice. Because of the low probability of escaping predation, the fitness of larger acorns estimated on this component was significantly lower than the fitness of smaller acorns (Fig. 3). Holm-oak postdispersal seed predators are extreme generalists, selecting food items based mainly on energy budgets (Hulme and Benkman 2002). Larger seeds are more nutritious, because the proportion of proteins and carbohydrate

TABLE 2. Estimate of phenotypic selection on acorn mass for each fitness component considered in this study. α refers to the logistic regression coefficient, $W(z)_{\mathrm{avg}}$ is the average survival probability of an acorn during each fitness component, and β_{avggrad} is the average selection gradient.

Fitness component	$\alpha \pm 1$ SE	P	$W(z)_{\mathrm{avg}}$	β_{avggrad}	R^2
Escape from postdispersal predation	−1.254 ± 0.563	**	0.0233	−0.029	0.04
Overall establishment[1]	0.228 ± 0.120	*	0.0889	0.018	0.08
Germination	0.135 ± 0.066	**	0.3750	0.032	0.01
Emergence	0.186 ± 0.128	ns	0.3609	0.043	0.01
First-summer survival	0.301 ± 0.213	**	0.6569	0.068	0.01

[1] Overall establishment refers to the probability to survive from germination to two-year seedling stage.

* $P < 0.05$; ** $P < 0.01$; ns, nonsignificant.

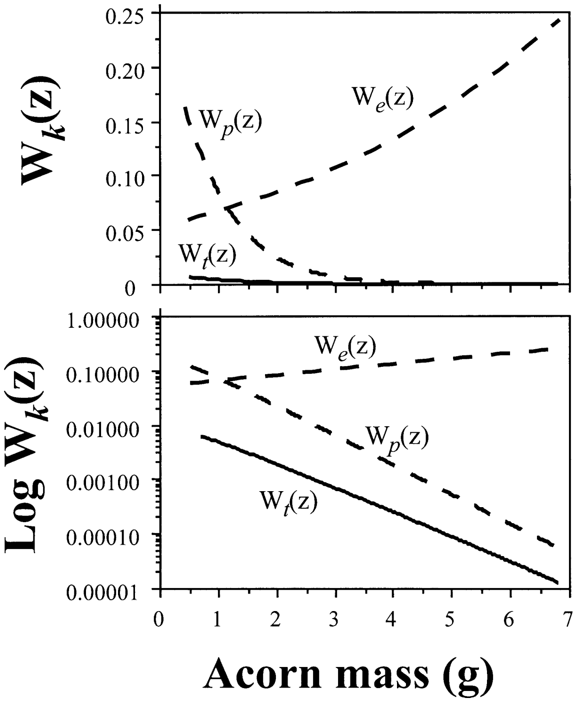

FIG. 3. Predicted fitness functions relating seed mass and fitness (estimate as expected probability of survival) for the main fitness components quantified in this study: escape from predation, $W_p(z)$; establishment, $W_e(z)$; and total fitness from dispersal to sapling, $W_t(z)$. The equations relating fitness and seed mass are: $W_p(z) = 0.254e^{-1.231(\text{acorn mass})}$; $W_e(z) = 0.053e^{0.228(\text{acorn mass})}$; $W_t(z) = 0.010e^{-0.944(\text{acorn mass})}$; $R^2 > 0.95$ in all cases.

reserves increases with greater seed mass in acorns, hence providing more available energy to seed consumers (Tripathi and Khan 1990; Bonfil 1998; Jansen et al. 2002).

The selective role of seed predators on seed size is still controversial (Hulme and Benkman 2002), although several studies have argued that seed consumers can be selective agents because they show a significant preference for seeds of a specific size (Jordano 1984; Cipollini and Stiles 1991; Reader 1993; Gómez and Zamora 1994; Szentesi and Jermy 1995; Moegenburg 1996; Geritz 1998; Jansen et al. 2002). Nevertheless, an increase in maternal fitness requires that this selective predatory feeding behavior can create interplant differences in predation rate and reproductive success (Hulme and Benkman 2002). My limited sample size of plants suggests that oaks producing smaller acorns suffered a lower predation rate during the study year than did oaks bearing larger acorns, according to the significant effect of the mother oak in predation probability (Table 1) and the significant effect of acorn size per oak on predation rate.

As a main consequence of the above-described conflicting selective forces, the net phenotypic selection acting on seed mass was negative (Fig. 3), indicating that selection at the study site is favoring not an increase but a decrease in seed mass. This occurs because to invest more resources in each propagule would result in higher mortality during life-cycle processes other than establishment, because mortality factors such as seed predators select against size. If this study were expanded to incorporate other fitness components of Holm-oaks, such as predispersal seed predation or seed dispersal, more conflicting selection pressures surely would be discovered. For instance, several studies have reported that predispersal seed predators prefer larger seeds (Gómez and Zamora 1994; Moegenburg 1996). *Quercus ilex* acorns are consumed before dispersal at the study site by the weevil *Curculio elephas*. If this weevil species also selects larger seeds, as some unpublished observations suggest (R. Bonal and M. Díaz, pers. comm.), the conflicting selection imposed by postdispersal predators could become even stronger. In addition, the acorns are dispersed at the study site mainly by the jay *G. glandarius* (Gómez 2003a). Bossema (1979) found that these jays also prefer larger acorns of *Quercus robur*. Consequently, it would be very interesting to quantify the relationship appearing between acorns size and dispersal probability or efficiency via jay behavior, to incorporate the potential phenotypic selection provoked by jays into the overall equation and analyze the putative existence of additional selective conflicts.

A potential caveat of this study is related to its short time span in contrast to the long life span of *Q. ilex*. Because selection can fluctuate between years in long-lived organisms, it is difficult to extrapolate the real effect of predators or any other agent on seed size from just a few years. It is thereby imperative to design long-term studies to account for potential variability between different selection events, because the net selection occurring on seed size during every selection event will surely depend on the relative importance of each partial selective pressure. Thus, the advantage of large seeds will be more apparent in years with low predation pressure than during years with high predation pressure. For example, many oaks establish episodically after mast-fruiting events, and it could be possible that during a mast year selection pressure by postdispersal predators relaxes. For this reason, it would be informative to estimate the phenotypic selection exerted by predators in mast and nonmast years. However, it appears that in my study system there is not much temporal fluctuation in predation pressure, because the main predators are wild boars and wood mice, which consume a large proportion of the total seed crop even during years with heavy acorn production (Gómez 2003b). In any case, no theoretical model about propagule size evolution has considered temporal variability in selective regimes (Rees 1997; Rees and Westoby 1997; Sakai and Harada 2001; Roff 2002).

To have any evolutionary significance, there must be heritable variation in acorn size. Significant variation in *Q. ilex* seed size was found in both study years. Furthermore, about 40% of this variation occurred between different oaks, a high proportion even considering the low number of oaks sampled. Intra- and interindividual variation in seed size has been shown for other plant species (e.g., Ganeshaiah and Uma Shaanker 1991; Hegde et al. 1991; Obeso 1993; Moegenburg 1996; Sakai et al. 1997; Turnbull et al. 1999; Simons and Johnston 2000). In some cases, the proportion of total phenotypic variance in seed size attributable to additive genetic variance has been proven negligible, much of the seed-size variation be due to environmental or maternal effects (Wolfe 1995; Leishman et al. 2000; Simons and Johnston 2000; Hulme and Benkman 2002). Nevertheless, high heritabilities in seed size have been reported in many species (Biere 1991;

Lynch and Walsh 1998) and are strongly supported by the long history of artificial selection for this trait in many crops (Lynch and Walsh 1998). As an additional support for the existence of additive genetic variation in seed mass, the quantitative trait loci affecting seed size have being successfully mapped in a diverse array of plant species (e.g., Lynch and Walsh 1998; Alonso-Blanco et al. 1999 and references therein).

The results reported here could have two major implications for studies on seed-size evolution and the theoretical analyses of the size-number trade-off. First, my data suggest that contrasting selective pressures may affect the evolution of seed size. When a phenotypic trait covaries with more than one fitness component, trade-offs due to conflicting selection pressures are possible, and several other biologists have already found conflicting selection regimes on seed size. For example, Moegenburg (1996) proposed that *Sabal palmetto* seed size is the evolutionary outcome of the conflict between invertebrate seed predators and seedling vigor. Furthermore, several authors have suggested that seed size may be an evolutionary response to the opposing pressures exerted by seedling establishment and wind-dispersal efficiency (Hedge et al. 1991; Ganeshaiah and Uma Shaanker 1991; Eriksson and Jakobsson 1999). Conflicting pressures has been detected on other phenotypic traits. For example, evidence is accumulating that several plant traits are the evolutionary outcome of opposing selection exerted by pollinators and herbivores on fecundity (Quesada et al. 1995; Niesenbaum 1996; Brody 1997; Strauss et al. 1999; Gómez and Zamora 2000; Galen and Cuba 2001; Gómez 2003c and references therein). All these findings suggest that intense selection on a trait during a fitness component can be disrupted or cancelled by conflicting factors acting at any other component. For this reason, to gain an accurate view of the selective pressures acting on that organism, it is advisable to consider the entire life cycle of the organisms, as well as the complete ecological scenario in which the interactions take place (Preziosi and Fairbairn 2000).

My results also suggest that the general assumption made by most theoretical models examining the evolution of offspring size—that success of each offspring is a fixed positive function of its size (Smith and Fretwell 1974; Lloyd 1987; Rees 1997; Rees and Westoby 1997; Sakai and Harada 2001)—needs to be reconsidered for some systems. A larger seed is not always beneficial for the fitness of the subsequent seedling, because the seed size in a given environment could be a compromise between increasing seedling performance and increasing escape from predators or improving dispersal efficiency. For example, Eriksson and Jakobsson (1999) predicted that selection should favor higher fecundity and smaller seeds when a seed size–recruitment relationship is absent or negative. Similarly, Geritz (1998) extended the Smith and Fretwell basic model to incorporate the effect of seed predators on seed size using an evolutionarily stable strategy approach, finding that the action of seed predators can sometimes favor high intraplant variability in seed mass. Under these circumstances, an optimal seed size may evolve without an offspring number-size trade-off, caused simply by conflicting selection acting on different fitness components (Roff 2002).

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