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## Species composition and heterogeneity of blowflies assemblages (Diptera: Calliphoridae) in urban-rural gradients at regional scale in Argentinean Patagonia

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### **ORIGINAL ARTICLE**

# Species composition and heterogeneity of blowflies assemblages (Diptera: Calliphoridae) in urban–rural gradients at regional scale in Argentinean Patagonia

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This work is aimed at describing the spatial distribution of Calliphoridae species along urban–rural gradients in the southern Patagonian steppe, and the influence of geographical factors on species composition and relative abundance. Blowfly assemblages from 12 localities of Santa Cruz and Tierra del Fuego, Argentina between 1997 and 2007 were studied. Samples were assigned to three categories of sites (wild sites, mid-urbanized and urbanized) and four environmental variables were recorded. Three dominant species account for 98% of all specimens, the wild *Compsomyiops fulvicrura*, and two synanthropic species: *Lucilia sericata* and *Calliphora vicina*. Additionally we tested the homogenization of urban calliphorid fauna by comparing the turnover of species among sites, but our findings showed similar heterogeneity of urban and non-urban assemblages.

El objetivo de este trabajo fue describir la distribución espacial de las Calliphoridae presentes en gradientes urbano-rurales de la estepa Patagónica austral, y la influencia de factores geográficos sobre su composición y abundancia relativa. Se registró la composición de califóridos en 12 localidades de Santa Cruz y Tierra del Fuego, Argentina entre 1997 y 2007. Las muestras fueron asignadas a tres categorías de sitio (urbano, semi-urbano y no habitados) y se registraron cuatro variables ambientales. Tres especies dominantes acumularon el 98% del total capturado de especimenes, la asinantrópica *Compsomyiops fulvicrura*, y dos especies sinantrópicas: *Lucilia sericata* y *Calliphora vicina*. Adicionalmente se puso a prueba la homogenización de la fauna de callifóridos urbana, mediante la comparación del recambio de especies entre sitios, pero nuestros resultados indicaron una similar heterogeneidad de las comunidades urbanas y no urbanas.

Keywords: Argentina; Calliphoridae; diversity; Patagonia

#### Introduction

Species composition and relative abundance of Calliphoridae are strongly influenced by human intervention on natural environments, especially when urbanization processes are involved. Related to these processes, the term synanthropy has been proposed to describe species associated with human settlements (Nuorteva 1963).

Synanthropy of calliphorid species has been considered for many locations of the world (Hwang & Turner 2005). However, few studies have focused on synanthropy in relation to geographical variables at a regional scale (Nuorteva 1963; Baumgartner & Greenberg 1985).

Studies of blowfly diversity along rural–urban gradients show that urban environments exhibit lower species diversity than wild environments (McKinney 2002, 2008). Species loss at urban sites may be attributed to several causes, such as isolation,

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distance to natural habitats, and the increased ecological disturbance displayed by human settlements (Faeth & Kane 1978; Blair & Launer 1997; McIntyre 2000). Moreover, those species regarded as "urban exploiters" would represent the most homogenized fauna, mainly composed by a small subset of cosmopolitan species adapted to urban conditions (Denys & Schmidt 1998; McKinney 2002, 2006). This trait attributed to urban muscoid flies communities suggests the existence of higher similarity between urban environments of different biogeographic regions than their natural adjacent ecosystems (McKinney 2006).

Currently, six blowfly species are known from the Patagonian steppe: *Calliphora vicina* (Robineau-Desvoidy), *Lucilia sericata* (Meigen), *Compsomyiops fulvicrura* (Robineau-Desvoidy), *Sarconesia chlorogaster* (Wiedemann), *Chlorobrachicoma versicolor* (Bigot) and *Protophormia terraenovae* (Robineau-Desvoidy) (Mariluis & Mulieri 2003; Schnack & Mariluis 2004; Mariluis & Schnack 2005).

The diversity of blowfly pest species in Patagonia and the identification of their preferred habitats have important medical and veterinary relevance. Species like P. terraenovae may be a potential parasite of cattle, sheep and reindeer (James 1947), and Ca. vicina is usually referred to as secondary myiasis agent, although, in some cases, it has been recorded as primary invader (Zumpt 1965; Delhaes et al. 2001). On the other hand, L. sericata was locally recorded as myiasis agent in man (Mariluis & Guarnera 1983; Visciarelli et al. 2007) and as forensic indicator as well (Oliva 2001). This species is responsible for the condition known as "blowfly strike". Females of L. sericata leave their eggs on carcasses, wounds and, in particular, on the wool of sheep soiled with urine, faeces or blood (Hall & Wall 1995). Although there is an enormous body of literature on myiasic blowfly agents, little attention has been played to the incidence of larval infestation in relation to the main sheep farming areas of South America, which includes the Patagonian Region.

This work is aimed at describing the spatial distribution of calliphorid species along urban–rural gradients in the southern Patagonian steppe, their degree of synanthropy and the influence of geographical factors such as latitude and elevation on species composition and relative abundance. In addition, we tested the homogenization of urban calliphorid fauna by comparing the turnover of species among sites exhibiting different levels of human influence ("beta diversity").

#### Materials and methods

#### Study area

The Patagonian steppe is a cool semi-desert ecosystem that occupies the southern portion of Argentina. The climate is semiarid with 100–270 mm of annual precipitations, concentrated in winter, and declining from west to east. In the southern portion of the study area, the so-called Magellanic steppe, the climate is semiarid to sub-humid, with oceanic influence. Annual rainfall is 350–400 mm. Cold weather and strong winds are common features all year long.

The vegetation of the entire area is mainly dominated by shrublands of *Chuquiraga*, *Nassauvia*, or *Senecio* spp., and grasslands of *Stipa*, *Poa*, and *Festuca* spp. (Cabrera 1971).

#### Sampling methods

This work was based on equal trapping effort of calliphorids in 36 sites covering a large part of the south Patagonian steppe. Blowflies are very mobile organisms which make them difficult to sample at randomly-selected locations. Thus, collecting methodology is mainly based on the use of aggregation sites, such as fly-attracting baits (Muirhead-Thomson 1968). We sampled blowflies during the late spring and summer (November to February), once a month per site. Seven hourly captures of adult flies (10:00–16:00 h) were taken with a hand net on a bait composed of 150 g of rotten cow meat (five days old). The baits were exposed for 15 minutes every hour. The sampling effort was the same at each site (one bait per site and seven netting events per date) and data collection took place when weather conditions were appropriate (i.e., sunny and dry).

Samples were taken in 12 localities of Santa Cruz and Tierra del Fuego provinces (Figure 1). Three sampling sites were selected at each location. The sampling sites were assigned alternatively to three categories according to their level of human alteration: (1) urbanized, (2) mid-urbanized, (3) wild sites (Table 1). The urbanized sites were located in the geographical center of cities and comprised those areas with paved streets and dense presence of buildings. The midurbanized sites were located at the city edges or in small towns. Finally, the wild sites were located in the steppe, 5–8 km away from urban limits. Blowflies were killed in glass vials with carbon tetrachloride and preserved in paper containers. The sampled flies are currently deposited at ANLIS, Instituto Nacional de Microbiología "Dr. Carlos Malbrán". Identification of specimens was made using the key included in Mariluis & Schnack (2002).

Four variables of potential environmental importance were recorded at each site: GPS coordinates, elevation, degree of urbanization and daily mean temperature.

#### Statistical analysis

Raw counts of calliphorids reflect the local fauna at the particular site and date, rather any fundamental differences among assemblages (MacLeod & Donnelly 1962). We performed an ANCOVA analysis including latitude and elevation as continuous predictors, and degree of urbanization (as categorical levels of the independent variables) to test the relationship between total abundance of calliphorids and environmental variables.

Relationships between faunistic composition and environmental variation were explored using Canonical Correspondence Analysis (CCA) (Braak 1986). In this analysis, linear combinations of environmental variables are selected to obtain the separation of the species distribution into the multivariate space. The proportional abundance of six species at each of



0

300 km

4. Puerto Deseado
5. Puerto San Julián
6. El Chaltén
7. Tres Lagos
8. Hipólito Yrigoyen
9. Gobernador Gregores
10. Puerto Santa Cruz
11. Río Gallegos
12. Río Grande

Figure 1. Map of Santa Cruz and Tierra del Fuego provinces showing the sampled locations in Argentinean Patagonia.

the 36 sites was used to build the species-site matrix. The environmental matrix included four variables: latitude, elevation and temperature as continuous numerical variables, and urbanization degree as an ordinal numerical variable.

Caleta Olivia
 Los Antiguos
 Perito Moreno

Relationship of urban degree and the presence of dominant species were analysed univariately. Thus, data of proportional abundance between types of sites were compared using one-way ANOVA. We assume there were no effects between years in which the samples were taken. For this reason, year-site cases were included as independent observations. In addition, a Synanthropy Index (SI) (Nuorteva 1963) was calculated for dominant species (those included in all three categories of urbanization) at eight locations.

Association between proportional abundance of individual species and latitude were performed using nonparametric Spearman Rank Correlation (Zar 1996). The same analysis was adopted to correlate latitude with those values of the SI calculated for the dominant species.

We analyzed faunistic variation and spatial distance and calculated the similarity between all possible pairs of sites within each category of urbanization

Table 1. Geographic locations and characteristics of sampling sites in Argentinean Patagonia.

Locality	Latitude S	Longitude W	Population size	Sampling period	Type of site sampled <sup>a</sup>
Río Gallegos	51°38′57″	69°14′20″	79,144	1997–1998	1, 2, 3
Puerto Santa Cruz	50°01′00″	68°31'00″	3397	1997-1998	1, 2, 3
Río Grande	53°48′45″	67°43′11″	55,131	1997-1998	1, 2, 3
Puerto Deseado	47°45′11″	65° 55′ 42″	10,237	2004-2005	1, 2, 3
Caleta Olivia	46°26′17″	67°32′05″	36,077	2004-2005	1, 2, 3
Puerto San Julián	49°17′60″	67°43′00″	6143	2004-2005	1, 2, 3
Los Antiguos	46°32′60″	71°37′00″	2047	2005-2006	2, 2, 3
Hipólito Yrigoyen	47°34′06″	71°44′24″	171	2005-2006	2, 2, 3
Perito Moreno	46°36′00″	70°55′00″	3588	2005-2006	1, 2, 3
El Chaltén	49°19′44″	72°55′48″	324	2006-2007	2, 3, 3
Tres Lagos	49°37′00″	71°30′00″	186	2006-2007	2, 2, 3
Gobernador Gregores	48°45′05″	70°14′40″	2519	2006-2007	1, 2, 3

<sup>a</sup>1, urbanized; 2, mid-urbanized; 3, wild sites.

using Percentage Similarity Index (PSI) (Krebs 1999). ANCOVA analysis was used to describe the effects on faunistic variation using category of urbanization (as dummy variables) and distance as independent variables. Data of PSI were normalized by arcsine transformation.

#### Results

Samples taken contained the six blowfly species previously recorded for the Patagonian steppe, and a total of 19,383 specimens were counted. Three dominant species account for 98% of all specimens. *Compsomyiops fulvicrura* was the most abundant species with 8110 individuals, followed by *L. sericata* and *Ca. vicina* with 6310 and 4597 individuals, respectively.

Latitude and elevation of sampling site had no effect on the abundance of blowflies. Both variables showed nonsignificant relationship with the abundance detected at the three levels of urbanization sites (ANCOVA  $F_{[4, 31]} = 1.180$ , P = 0.338) (Table 2).

The CCA showed a significant relationship between the calliphorid assemblage and the environmental measures of urbanization degree, latitude, temperature and elevation at the individual sampling sites. The first two

Table 2. Results of the ANCOVA model testing the effects of environmental variables on blowfly abundance.

$\begin{tabular}{ c c c c c c c c c c c c c c c c c c c$						
Intercept         1         93.4         93.4         0.329         0.3           Latitude         1         225.7         225.7         0.796         0.3           Elevation         1         285.7         285.7         1.003         0.3           Urbanization         2         569.6         284.8         1.003         0.3           category         Error         31         8795.3         283.7           Total         35         10,134.6         10,134.6		df	s.s. <sup>a</sup>	m.s. <sup>b</sup>	F	Р
Latitude 1 225.7 225.7 0.796 0.3 Elevation 1 285.7 285.7 1.003 0.3 Urbanization 2 569.6 284.8 1.003 0.3 category Error 31 8795.3 283.7 Total 35 10,134.6	Intercept	1	93.4	93.4	0.329	0.570
Elevation         1         285.7         285.7         1.003         0.3           Urbanization         2         569.6         284.8         1.003         0.3           category         2         569.6         283.7         1.003         0.3           Total         35         10,134.6         10         10         10	Latitude	1	225.7	225.7	0.796	0.379
Urbanization 2 569.6 284.8 1.003 0.3 category Error 31 8795.3 283.7 Total 35 10,134.6	Elevation	1	285.7	285.7	1.003	0.323
Error318795.3283.7Total3510,134.6	Urbanization category	2	569.6	284.8	1.003	0.378
Total 35 10,134.6	Error	31	8795.3	283.7		
	Total	35	10,134.6			

<sup>a</sup>s.s. = sums of squares; <sup>b</sup>m.s. = mean squares.

ordination axes explain 38.2% and 13.3% of the variance data. A Monte-Carlo test (with 99 permutations) indicated that species distribution along the first two axes was not at random (Axis I, P = 0.01; Axis II, P = 0.01). Moreover, these axes were successful in separating the most urbanized sites (upper and left quadrant), the mid-urbanized (center) and the wild sites (lower and right) (Figure 2). On the other hand, the southernmost sites were placed at the negative half of both axes. The length and direction of the arrows (Figure 2) indicate that the mean temperature and latitude are redundant measures, and that elevation shows a minor contribution to separate data of sites or species.

The synanthropic degree of the species was ranked according to the urbanization category variable. While *Ca. vicina* and *L. sericata* showed more synanthropic habits, with the later species associated to the northern sites, *Co. fulvicrura* and *S. chlorogaster* were associated with wild sites. The third group of species (*Ch. versicolor* and *P. terraenovae*) was associated with mid-urbanized sites and were more abundant in the southern portion of the region (Figure 2).

The relative abundance of a given species along the latitudinal gradient was assessed by using correlation coefficients at each category of urbanization (Table 3). Obtained results showed that *Ca. vicina* was more abundant at higher latitudes, especially for nonurbanized sites, while *L. sericata* exhibited lower levels of abundance in the southern urban sites. *Sarconesia chlorogaster* showed higher density at lower latitudes, while *P. terraenovae* was significantly associated to high latitudes. Finally, *Co. fulvicrura* and *Ch. versicolor* did not show a definite trend in the study sites (Table 3).

Among the dominant species, the most successful colonizers of urban sites were *Ca. vicina* (ANOVA,  $F_{[2, 33]} = 5.502$ , P = 0.008) and *L. sericata* (ANOVA,  $F_{[2, 33]} = 4.952$ , P = 0.013). These synanthropic species



Figure 2. Ordination diagram of the first two axes of CCA for 36 samples sites (A), and six blowfly species and four environmental variables (B) recorded in South Argentinean Patagonia. Arrows represent directions of greatest change in environmental variables.

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	Urban $(n = 8)$	Isolate dwellings $(n = 15)$	Wild $(n = 13)$	All sites $(n = 36)$
Calliphora vicina	r = 0.517	r = 0.742	r = 0.622	r = 0.536
*	P = 0.085	P = 0.006	P = 0.031	P = 0.001
Lucilia sericata	r = -0.827	r = -0.153	r = 0.017	r = -0.268
	P = 0.001	P = 0.634	P = 0.956	P = 0.113
Compsomyiops fulvicrura	r = -0.196	r = -0.446	r = -0.242	r = -0.282
	P = 0.541	P = 0.146	P = 0.447	P = 0.096
Sarconesia chlorogaste	r = -0.651	r = -0.772	r = -0.713	r = -0.691
	P = 0.022	P = 0.003	P = 0.009	P < 0.001
Protophormia terraenovae	r = 0.663	r = 0.652	r = 0.663	r = 0.658
*	P = 0.019	P = 0.021	P = 0.019	P < 0.001
Chlorobrachicoma versicolor	r = 0.257	r = 0.350	r = -0.271	r = 0.154
	P = 0.421	P = 0.265	P = 0.393	P = 0.370

Table 3. Correlation coefficients (Spearman rank correlation) obtained between proportional abundance and latitude for blowfly species.

exhibited higher abundance in urban sites (Figure 3). The opposite trends was displayed by *Co. fulvicrura* (ANOVA,  $F_{[2, 33]} = 7.237$ , P = 0.002) (Figure 3). All these results were corroborated with the SI calculated for the three species (Table 4). However, the SI calculated for these species showed different trends along the latitudinal gradient. While *L. sericata* and *Co. fulvicrura* did not change their values of SI, *Ca. vicina* exhibited lower SI values at higher latitudes (Table 4). Numerical variation of SI recorded for *Ca. vicina* can be attributable to its higher abundance at non-urban sites in the southern locations.

Finally, to test the urban homogenization of the calliphorid assemblages, we used a linear model to describe the relationships between faunistic similarity and distance between pairs of sites within each category of urbanization. Analysis of covariance was performed in order to determine any differences in the regression lines for the urbanized, mid-urbanized and wild sites. The result obtained (Table 5) showed similar negative slopes relating faunistic similarity (PSI) and distance between sites for the three categories of urbanization (e.g. pair of sites separated by the same distance showed similar faunistic differences, independently to the category of urbanization) (Figure 4).

#### Discussion

Results suggest that the three dominant blowfly species are spatially segregated according to two main environmental factors: the urban-rural and the latitudinal gradients. It seems that along the urban-rural gradient *Co. fulvicrura* exhibits a strong preference for noninhabited areas in contrast to the urban exploiters, like *Ca. vicina* and *L. sericata*. These trends have been previously observed in Patagonian localities while describing the association of *Ca. vicina* and *L. sericata* to human settlements, and the preference of *Co. fulvicrura* for environments less affected by human intervention (Schnack & Mariluis 2004; Mariluis et al. 2008).

In warmer environments such as the Buenos Aires province, synanthropic species show temporal variations, where L. sericata is mainly present during warm months and Ca. vicina is more abundant during cold months (Mariluis & Schnack 1986). In Santa Cruz and Tierra del Fuego provinces, these species showed geographic segregation. According to this, the warmloving species, L. sericata, was better represented in northern urbanized areas of Santa Cruz province, Ca. vicina was strongly dominant south of Santa Cruz and Tierra del Fuego provinces. This segregation is probably related to temperature variations observed across the latitudinal gradient. In northern locations, such as Puerto Deseado, historical records of temperature during November to February ranged between 7 and 22°C, while in southern locations, such as Río Grande, historical records of temperature during November to February ranged between 3 and 14°C (Instituto Gegráfico Militar 1998).

According to the recorded data, latitude strongly influenced the degree of synanthropy of the dominant species. This trait was specially displayed by *Ca. vicina* which behaved as an urban exploiter at northern locations, being a more generalist species towards the southern locations. Furthermore, *P. terraenovae* is only present at the Magellanic portion of the study area. Both cosmopolitan species account for 85% of all captures in the three sites at Rio Grande (Tierra del Fuego), being therefore good indicators of an extremely disturbed community.

Mack et al. (2000) suggest that community resistance to invasions increases in proportion to the number of species in the community (species richness).



Figure 3. Proportional abundance (mean  $\pm$ SE) of dominant blowfly species in South Argentinean Patagonia. Different letters above the bars indicate significant differences.

Table 4. Synanthropic Index (SI) (mean  $\pm$ SE) and correlation coefficient (Spearman) between the SI calculated for dominant Calliphoridae species and latitude of sampled locations.

	SI	r s	п	Р
Calliphora vicina	72 (±25)	-0.786	8	< 0.05
Lucilia sericata	49 (±37)	-0.643	8	NS
Compsomyiops fulvicrura	0 (±37)	-0.214	7	NS

According to this hypothesis, those species-rich communities are more "stable". Conversely, the low number of native species makes the communities more vulnerable to alien species invasions. This suggestion is conceptually related to the vacant niche hypothesis (Elton 1958). In this sense, the wild Patagonian calliphorid communities have lower richness of native species compared to other areas of the Neotropics. The presence of *P. terraenovae* and the wider spatial niche of *Ca. vicina* along the urbanization gradient in the Magellanic portion of the Patagonian Region can be explained by the referred intrinsic vulnerability exhibited by the wild blowfly communities susceptible to be invaded by non-native species.

Blowfly assemblages are affected by many factors. Among abiotic components, temperature and photoperiod are the most important, affecting significantly the rate of development of an individual species (Wall et al. 2000; Pitts & Wall 2005). Both, *Ca. vicina* and *P. terraenovae* are well adapted to the climatic conditions of the extreme southern continental portion. Furthermore, *P. terraenovae* has a Holarctic distribution and is very common in the colder regions. In fact, this species exhibits, among calliphorids, the highest tolerance to extreme low temperatures (Grassberger & Reiter 2002). Similarly, *Ca. vicina* was recorded as thermophobic, being dominant in cold and wet habitats (Martinez-Sanchez et al. 2000). This behavior was detected in both temporal and spatial scales, with the species showing significant population increase in winter (Martinez-Sanchez et al. 2000) or occupying higher portions of altitudinal gradients, the latter interpreted as surrogate of climate (Baz et al. 2007).

Concerning the biotic homogenization due to the urbanization process, several factors may influence the heterogeneity pattern observed for urban and nonurban surveyed communities. One explanation may be a resource shift along the geographical space exhibited by some calliphorid species (especially for *Ca. vicina*). Another explanation might be the absence of other, biologically similar species along the latitudinal gradient. These factors might add a similar degree of variability to both urbanized and non-urbanized calliphorid assemblages.

Despite the spatial segregation detected for urban exploiters and wild species we cannot detect higher levels of similarity for urban environments in comparison



Figure 4. Effect of the distance between sampled sites on similarity of blowfly communities.

Table 5. ANCOVA, separate slopes model testing the effects of distance on similarity of blowfly communities.

	df	s.s. <sup>a</sup>	m.s. <sup>b</sup>	F	Р
Intercept	1	54.71	54.71	988.19	< 0.001
Distance	1	3.68	3.68	66.55	< 0.001
Urbanization category	2	0.14	0.07	1.26	0.284
Error	207	11.46	0.05		
Total	210	15.42			

 $^{a}$ s.s. = sums of squares;  $^{b}$ m.s. = mean squares.

to the wild or mid-urbanized sites at South Patagonia. In other words, species turnover was similar for the different type of habitats (e.g. urban or wild). Only the increasing distance between sites contributes most clearly to proportional changes in beta diversity. However, it would be interesting to test the homogeneity of urban exploiters in more comprehensive regional or global scales containing several and different biogeographic areas.

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