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**Euglossine bees as potential bio-indicators of coffee farms: Does forest access, on a seasonal basis, affect abundance?**

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**Abstract:** In order to understand the implications of agriculture on the environment, ecosystem health must be measured. Observing the presence of a biological indicator within an ecosystem is one such method. In this study, male euglossine bees were observed using as attractant cineole 1:8, at adjacent organic (La Paz) and conventional (La Carena) coffee farms near the Northern Barranca River, San Ramón, Alajuela, Costa Rica. Simultaneous data collections were conducted on both farms in April 2004 (late-dry season) and June 2004 (early-rainy season) and combined with the findings of August 2004 (mid-rainy season). These observations show that orchid bees are a viable bio-indicator of organic farm health on a seasonal basis. In the dry season there was no significant difference in orchid bee abundance between the two farms. There is a strong tendency for more bees during the rainy season, suggesting that orchid bee abundance is linked to seasonality and forest access. Rev. Biol. Trop. 54 (4): 1189-1195. Epub 2006 Dec. 15.

**Key words:** orchid bees, *Euglossa*, deforestation, organic agriculture, bio-indicator, Costa Rica.

The onset of commercial agriculture and increasing urbanization has a considerable impact on natural ecosystems throughout the developing world. In Costa Rica, the national economy relies heavily upon agricultural exports, where coffee was the first successful export and today remains the number two crop for exportation, just behind bananas. Forest ecosystems across the country have been progressively converted to commercial farms and pasturelands. Besides the altered water flow, soil erosion, and loss of biodiversity caused by this deforestation (Schelhas 1996), the negative implications of conventional agriculture include pesticide residues, loss of soil productivity, and groundwater contamination. According to Humphries et al. (1995), biological diversity around the world is being lost at 1 000 times the natural extinction rate as a result of the alteration of natural habitats, pollution, and natural habitat degradation caused by human activities. This manipulation of the land is creating environmental disturbances that should be quantified.

Saunders et al. (1990) state that integrated landscape management is critical for conservation of biodiversity on a macroscale. Organic farming, in which agroecology is considered in the context of sustainable production techniques, is one such alternative. The International Federation of Organic Agricultural Movements (IFOAM) defined organic farming as agricultural systems that promote environmentally, socially, and economically sound production (Stolton et al. 2000). These systems value local soil fertility within natural ecological cycles, and prohibit the use of synthetic chemical fertilizers, pesticides, and pharmaceuticals in an effort to limit harmful external inputs.
Beginning in the 1970s, organic agriculture has progressively gained esteem throughout the world as a viable means of production as well as a healthier alternative for both humans and the natural environment (Hedström et al., 2006). Due to growing international markets for organic products, including coffee, organic farming in Costa Rica has emerged as a financially viable and “eco-friendly” alternative to conventional farming.

In order to quantitatively compare the environmental health of organic and conventional farming practices in Costa Rica, effective biological indicators for this habitat need to be established. The use of biological indicators is one of the simplest forms of habitat assessment. Biological indicators are used as measurable surrogates for monitoring environmental health. According to Noss (1990), reliable indicators should be: sensitive to provide an early warning of change; distributed over a broad geographic area; provide a continuous assessment over a wide range of states; independent of sample size; easy and effective to measure and collect; and able to differentiate between natural cycles or trends and those induced by anthropogenic stress. As suggested by Roubik and Hanson (2004), orchid bees are supreme research subjects. With the advent of artificial baits for attracting males, orchid bees have become easy to study, and their relatively large size and spectacular colors also make them more observable than most insects.

Male orchid bees are proposed here as potential biological indicators of tropical ecosystems, specifically the ecosystems of organic and conventional coffee plantations. According to Roubik and Hansson (2004), the Euglossinae subfamily (Hymenoptera; Apidae), is composed of five genera (Aglae, Eufriesea, Euglossa, Eulaena, and Exaerete). Euglossa, with 40 species in Costa Rica (only three endemic species), contains a total of 103 described species in the Neotropics. They are long distance pollinators of low to middle-elevation plants across mainland tropical America, comprising 20-30 % of the bee community’s species in lowland forests (Ackerman 1983, Roubik and Hanson 2004). According to Dressler (1982), large numbers of males may be collected without seriously affecting the reproduction and survival of the population.

Male euglossine bees possess a highly sensitive sense of smell (Schiestl and Roubik 2003) that may allow them to detect chemical disruptions in their habitat such as synthetic pesticides and herbicides. They are highly attracted to perfumes of plants other than orchids, which they collect on specialized hairs on their forelegs and hind tibias for unknown use (Roubik and Hanson 2004). They have a memory for scents relative to their location and can trace the origin of an odor from up to 1 km away (Dressler 1982). According to a study by Ackerman (1983) in central Panama, species richness and bee abundance are correlated; both fluctuate seasonally and peak in the early wet season. Individuals are known to fly long distances, up to 2.5 km, in response to a perfume bait (Dressler 1968), and according to Janzen (1981), will seek a variety of resources in different and distant habitats when their original habitat becomes seasonally severe. Under ordinary circumstances, every orchid bee can cover an area of over 1 000 km² a day (Roubik and Hansen 2004). Janzen (1981) suggests that distances in excess of 20 km are normal for foraging, and greater distances are feasible when resources become scarce. However, according to Ackerman (1983), species composition, evenness, and dominance ranks were virtually non-seasonal, so the male euglossine bee community seems to have some structural continuity.

The hypothesis of our study was based on two assumptions: 1) organic farms are healthier ecosystems than conventional farms, as organic farms seek to mimic the natural diversity of unaltered habitat, and 2) orchid bees may serve as viable biological indicators of ecosystem health because they exhibit many of the aforementioned qualities of biological indicators. Variables such as seasonality has been taken into account to project the most effective conditions for using euglossine bees as a biological indicator of agricultural ecosystem health. We
expected to demonstrate that organic agriculture is more effective than conventional agriculture in maintaining tropical ecological health. The results of the study conducted in August 2004 (mid-rainy period), were compiled with the previous findings from April 2004 (late-dry period) and June 2004 (early-rainy period). Our hypothesis is that more bees will be found within the organic farm site, implying the organic agriculture methods are less disruptive to the functions of a natural ecosystem.

MATERIALS AND METHODS

Data was collected on two adjacent coffee farms, La Paz (organic) and Cardena Farm (conventional), located near the Northern Barranca River (Norte del Río Barranca), San Ramón, Alajuela Province, Costa Rica (10° 06’ N; 84° 28’ W). Both farms are at an elevation of about 1 000 m above sea level, with an average temperature of 21 °C. There is an approximate 10 m “buffer zone” between the two farms (Fig. 1), with coffee plants inside the zone still considered conventionally-farmed coffee. The Corn Plant (Dracaena fragrans), “caña india” in Spanish, originally introduced from West Africa, is used to bolster the buffer zone. Both farms use a variety of shade tree species, including the Coral Bean (Erythrina sp.) and Guamo (Inga sp.), commonly known in Costa Rica as “poró” and “guaba”, respectively, which enrich the soil through nitrogen fixation. The farms are about 20 km from La Balsa Cloud Forest Reserve, which gradually changes into old secondary forest (Fig. 1). During the dry season (December-April) the patch of deciduous trees on the studied farms and La Balsa forest are relatively bare, yet the wet season (May-November) experiences an explosion of life as the dormant trees produce new growth.

La Paz is a 20 ha, mostly shade-grown coffee farm, owned by the export company Cafetalera Lomas al Río. Approximately 15 ha of the farm land is set aside for growing coffee. The farm was officially certified as organic by Eco-Lógica, a Costa Rican certification firm, following a five-year period of abandonment. Compost is the primary means of fertilization, containing coffee hulls from each past harvest and earthworms added to facilitate decomposition. According to the local manager of the organic farm (M. Mora, pers. comm.), an organic soil mixture is spread in 5 cm layers around the base of the coffee plants at routine intervals. The Cardena Farm is a private 1.5 ha plot of a conglomerate of conventional coffee farms. The Cardena Farm, owned by Mr. Fulvio Cardena Montero, uses a strong chemical herbicide once a year in May, and insecticides is used twice a year during the rainy season (M. Mora, pers. comm.).

Data were collected on April 20-21, June 29-30, and August 3-4, 2004, at four stations on each farm (eight total data collection sites) using chemical baits to attract male euglossine bees. Each local study station was spaced 50 m apart (April and June, 2004), and 100 m apart (August 2004), and 50 m into the farm from the edge. The distance between the two studied coffee farms was about 1 km apart. Eucalyptus oil (cineole 1:8), a pheromone substitute, was applied with an eyedropper to a coffee filter located on a tree at breast height (approx. 1.5 m). Five drops were reapplied to the coffee filter every 15 min. It was ensured that the drops were fully absorbed by the filter paper.
RESULTS

The total accumulated numbers of observed euglossine bee during the late-dry season (April 2004) were the same (Fig. 2). However, in both studies during the wet season (June 2004 and August 2004), a higher number of bees was observed in the organic as compared to the conventional farm (Fig. 2). The highest cumulative number of bees was observed within the organic farm during mid-wet season (August 2004, cf. Fig. 3). On August 3, a total of 62 bees were observed within the organic farm, while 21 bees were seen in the conventional farm. The following day, a total of 75 bees were observed at the organic farm land, while 26 bees were observed at the conventional farm (Fig. 4).

The t-calculation for August 3, 2004 was 4.57 with df= 6 and the tc = 2.015 (p= 0.003). The to for August 4 was 2.482, the df was 9, and the tc was 2.015 (p= 0.028). The null hypothesis could be rejected because the to values were greater than the tc values.

In each of the two days in August 2004, a higher average number of bees on the organic farm was observed at one time per 15’ min interval. On August 3, an average of 10 bees was seen on the organic farm while an average of three bees was seen on the conventional farm (Fig. 2). The following day, an average of 13 bees was seen on the organic farm while an average of four bees was seen on the conventional farm (Fig. 3). On the second day, observations for the last 15 min (11:15-11:30) occurred under light rain showers.

Fig. 2. Accumulated numbers of observed euglossine bees on the organic and conventional farms during the wet and dry seasons. Data was collected in April (late-dry), June (early-wet), and August (mid-wet) season, 2004, near San Ramón, Alajuela, Costa Rica.

Fig. 3. Percentage of the total accumulated number of euglossine bees observed during the mid-wet season at organic and conventional coffee farms near San Ramón, Alajuela, Costa Rica (on August 3 and 4, 2004).
DISCUSSION

Data from the mid-wet season (August, 2004) demonstrated significant support for the hypothesis that euglossine bees are effective indicators of farm health during Costa Rica’s wet season. Hence the findings show a much higher abundance of euglossine bees within the organic farm, and the t-test further supports the hypothesis. When the results from late-wet (April) and early-wet (June) seasons were taken into account, more intricate conclusions linked to seasonality were drawn. The most significant difference in abundance between the two farms was found in the studies during the wet season. The months of July and August are in the midst of the Costa Rican wet season.

When observations were conducted during the late-dry season in April 2004, there was no difference in bees observed on the organic and conventional farms (Fig. 4). By the end of June 2004 (early-wet season), however, a considerable difference in bee abundance was found between the two farms, with many more bees observed in the organic environment. The August 2004 data (mid-wet season) continued this trend. These findings suggest that orchid bees in lowland Costa Rica were most abundant during the midst of the wet season.

A study by Ackerman (1983) on Barro Colorado Island in central Panama confirmed that euglossine bee activity may fluctuate on a seasonal basis. A similar study, also conducted in Panama (Roubik 1993), found that bees are most abundant during the mid-dry season to early-wet season. This trend was explained by the availability of resources for male euglossines, since orchids and their fragrances are ubiquitous during the wet season. In addition, during two of the 15 min observations on August 4, 2004, it began to rain. Orchid bees seek shelter and are seldom seen during rain (Hedström, unpubl. data), a variable that could affect the total number of bees observed.

The apparent seasonal variation in male euglossine abundance and their capacity for long flight distances (Roubik and Hanson 2004), make the issue of forest cover an important variable that must be taken into account in their use as biological indicators for sustainable agriculture. Euglossines have a preference for dense and humid forest (Dressler 1982, Roubik and Hanson 2004). The findings from an earlier study in June 2004 (cf Fig. 5, Hedström, unpubl. data) within and in the surroundings of the organic multi-crop farm Luna Nueva, La Tigrá, San Carlos, Costa Rica, supported this conclusion. However, the willingness of the orchid bees to travel long distances to obtain resources, especially in the context of fragmented forest patches, may explain the presence of orchid bees on farms with a...
proximity to primary and/or secondary forest. The wet season is particularly important for this migration process, as forest cover may provide a biological corridor for bees to travel within. In the case of La Paz and Cardena farms, the closest forest, La Balsa, is approximately 20 km away, separated by other small farms and pasture lands (Fig. 1). After reaching La Balsa, the next substantial forest habitat is found at Monteverde Cloud Forest Reserve. During the dry season, the deciduous trees of the intermediate small farms and pasture area lose their leaves, thus deterring orchid bees to leave the forest in search of orchids and other resources. During the wet season, however, these trees can probably act as a biological corridor, allowing bees to migrate and exhibit their preference for an organic farm over a conventional farm.

Previous studies by Roubik (1993, 2001) have shown that higher numbers of male bees may occur in “hot spots”, presumably due to physical factors, nearby concentration of resources, or nests and male resting or display sites. Any absence in their populations after a major forest clearing seemed to not be due to a suddenly restricted habitat size (Becker et al. 1991, Cane 2001), but to a high level of disturbance (Roubik and Hanson 2004).

Once the process of regeneration begins and disturbance diminishes, the bees may spread to formerly isolated patches. If food and nesting resources become especially abundant, secondary vegetation may even support a higher diversity of euglossines (Roubik and Hanson 2004). Therefore, it is not necessarily the size or age of the vegetation, but rather the resource quality, which matters to euglossine bees. Seasonality is directly related to this variable, as the wet season provides more forest and vegetation cover for bees to travel long distances. The remarkable stability of orchid bee populations, even in the face of ongoing deforestation, may make them ideal to use to monitor Costa Rican agroecology. However, male euglossines seem to make effective biological indicators only if that their habitat has not been altered to the extent that they cannot travel to the monitored area.

In conclusion, male euglossine bees were observed using a known attractant (cineole 1:8) at organic and conventional coffee farms. These observations have shown orchid bees to be a viable bio-indicator of organic farm health on a seasonal basis. Hence, orchid bees may offer an effective means for measuring the health of an ecosystem. They are particularly useful during the rainy season, when the area between farm and forest is green, providing a biological corridor by which the bees may travel. It is important to note that both farms sit 20 km from the nearest forest (Fig. 1) and use the same diversity of shade trees (Erytirina sp. and Inga sp.) and coffee plants, yet the orchid bees still were more abundant in the organic farm. Since the orchid bees have a acute sense of smell, it is possible that the unnatural odors of agricultural chemicals used in conventional coffee farms interfere with their search for natural fragrances. Our findings demonstrated that euglossine bees may have a natural preference for undisturbed habitat, and thus will choose to forage in an organic coffee farm over a conventional coffee farm. However, seasonality and its corollary, forest cover, are critical variables of this assessment. It seems that euglossine bees will only travel in large numbers during the wet

Fig. 5. Total accumulated numbers of observed male euglossine bees in pristine forest, organic farm, and pasture land within and in the surroundings of the organic multi-crop farm Luna Nueva, La Tigra, San Carlos, Costa Rica, on June 23 and June 24, 2004.

Fig. 5. Número total acumulativo de machos de abejas euglosinas en bosque prístino, cultivos orgánicos y pastizales, dentro y en los alrededores de los cultivos orgánicos multicosecha: Luna Nueva, La Tigra, San Carlos; Costa Rica; 23 y 24 de junio del 2004.
season, traveling through deciduous foliage between the forest and the farm. It was only during this wet season migration that a clear preference for organic versus conventional farm ecosystems was shown. Therefore, it is quite possible that orchid bees are an effective biological indicator of organic coffee farms during the wet season only.

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