# Diptera of sanitary importance associated with composting of biosolids in Argentina

# Dípteros de importancia sanitaria asociados al compostaje de biosólidos en Argentina

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## **Keywords**

Composting. Diptera. Ecology of vectors. Biosolids.

## Abstract

## Objective

Odorous compounds produced at the biosolids composting plant in Bariloche (NW Patagonia) attract a variety of insects, mainly belonging to the order Diptera. In order to characterize these flies, collected specimens were taxonomically identified, their community characteristics were described and their sanitary and synanthropic importance and autochthonous or introduced character were determined.

## Methods

Sampling was performed from October 1999 until March 2000. Adults were collected using an entomological net, and larvae and puparia were obtained from the composting material and incubated to obtain adults. Richness, abundance and sex ratio were calculated.

#### Results

A total of 9 taxa of Diptera were identified: Sarconesia chlorogaster, Phaenicia sericata, Calliphora vicina, Cochliomya macellaria, Ophyra sp., Muscina stabulans, Musca domestica, Sarcophaga sp and Fannia sp. Specimens of Anthomyiidae, Acaliptratae and one larva of Eristalis tenax were also found. Ophyra sp. was the most abundant taxa. All the captured Diptera belonged to introduced taxa. Most of them are considered to be eusynanthropic and/or hemisynanthropic and have sanitary importance as they may cause myiasis and pseudomyiasis. The high number of females registered and the finding of immature stages indicated that flies can develop their complete life cycle on biosolid composting windrows.

#### Conclusions

The characterization of flies obtained in this study may be useful for defining locations of urban or semi-urban composting facilities. It also highlights the importance of sanitary precautions at such plants.

#### **Descriptores**

Compostaje. Diptera. Ecología de vectores. Biosólidos.

#### Resumen

#### **Objetivo**

Los compuestos odoríferos producidos en la Planta de Compostaje de Biosólidos de Bariloche (NO Patagonia) atraen diferentes insectos, principalmente moscas (Orden Diptera). Con el objeto de caracterizarlas, se colectaron especímenes que fueron identificados taxonómicamente. Se describieron sus características comunitarias y se determinó su importancia sanitaria y su carácter autóctono o introducido.

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#### Métodos

El muestreo se realizó desde octubre de 1999 a marzo de 2000. Los adultos se colectaron utilizando una red entomológica. Las larvas y las pupas, obtenidas de las pilas de compostaje, se incubaron para obtener adultos. Se calcularon riqueza, abundancia y relación de sexos.

#### Resultados

Se identificaron 9 taxones de Diptera: Sarconesia chlorogaster, Phaenicia sericata, Calliphora vicina, Cochliomya macellaria, Ophyra sp, Muscina stabulans, Musca domestica, Sarcophaga sp y Fannia sp. También se capturaron especímenes de Anthomyiidae, Acaliptratae y una larva de Eristalis tenax. Ophyra sp fue el taxón más abundante. Todos los especímenes capturados pertenecieron a taxones introducidos y la mayoría de ellos, son eusinantrópicos y/o hemisinantrótpicos. Además, tienen importancia sanitaria dado que pueden causar myiasis y pseudomyiasis. La cantidad de hembras registrada y el hallazgo de estadios inmaduros indican que estas moscas pueden desarrollar su ciclo completo de vida en las pilas de compostaje.

#### **Conclusiones**

La caracterización de las moscas encontradas en la Planta de Compostaje de Bariloche puede ser de utilidad para definir la instalación en zonas urbanas o semi-urbanas de este tipo de plantas. Adicionalmente, aporta información sobre aspectos sanitarios en esta clase de emprendimientos.

#### INTRODUCTION

Composting is one of the best alternatives for the beneficial land use of biosolids. This extensive world-wide practice transforms potentially risky wastes into an odorless, innocuous and stable organic amendment, called "compost". An adequate composting process ensures effective pathogen and odor reduction and, consequently, the attraction of vectors such as insects, birds or rodents decreases drastically.

At the biosolids composting plant in Bariloche (NW Patagonia), 450-500 m<sup>3</sup> of biosolids per month have been composted outdoors in turning windrows since 1998.9 Odorous compounds are naturally produced during this process, especially during the thermophilic period (temperatures above 45°C), when the greatest biological activity takes place, due to degradation of labile compounds. Odors are emitted as moist aerosols when windrows are turned for aeration and may contain appreciable levels of volatile organic acids.<sup>2</sup> This odor emission restricts the installation of composting facilities near urban centers, and is a cause for concern because it attracts potential disease-transmitting vectors. Among these are sarcosaprophagous and/or coprophagous insects, especially flies, which become important as the nexus between different infectious foci and man and his products.3,9,14

Although studies on Diptera in relation to sewage sludge are scarce, the most important families cited are Psychodidae, Syrphidae, Ulidiidae, Biophilidae, Anthomyiidae, Fanniidae, Tachinidae, Muscidae, Sarcophagidae and Calliphoridae, and especially the last three, with a high number of genera and species involved.<sup>3</sup> Members of Calliphoridae have acquired great importance because of their invasion of the Americas,1 which has produced significant changes in local fly communities, such as those recorded in urban ecosystems of Buenos Aires, Argentina.8 Flies can transport agents in their digestive system, and on their legs and wings, that may cause illness to humans and animals. Generally, the pathogenic organisms involved are viruses, bacteria, protozoan cysts and invertebrate eggs (trematodes, cestodes and nematodes). According to their synanthropy, flies can be classified as asynanthropic, hemisynanthropic and eusynanthropic.<sup>10</sup> The last two categories can complete their whole development within the habitats of man and domestic animals, thereby potentially acting as disease vectors.

In Argentina, sludge treatment facilities have only recently been constructed and therefore there is no information about the attraction of vectors. The synanthropy of only a few species has been determined in the Andean Patagonian region.<sup>15</sup>

The objectives of the present work were: i) to identify the Diptera taxa associated with biosolid composting windrows and describe their community characteristics; and ii) to define their sanitary and synanthropic importance and their autochthonous or introduced character.

#### **METHODS**

This study was carried out at the biosolids composting plant in Bariloche, located in a semi-urban area 8 km south of the town (41°05'S – 71°20'W). Biosolids derived from Bariloche's sewage treatment plant are composted outdoors in turning windrows (10-15 m long, 3-4 m wide and 1.6 m high) on concrete platforms, with an open drainage system for lixiviates (Figure). Wood shavings, yard trimmings and recycled windrow material (co-compost) are used as bulking agents at 1:1 ratio by volume, approximately.

The sampling was conducted from October 1999 until March 2000. A new windrow was selected each month and Diptera adults were sampled in January, February and March during the thermophilic composting period (weeks 1, 2 and 3). During each week, three consecutive days were chosen to collect Diptera specimens. To evaluate the evolution of the thermophilic period on the Diptera community, sampling weeks were grouped in the composting weeks (I, II and III) according to the same composting period, independent of the fact that they belonged to windrows from different months (Table 1).

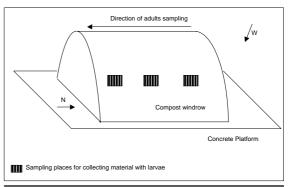
At least 100 flies were randomly caught every day using an entomological net, from all over the windrow (Figure). Specimens were killed using ethyl ether vapor in killer bottles and were then preserved dry on entomological beds, individualized by day of sampling.

Approximately 1kg of material was taken from the windrows (Figure) in October, November, December and February, in order to obtain larvae. The material was put into plastic bags and taken to the laboratory for separating the larvae. Specimens were incubated with windrow material at 25°C, in open plastic bottles covered with a thin cloth and periodic checkings were done in order to detect pupation. Puparia obtained from larva incubation and those found in the windrow material were individually put into open plastic bottles covered with a thin cloth and regularly checked to detect adults.

Minimum and maximum ambient temperatures were registered for each sampling day and monthly averages were calculated.

Table 1 - Sampling weeks grouped by composting weeks.

Sampling month		January			Month February			March		
Sampling weeks	1	2	<sup>′</sup> 3	4	5	6	7	8	9	
Composting week I Composting week II Composting week III	X	Х	X	X	Х	Х	X	X	X	
Windrows used		N 240	)	1	<b>V</b> 24	9	N	<b>1</b> 25	9	



**Figure** - Detail of windrow with sites and direction of sampling.

Diptera adults and puparia were identified<sup>11,17</sup> and adults were also sexed. In addition, taxa were characterized according to their sanitary and synanthropic characteristics and their autochthonous or introduced status.<sup>8,10,13,15,17</sup>

Richness and abundance values<sup>6</sup> were calculated for total, monthly and composting week data. Additionally, sampling week values were calculated for richness.

A  $\chi^2$  test (P<0.05) was applied to evaluate the female-male ratio.

# **RESULTS**

The maximum and minimum temperature averages were 24.4 and 5.1°C for January, 25.7 and 7°C for February and 23.3 and 6°C for March, respectively.

All sampled adults belong to the Schizophora division, of which 2,073 corresponded to the Caliptratae section and 811 to the Acaliptratae section (Table 2). In the latter section, a more accurate identification could not be reached because of taxonomic difficulties. Within the Caliptratae section, 5 families were identified, but the specimens of Anthomyiidae could not be identified down to further levels, and the specimens of Fanniidae and Sarcophagidae only to genus level (Table 2). Muscidae (2 species) and Calliphoridae (4 species) were the best-represented families, not only in relation to the number of species but also to their presence during the sampling period. Ophyra sp (996), Cochliomyia macellaria (534), Muscina stabulans (233) and Fannia sp (216) were the bestrepresented taxa.

The total number of males (520) differed significantly from the total number of females (1430) ( $\chi^2$  =497.81; P<0.05; D.L.=1), and this difference was also significant for the sampling months (Table 3) and

Table 2 - Number and taxa of Diptera collected during sampling and composting weeks.

Sampling weeks Composting weeks	Week 1 Weekl	January Week 2	Week 3	Week 4 Weekl	February Week 5	Week 6	Week 7 WeekI	March Week 8	Week 9	Tota
Composing weeks	VVCCKI	WeekII		VVCCKI	WeekII		VVCCKI	WeekII		
Taxa			WeekIII			WeekIII			WeekIII	
Caliptratae										
Calliphoridae										
Sarconesia chlorogaster (Wiedemann, 1830)	0	0	0	0	0	1	0	1	0	2
Cochliomya macellaria (Fabricius, 1775)	101	151	59	158	14	28	22	0	1	534
Phaenicia sericata (Meigen, 1826)	0	2	1	4	0	0	7	0	2	16
Calliphora vicina (Robineau-Désvoidy, 1830)	0	0	0	0	0	0	2	2	2	6
Ophyra sp	228	114	205	77	116	102	102	42	10	996
Muscidae										
Muscina stabulans (Fallén, 1817)	1	12	29	9	20	100	25	33	4	233
Musca domestica (Linné, 1758)	0	2	4	11	9	8	8	11	5	58
Sarcophagidae										
Sarcophaga sp	1	0	0	2	0	0	0	0	0	3
Fannidae										
Fannia sp	5	1	6	24	27	19	57	73	4	216
Anthomyiidae	0	1	0	0	1	2	1	4	0	9
Acaliptratae	0	31	37	46	11	108	105	144	230	811
Total										2,884

composting week I ( $\chi^2$ =341.82; P<0.05; D.L.=1), week II ( $\chi^2$ =156.5; P<0.05; D.L.=1) and week III ( $\chi^2$ =44.77; P<0.05; D.L.=1).

After 7 to 10 days of larva incubation, nine puparia were obtained. All of them metamorphosed to adults: 7 corresponded to *Ophyra* sp (3 females and 4 males), with an incubation period of 13 to 15 days, and 3 to *C. macellaria* (2 females and 1 male) with an incubation period of 7 to 13 days. During the sampling period, only one larva of *Eristalis tenax* (Syrphidae) was found in windrow lixiviates but no adults were captured.

The total richness of the Diptera community was 9 (Table 2). However, this value should have been higher, because identification of genus and species for Anthomyiidae and Acaliptratae could not be performed. The richness averages for the sampling weeks increased over the course of the months: January (5.7), February (6.0) and March (6.7) but rich-

ness averages for composting weeks did not sustain this trend for the same period.

Among the Caliptratae, the highest value for total abundance was for *Ophyra* sp, followed by *C. macellaria* (Table 4). *Muscina stabulans* and *Fannia* sp presented similar abundance values, but much lower than for the former two taxa. Analysis of monthly abundance showed that the highest values for *C. macellaria* and *Ophyra* sp were in January and February, and for *Ophyra* sp and *Fannia* sp in March. In the composting weeks, the highest values were always for *Ophyra* sp. The other Caliptratae taxa: *S. chlorogaster* (0.0007), *P. sericata* (0.0055), *C. vicina* (0.0021), *M. domestica* (0.002) and *Sarcophaga* sp (0.001) showed very low values of total abundance. The remaining abundance values corresponded to specimens of Anthomyiidae and Acaliptratae.

All adult Diptera collected belonged to introduced

Table 3 - Male and female specimens of the Caliptratae and Acaliptratae sections collected during sampling weeks.

M F
tal Total
2
34 500
4 12
1 5
6 533
9 196
6 34
3
0 137
8
20 1,430
,

\*Significant difference for P<0.05; D.L.=1

M = Male; F = Female

taxa and corresponded to eusynanthropic or/and hemisynanthropic species. They could be also classified by their medical and veterinary importance, except for S. chlorogaster (Table 5).

## **DISCUSSION**

Six of the families recorded at the biosolids composting plant in Bariloche - Calliphoridae, Muscidae, Fanniidae, Sarcophagidae, Anthomyiidae and Syrphidae (Tables 2 and 5) - were the same as registered in a similar study performed in Poland.<sup>3</sup> Except for the calliphorids S. chlorogaster, P. sericata, C. vicina and Ophyra sp,8,15 this is the first time that all the other taxa have been cited in the northwestern Patagonian region.

Only one larva of Eristalis tenax was captured and no adults were observed, in spite of the facts that lixiviates are an appropriate substrate for such larvae and the pastures and woods surrounding the composting plant are a good source of nectar for the adults. All these characteristics and the previous records of human pseudomyiasis caused by this species in Bariloche<sup>4</sup> would have allowed the assumption that this species would be present more frequently at the composting plant, but our data suggested that adults might be visiting the biosolid windrows only sporadically.

Biosolid composting windrows constitute an appropriate location for fly feeding as well as for egg laying and the development of immature stages (3 larval stages and puparium). In treatment plants in Poland, the number of females found was significantly higher than the males,<sup>3</sup> and the same situation was registered at the Bariloche plant. These numbers of females and puparia, and Muscidae and Calliphoridae adults obtained in the laboratory, are indicative of the role of windrows in the life cycle of these flies.

The abundance of fly taxa could be related to their diet and environmental conditions, especially temperature and wind. Ophyra sp and C. macellaria, the most abundant Caliptratae taxa, decreased in abundance over the whole sampling period, in spite of variations in ambient temperature. For both species, other factors could be involved, such as larval predation by other Diptera and their own life cycle. In taxa such as M. stabulans and Fannia sp, the abundance was related to the temperature as has been found for the same taxocoenosis in other places in Argentina. 13 Likewise, at the Bariloche composting plant in the summer of 2000, the highest abundance in February was recorded for M. stabulans (highest temperature averages), and in March for Fannia sp (lowest temperature averages).

The richness found in the biosolids composting plant had intermediate values in comparison with those from sewage plants in Poland<sup>3</sup> and those from chicken and turkey dung. 13,16 The occasional presence of some taxa like Sarcophaga sp may explain the differences between total richness and monthly and composting week averages.

Flies associated with sewage and homeotherm vertebrate dung are potential vectors for taeniasis, oxyuriasis, trichuriasis and ascaridiasis.<sup>3,13,16</sup> For example, Calliphoridae related to dog dung contaminated with eggs of Taenia hydatigena can transport them to lambs.5 Eight of the nine adult taxa captured, were of

Table 4 - Total, monthly and composting week abundance values for the most frequent Diptera taxa.

Taxa	Total		Month		Composting weeks			
		Jan	Feb	Mar	1	' II °	III	
C. macellaria	0.1852	0.3138	0.2018	0.0232	0.2821	0.1792	0.0910	
Fannia sp	0.0749	0.0121	0.0706	0.1352	0.0863	0.1151	0.0248	
Ophyra sp	0.3454	0.5520	0.2977	0.1554	0.4086	0.2953	0.3278	
M. stabulans	0.0808	0.0424	0.1302	0.0626	0.0351	0.0706	0.1375	

Table 5 - Sanitary importance and synanthropic characteristics of collected Diptera.

	Sanitary i	mportance	Synanthropic characteristics			
Taxa	Medical '	' Veterinary	Eusynanthropic	Hemisynanthropic		
S. chlorogaster (Calliphoridae)	?	?	Χ	-		
C. macellaria (Calliphoridae)	yes	yes	-	X		
P. sericata (Calliphoridae)	yes	yes	X	X		
C. vicina (Calliphoridae)	yes	ves	X	X		
Ophyra sp (Muscidae)	yes	´-	X	X		
M. s'tabulans (Muscidae)	yes	-	X	-		
M. domestica (Muscidae)	yes	yes	X	X		
Fannia sp (Fannidae)	yes	´-	X	-		
Sarcophaga sp (Sarcophagidae)	ýes	yes	-	X		
E. tenax (Syrphidae)*	yes	yes	?	?		

<sup>\*</sup>One larva collected.

medical and veterinary importance because of their role as potential vectors of helminth and other pathogen infections.<sup>17</sup> This indicates the need for controlling Diptera populations during the spring and summer periods at the plant, as well as ensuring sanitary precautions for composting plant workers. In addition to this, Calliphoridae and Muscidae have flight radiuses that exceed 30 km per day and they can include wide areas in their daily movements, thereby spreading potential infections. The lack of an accurate taxonomic identification for Anthomyiidae and Acaliptratae species hindered the definition of their sanitary importance.

Many Diptera related to decaying organic matter could cause myiasis and pseudomyiasis to man and animals. Species of Diptera adults and/or larvae captured at the Bariloche plant have sanitary importance (Table 5). Phaenicia sericata and species of Cochliomyia such as C. hominivorax are obligatory or facultative parasites that cause epidermal myiasis in humans and domestic and wild animals. 12 E. tenax is another species that can produce pseudomyiasis, and even though only one larva was found, human cases have been reported in the Andean Patagonian region.4 Although investigations have indicated the importance of helminth egg distribution by flies,5 this role has not been corroborated either in composting plants or in sewage plants.3 Likewise, birds and other insects feed on Diptera adults and larvae; hence, they also constitute potential vectors for various diseases. At the biosolids composting plant, these potential vectors may be birds, like Tachycineta leucopyga (Hirundinidae) and *Polyborus chimango* (Falconidae), which were observed feeding on Diptera larvae and adults on or over the windrows.

Most of the captured Diptera were eusynanthropic and/or hemisynanthropic, with close or scarce contact with human and domestic animals, respectively.

The presence of these Diptera taxa at the biosolids composting plant indicates that its location has transformed a natural asynanthropic or hemisynanthropic site into a eusynanthropic one.

M. domestica, a species found extremely close to human activities, was found in low numbers at the Bariloche plant in spite of being the second in importance at sewage plants in Poland.3 With regard to the other captured species, data for comparing with Calliphoridae in a eusynanthropic habitat in Patagonia was only found for P. sericata and C. vicina.8,15 Alternating dominance between these species was recorded, a fact that possibly favors niche exclusion and avoids competition.8,15 In the composting plant, P. sericata was dominant over C. vicina, although an inverse relationship has previously been registered in the region.<sup>8,15</sup> Likewise, both species are poorly represented, probably due to niche competition with other numerically greater taxa. The microenvironmental conditions of the composting process could explain the differences in the numbers of M. domestica and the dominance relationship of C. vicina and P. sericata found in this study, in comparison with the results presented by other authors.8,15

The sanitary and epidemiological characteristics of most of the Diptera captured at the biosolids composting plant in Bariloche may be useful for defining locations for urban or semi-urban plants. They also highlight the importance of sanitary precautions at composting facilities.

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## **REFERENCES**

- Amorim JA, Ribeiro OB. Distinction among de puparia of 3 blow flies species (Diptera: Calliphoridae) frequently found on unburied corpses. *Mem Inst* Oswaldo Cruz 2001;96:781-4.
- Brinton WF. Volatile organic acids in compost: production and odorant aspects. Comp Sci Util 1998;6:75-82.
- Juriš P, Világiová I, Plachý P. Importance of flies (Diptera-Brachycera) in the distribution of helminth eggs from municipal sewage treatment plants. Vet Med Czech 1995;40:289-92.
- Kun M, Kreiter A, Semenas L. Myiasis gastrointestinal humana por Eristalis tenax. Rev Saúde Pública 1998;32:367-9.
- Lawson JR, Gemmell MA. The potential role of blowflies in the transmission of taeniid tapeworm eggs. *Parasitology* 1995;91:129-43.
- Mariluis JC. Clave para la identificación de los Calliphoridae de la República Argentina (Diptera). Rev Soc Ent Arg 1981;40:27-30.

- 7. Mariluis JC, Lagar MC, Bellegarde EJ. Diseminación de enteroparásitos por Calliphoridae (Insecta, Diptera). *Mem Inst Oswaldo Cruz* 1989;84:349-51.
- 8. Mariluis JC, Schnack JA. Elenco específico y aspectos ecológicos de Calliphoridae (Insecta, Diptera) de San Carlos de Bariloche, Argentina. *Bol R Soc Esp Hist Nat (Sec Biol)* 1996;92:203-13.
- Mazzarino MJ, Laos F. Composting biosolids in Patagonia. *BioCycle* 2000;41:83-5.
- Nuorteva P. Synanthropy of Blowflies (Diptera, Calliphoridae) in Finland. Ann Entomol Fenn 1963;29:1-49.
- 11. Oliva A. Insectos de interés forense de Buenos Aires (Argentina). Primera lista ilustrada y datos bionómicos. Rev Mus Arg C Nat "Bernardino Rivadavia". Entomología 1997;7:13-59.
- 12. Otranto D. The immunology of myiasis: parasite survival and host defense strategies. *Tr Parasitol* 2001;17:178-82.

- Perotti A. Moscas sinantrópicas (Diptera: Muscidae y Fanniidae) asociadas a producciones avícolas del Centro-Sudeste bonaerense. Nat Neotrop 1998;29:145-54.
- 14. Robinson WH. *Insect and mite pests in the human environment*. In: Robinson, WH. Urban Entomology. London: Chapman and Hall; 1996. p. 285-320.
- Schnack JA, Mariluis JC, Spinelli GR, Muzón J. Ecological aspects of urban blowflies in midwest Argentinian Patagonia (Diptera: Calliphoridae). Rev Soc Entom Arg 1998;57:127-30.
- Szelagiewicz M, Sokol R, Spodniewska A. Participation of insects in spreading parasitic invasion in guess reared in a traditional way. *Nat Sci* 1998;1:71-9.
- 17. World Health Organization (WHO). Moscas de importancia para la salud pública y su control. Washington (DC); 1962. (Publ. Cient. 61).