**Colombia – Protocol**

**Location**: Boyacá: Three hours from Bogotá

**Crop: Agraz, Mortiño (*Vaccinium meridionale*)**

**System**: small scale organic farmers. All the sites are within natural or semi-natural habitats of *Quercus humboldtii*

**Cultural practices:** The only management practice consist in removing native "weeds" in some farms. Also some organic fertilization in very few farms. The plants grow naturally, they are generally not sown by the farmers. Each site consist in aprox. 1000 plants at a density of 2000 plants per ha.

**Flowering period:** an extremely asynchronic crop in terms of flowering. On the same plants "new" flowers and rape fruits. Therefore the sampling must be performed by tagging the flowers and fruits to be observed. The flower unit to be measured are "racimes" consisting in 10-20 flowers each.

**Predictors**:

* Honey bees: two levels (with vs. without honey bees within a 2km radius). In each site, the researchers will register the number of colonies within a 2km radius and also the distance of the hives to the site. The researchers will try to reach 3 hives with similar and good colony strength within a 200m radius from site.
* Density of wild insects: measured according to the protocol (see below)

The researchers will measure in each site the elevation, the density of wild insects and the degree of "weed" removal (as percentage). The researchers will also measure the diversity of plants and the cover of these plants within the sampling plot.

These variables could be employed in a regression model with categorical variables, e.g. to evaluate the interactive effects of wild insects and honey bees on crop yield. The sites with both treatments (high vs. Low honey bees) should be at similar elevations, receive similar management and share similar densities of wild insects. The same idea applies to other variables that are relevant for crop pollination. The sites with the different treatments should be spatially mixed.

Before performing the flower and pollinator samplings, the researchers will measure all these co-variates in16-24 sites. With these co-variables measured the researchers will select the sites to be assigned to the honey-bee treatments.

**Replication**: 6-10 sites with honey bees and 6-10 sites without honey bees, 12-20 sites in total

**Mesh treatment:** in each site 50 racimes of flowers in each of four plants are going to be excluded from pollinators during flowering. The four plants should be at least 5 meters apart. The mesh should be installed just before the opening of the flowers and removed when the flowering period is finished. Also the flowers should not have "holes". The mesh should not change climatic or other environmental characteristics and should allow wind pollination.

50 racimnes per plant, 4 plants per site, 12-20 sites, therefore 2400-4000 exclosures. We measure yield quality and quantity in these exclosures (see below).

**Variables to measure (see data sheets):**

1. **Site characteristics:**

To will be measured only once per site. The researchers will also measure the diversity of plants and the cover of these plants within the sampling plot. All the sites are located within a natural habitat patch.

1. **Pollinator density**
2. **Pollinator diversity**
3. **Flower density and phenology**

The three aspects above are going to be measured 4 times for each of 12-20 fields, therefore, 48-80 measurments in total: one morning, two mid-days, one afternoon. Each field will be visited once per two weeks.

1. **Crop yield quantity and quality:**

1 harvest, 12-20 fields.

8 trees per open treatment. Four of these 4 trees will have racimes with exclosures from which the researchers will also harvest (i.e. On these 4 trees they will harvest in the open and the enclosure racimes).

**Work schedule**

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| --- | --- | --- | --- | --- | --- | --- | --- |
| **Variables to measure** | **J** | **J** | **A** | **S** | **O** | **N** | **D** |
| Site characteristics and selection | X |  |  |  |  |  |  |
| Pollinator density |  | X | X |  |  |  |  |
| Pollinator diversity |  | X | X |  |  |  |  |
| Flower density and phenology |  | X | X |  |  |  |  |
| Crop yield quantity and quality |  |  |  |  |  |  | X |

**Data sheets**: see data sheets attached. These data sheets show the variables to be measured and they are prepared to understand the design and the differences and similarities of this design to other studies in other countries. The researchers will shorten these data sheets to get better formats for field sampling.

**More details:** are explained in the "Protocol to detect and assess pollination deficits in crops: a handbook for its use".