

# Can pollination quality limit reproductive output in a self-compatible species? An experimental test with *Narcissus longispathus*

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## BACKGROUND

Pollen limitation has been traditionally characterized by plants receiving an insufficient number of pollen grains to fertilize all their ovules (*quantity limitation*). However, although pollination can also affect the *quality* of the seeds produced by a mother plant, very few studies have considered the possible implications of the “*quality component of pollen limitation*”, (i.e., reduced reproductive output caused by poor-quality pollination; Aizen and Harder 2007).



Fig. 1. *Narcissus longispathus* Pugsley (Amaryllidaceae)

## HYPOTHESIS

One possible way to measure the quality of the seeds produced by plants is to estimate the proportions of seeds produced by self-fertilization and outcrossing. In a self-compatible species incapable of autonomous selfing, in which reproductive output is being limited by pollen quality, we expect to find higher outcrossing rates when arrival of self pollen is prevented, or when arrival of outcross pollen is promoted, in comparison with outcrossing rates obtained from plants under natural pollination conditions.

In this study, we test this prediction and investigate the quality and quantity components of pollen limitation in one population of the endemic daffodil *Narcissus longispathus* (Fig. 1).

## METHODS

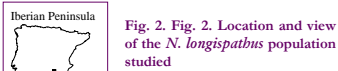


Fig. 2. Fig. 2. Location and view of the *N. longispathus* population studied



**Study species.** *Narcissus longispathus* is a perennial geophyte restricted to a few mountain ranges in southeastern Iberian Peninsula. Flowering plants produce almost invariably a single hermaphrodite flower per inflorescence. The species is self-compatible but in the absence of pollinators flowers hardly ever set seed (Herrera 1995). Flowering takes place in late winter-early spring, a period in which the activity of the species' main pollinator (*Andrena bicolor*, Andrenidae) is frequently reduced. Despite this, Herrera (1995) previously found that seed production is only weakly pollen limited (*quantity limitation*).

**Experimental design.** During 2006 flowering season, we selected 90 flowering plants from a *N. longispathus* population located in Sierra de Cazorla mountains (Jaén Province, Spain, Fig. 2). Each plant was randomly assigned to one of the three following treatments: emasculation (low quality pollen prevented), outcross pollen addition (high quality pollen promoted), and control (natural pollination conditions) (Fig. 3).

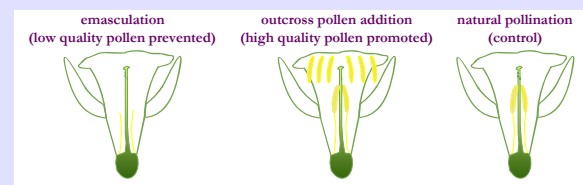


Fig. 3. Schematic representation of the three treatments performed on *N. longispathus* flowers

**Measures of reproductive output.** From each plant we quantified fruit and seed production. Seeds were collected and assayed for six electrophoretic loci using previously described methods (Medrano et al. 2005). We estimated the proportion of seeds produced by outcrossing ( $t$ ) by applying the mixed model (Ritland 1986) to progeny arrays of 8-36 seeds collected from 17-25 plants per treatment (67 maternal plants and 1156 seeds, in total).

## RESULTS

### 1.- Quantitative measures of reproductive output

Emasculated flowers or those supplemented with outcross pollen did not produce more fruits or seeds than naturally pollinated ones (Fig. 4)

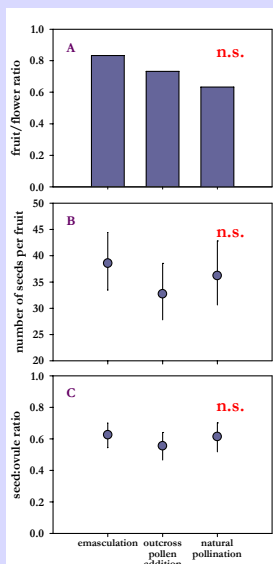


Fig. 4. Quantitative measures of female fertility obtained from the three experimental groups of plants included in this study. A) Proportion of flowers setting fruit, B) Mean number of seeds produced by fruit and C) Mean proportion of ovules that set seeds. Vertical lines indicate 95% CI. (n.s =  $P > 0.05$ ).

### 2.- Qualitative measures of reproductive output

When seed progenies from all plants were pooled by treatment, average outcrossing rate of plants in which arrival of low quality pollen was prevented (emasculated) or arrival of high quality pollen was promoted (outcross pollen added), were both slightly higher than the outcrossing rates of plants under natural pollination conditions (Table 1). However, there was extensive overlap of the 95% CI among all groups, suggesting that differences did not reach statistical significance.

Treatment	Multilocus outcrossing rate $t_m$	Single locus outcrossing rate $t_s$	Correlation of paternity $r_p$	Effective number of mates ( $1/r_p$ )
Emasculation	0.832 (0.026)	0.713 (0.025)	0.339 (0.048)	2.96
Outcross pollen addition	0.784 (0.032)	0.738 (0.032)	0.169 (0.045)	5.92
Natural pollination	0.767 (0.035)	0.751 (0.038)	0.238 (0.069)	4.20

Table 1. Maximum likelihood estimates of mating system parameters obtained for the three groups of *N. longispathus* experimental plants. (SEs in parentheses).

When outcrossing rates were estimated separately for individual plants, a broad range of family-level maximum likelihood outcrossing rate was obtained in all treatments (Fig. 5).

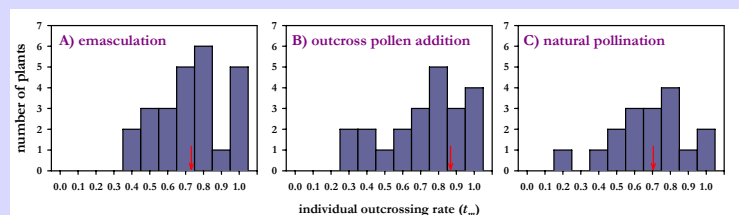


Fig. 5. Frequency distribution of family maximum likelihood outcrossing rate ( $t_m$ ) in the three experimental groups of plants. (Red arrows indicate median values).

## CONCLUSIONS

- In accordance with previous findings, in *N. longispathus* pollination does not limit quantitatively fruit or seed production.
- Our results do not support the pollen quality limitation hypothesis either. This may be attributed to:
  - Low statistical power, given the broad variation observed in individual outcrossing rates.
  - Extensive genetic relatedness among plants of the same population, which may reduce or even obliterate the contrast in *quality* between self and cross pollen. High levels of biparental inbreeding, as denoted by both the differences ( $t_m - t_s$ ) and  $(1 - t_m)$  for emasculated flowers, are compatible with this interpretation.

## REFERENCES CITED

- Aizen and Harder. 2007. Expanding the limits of pollen-limitation concept: effects of pollen quantity and quality. *Ecology* 88: 271-281.
- Herrera. 1995. Floral biology, microclimate, and pollination by ectothermic bees in an early-blooming herb. *Ecology* 76: 218-228.
- Medrano, Herrera and Barrett. 2005. Herkogamy and mating patterns in the self-compatible daffodil *Narcissus longispathus*. *Annals of Botany* 95: 1105-1111.
- Ritland. 1986. Joint maximum likelihood estimation of genetic and mating structure using open-pollinated progenies. *Biometrics* 42: 25-43.