In this chapter I discuss the results obtained in chapters 2 to 5 and highlight the most important findings. Throughout this chapter, I point out which further approaches in studying variation in pollen-ovule ratio appear meaningful.

### 6.1 Pollen-ovule ratios and Charnov’s model

Charnov’s (1982) idea that pollen-ovule ratios are a direct consequence of resource allocation to female and male sex function has only been considered further on a theoretical basis in a small number of articles. Queller (1984) pointed out that while most of the theoretical work on sex allocation is grounded in an intraspecific context, pollen-ovule ratios and the mathematical model of Charnov provide an interesting opportunity to put sex allocation in an interspecific context.

Furthermore, Queller showed theoretically that though Charnov’s and Cruden’s views appear to be contradictory at first, they are in fact not mutually exclusive and the findings of Cruden (1977; Cruden and Jensen 1979; Cruden and Miller-Ward 1981) are in line with sex allocation theory.

For some further considerations recall equation 1.3, which can be rewritten as

\[
\log \frac{P}{O} = \log \frac{C_2}{C_1} + \log \left( \frac{r}{1-r} \right) \tag{6.1}
\]

since \( \log C_2 - \log C_1 = \log(C_2/C_1) \). The \( \log(r/1-r) \) term accounts for variation in pollen-ovule ratios caused by different ways of packaging male and female resources within any given \( \log(C_2/C_1) \). In the analyses in chapter 2 and 3 I have assumed that within a given mating system \( \log(r/1-r) \) is constant, based on the notion of Charnov (1982) that within a mating system allocation reflects an evolutionary stable strategy. This assumption seemed to be valid as no significant differences between slopes of the pollen-ovule ratio – seed size relationship or between slopes of the pollen-ovule ratio – pollen size relationship were found.

De Jong and Klinkhammer (2005, p. 137) extended the mathematical model of Charnov by adding a term to account for the fact that in many plants not all ovules develop into a seed. There was not sufficient data on seed/ovule ratios for the species in our analyses to explicitly account for such a possible effect. However, such an effect does not seem to influence our general results. Given that outcrossing species are expected to have smaller seed ovule ratios than selfing species (Wiens 1984), we would expect the slope parameters predicted by Charnov’s model to differ between these groups. As mentioned above, a difference among slopes of different mating systems was not found for the relationship between seed size and pollen-ovule ratio (chapter 2) neither for the relationship between pollen size and pollen-ovule ratio (chapter 3).
There are two further factors that might influence the trait relationships predicted by Charnov’s model that were not studied in detail in this thesis. Firstly, the assumption of sex allocation theory, that male and female investments are drawn from the same limited resource pool may be wrong because investment in maturing seeds can also be assimilated from post-flowering photosynthesis (Seger and Eckhart 1996). We could not account for a possible effect of this phenomenon in our comparative analyses. Second, allocation to male and female function is hypothesized to alter with plant size for several reasons (see review in de Jong and Klinkhammer 2005, chapter 9). In the case of the seed size – pollen-ovule relationship we could not detect an influence of plant size (chapter 2).

Clearly, the relationship between seed size and pollen-ovule ratio and pollen size and pollen-ovule ratio is less clear-cut than would be expected from Charnov’s model. The deviation of the data from the predictions are caused by two major aspects: (1) The assumptions of the model are not met by the actual biological data and (2) the pollen-ovule ratio is part of a complex system of intercorrelated traits which are subject to different selection pressures. I suppose that Charnov’s “mechanistic” model of the allocation to seeds and pollen grains and its resulting pollen-ovule ratios provide the basis for understanding interspecific variation in pollen-ovule ratios but that this basis is altered according to the way the size and number of ovules/seeds and pollen grains adapt to a wide spectrum of specific conditions (Figure 6.1). There is strong evidence for a size/number trade off for seeds (Shipley and Dion 1992; Jakobsson and Eriksson 2000; but see Moles et al. 2004a; Greenway and Harder 2007) as well as for pollen grains (Vonhof and Harder 1995; Yang and Guo 2004). Hence, a factor influencing pollen size indirectly affects pollen number and vice versa, and a factor influencing seed size indirectly affects seed number and vice versa. Some factors, like DNA content or plant size, may influence seed and pollen characteristics, while others primarily affect either seed traits or pollen traits (e.g. pistil length). Since the pollen-ovule ratio is a mere mathematical construct, none of the factors governs the pollen-ovule ratio directly. The effect is always mediated through sizes and numbers of pollen and ovules/seeds. However, I found a striking difference between analyses of the correlation of pollen-ovule ratio with seed size, and the correlation of pollen-ovule ratio with pollen size. The latter could be explained by the trade off between pollen size and number alone (chapter 3) while for the former, seed size had an effect on pollen-ovule ratios independent of the seed size/seed number trade-off (chapter 2).

The direct relation between seed size and pollen-ovule ratio (chapter 2) is especially intriguing. This relationship suggests a possible link that connects mating system biology and seed ecology as was already mentioned by Preston (1986). He stated that “if this correlation [between seed size and the pollen-ovule ratio] indicates that resources allocated to male and female function are not independent, then factors influencing variation in seed size, such as seed dispersability and seedling establishment, might also influence the pollen-ovule ratio.” The positive association between seed mass and the pollen-ovule ratio is also interesting from a plant’s strategic viewpoint. Species with large seeds, i.e. a high competition ability but low dispersal po-
tential and short longevity in the soil, tend to be predominantly outcrossing. Small seeded species with low competition ability, high dispersal potential, and high longevity in the soil are predominantly selfing. This view may be simplistic. For instance, also relatively heavy seeds can disperse over great distances with the help of assisting structures like wings. However, it shows a general pattern that has not received much attention though it might bear implications for the reproductive strategies found in plants. Except for the well studied correlation between dioecy and fleshy fruits (Bawa 1980; Thomson and Brunet 1990; Vamosi and Vamosi 2004), pollination ecology and seed ecology are rarely studied in an integrated approach, maybe as a result of the traditional separation of these two subjects. Though the processes of pollination, seed dispersal, and establishment are phenologically separated, the involved characters are affected by similar factors and are components of the same functional and morphological structures. For further studying interspecific variation in pollen-ovule ratios more data should be collected to be able to analyze the factors involved (see Figure 6.1) in a fully integrated approach.

![Figure 6.1](image)

**Figure 6.1** Empirical and hypothetical relationships between seed size, seed number, pollen size, pollen number, and pollen-ovule ratio. The traits are shown as components of an integrated complex. Investment of resources to male and female function is limited to investment in seeds and pollen grains. The size and number of pollen grains and seeds are governed by numerous factors that indirectly also influence variation in pollen-ovule ratios. Solid lines depict established direct relationships (chapters 1 and 2). A negative relationship between pollen size and pollen-ovule ratio (dashed arrow) is predicted by Charnov's model, but only exists indirectly (chapter 2).
6.2 The effect of phylogeny and taxonomy

This thesis provides the first study of the relationship between pollen-ovule ratio and other plant traits that is explicitly based on phylogenetically informed comparative analyses. In a critical review of PIC, Price (1997) objected as a major problem of any PIC analysis and cross species analysis that correlation between traits may be spurious, i.e. confounded by unmeasured variables and thus correlation does not imply causal relationships. That is, a correlation between two traits may simply be observed because both traits are associated with one or several other traits that are not included in the analyses. I tried to minimize this problem by including variables that were likely to covary with the traits studied and by using partial correlation (chapters 2 and 3), and multiple regression and hierarchical partitioning (chapter 4) to analyze trait associations. Price (1997) also opposed authors that advocate to use only PCM for evolutionary questions by stating that “Differences between species and contrast correlations are likely to be of biological significance, suggesting predictable patterns of change...”. Throughout chapter 2 - 4 most of the results for cross species analysis are similar or even nearly identical to the results of PCM. Hence, the observed patterns are not only a product of current adaptation but of processes that maintained throughout evolutionary history. This is especially the case when data were analyzed with PIC (chapters 2 and 3) because a correlation of contrasts measures if the change in a variable is correlated with the change in another variable along the branches of a phylogenetic tree.

In chapter 5 a statistical approach was applied that could also be used to account for the effect of phylogeny, especially when the phylogeny of the studied species is unknown. The method of mixed effect models divides the explanatory variables into random and fixed effects, allowing to account for variation within groups that are defined by the random effects. In chapter 5, however, the method of linear mixed effect models was used to increase the accuracy of predicting pollen-ovule ratios from seed mass by accounting for the variation in the seed size – pollen-ovule ratio relationship within taxonomical groups. Predicted pollen-ovule ratios from mixed effect models were in deed more accurate than predicted pollen-ovule ratios from regression models that did not account for the effect of taxonomy. This result reflects the different patterns in the relationship between the pollen ovule ratio and seed size and emphasizes the importance of considering pollen-ovule ratios and its relationship with other traits in a taxonomical context. It is possible to find major general patterns across a great range of taxonomic groups but investigated at lower taxa levels these patterns can be absent or even reversed (chapter 2 and 3).

6.3 Correlated evolution

The traits that I found to correlate with the pollen-ovule ratio can evolve in a correlated way with the pollen-ovule ratio for two general reasons (Felsenstein 2004, p. 420): (1) The traits are genetically correlated or (2) the selection pressure on the traits are correlated or identical. These two possibilities are not mutually exclusive.
Genetic correlation is given when a selection for the increase or decrease in one trait is followed by a systematic change in the second trait because the two traits are linked by two or more genes or because of pleiotropic effects, i.e. two or more traits are the product of one single gene (Lande and Arnold 1983). A possible genetic correlation between seed size or pollen size and the pollen-ovule ratio has not been investigated so far. Whether such correlations should be expected remains speculation since studies of genetic correlations between male and female investment have gained equivocal results (Charlesworth and Morgan 1991; Brunet 1992; Fenster and Carr 1997; Mazer et al. 1999; Campbell 2000). Mazer (1998) also proposed that results for such investigations may be dependent on whether the study was conducted on an intra-floral or an intra-individual level. Evidence for genetic correlation can only be provided by selection experiments. The results for the interspecific phenotypic correlation between seed size and pollen-ovule ratio (chapter 2) suggest that selection experiments could be a next step for investigating the relationship of the pollen-ovule ratio with floral traits on an intraspecific level.

A possible common selection pressure on seed size and pollen-ovule ratio was investigated in chapter 4, studying the effect of habitat disturbance on interspecific variation in pollen-ovule ratios. However, different degrees of habitat disturbance only affect seed size but not pollen-ovule ratios. Though the intentional question of this paper, if an effect of habitat disturbance on pollen-ovule ratio is accounted for by covariation with seed size, was not directly suited to answer the problem mentioned above, the unexpected result suggests that habitat disturbance is not a common selection pressure on seed size and pollen-ovule ratios. Other environmental variables are conceivable as selection pressure on both, seed size and pollen-ovule ratios. For instance, seed size is correlated with latitude (Moles and Westoby 2003; Moles et al. 2004b). Although the foundations for this correlation are not yet completely understood it would be interesting to investigate if a correlation with latitude also exists for the pollen-ovule ratio.

6.4 Conclusions

This thesis is the first work that attempts to explain the interspecific variation in pollen-ovule ratios by analyzing large numbers of species with help of the comparative method while including information on the phylogeny of the species under study. With this approach it was possible to analyse which factors govern pollen-ovule ratios among current species but also if correlation between pollen-ovule ratios and other traits are the result of evolutionary history.

The following conclusions can be drawn from results in chapters 2-5:

- Charnov’s model for the allocation of resources to seeds and pollen grains can only partially explain variation in pollen-ovule ratios. Contradicting the model, pollen-ovule ratios do not decline with increasing pollen size after accounting for the effect of pollen grain number. In contrast, pollen-ovule ratio does scale with seed size as predicted by Charnov’s model, even after account-
ing for the effect of seed number. Because it suggests a link between pollination/mating system biology and seed ecology, the association between seed size and pollen-ovule ratio merits further research.

- There is no difference in average pollen-ovule ratios between disturbed and undisturbed habitats. Thus, pollen-ovule ratios do not support the reproductive assurance hypothesis. Instead, pollen-ovule ratios reflect a wide range of sexual reproduction strategies in both types of habitat.

- Pollination type, which can be viewed as a qualitative measure of pollination efficiency, influences pollen-ovule ratios. Since this relationship is also evident when accounting for the effect of seed size, Cruden’s statement that “pollen-ovule ratios reflect the likelihood of sufficient pollen grains reaching each stigma to result in maximum seed set” is valid. Cruden’s and Charnov’s view on variation in pollen-ovule ratios are not mutually exclusive and complement each other.

- The detected patterns in pollen-ovule ratio variation are not only evident for current species but also are evolutionary correlated. That is, changes in pollen-ovule ratios through evolutionary time are accompanied by changes in traits like seed size or pollination type.

- When taxonomical variation is accounted for, seed size can be used as an accurate estimator of pollen-ovule ratios.
6.5 References


