Universidade Federal do Rio de Janeiro
Instituto de Biodiversidade e Sustentabilidade
Programa de Pós-Graduação em Ciências Ambientais e Conservação

Juan David Rojas Arias

Effect of pipelines on landscape connectivity for long-furred woolly opossum (*Marmosa paraguayana*) in Atlantic Forest

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Dissertação de Mestrado apresentada ao Programa de Pós-Graduação em Ciências Ambientais e Conservação, Universidade Federal do Rio de Janeiro, como requisito para obtenção do título de Mestre em Ciências Ambientais e Conservação.

Candidate: Juan David Rojas Arias

Advisors: Dr. Caryne Aparecida de Carvalho Braga

Co-Advisor: Dr. Pablo Rodrigues Gonçalves

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Appreciating the land is the only true hope of survival.

-Anonymous-

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Effect of pipelines on landscape connectivity for long-furred woolly opossum (*Marmosa paraguayana)* in the Atlantic Forest

ABSTRACT

The Atlantic Forest is one of the five most important hot spot for the conservation of biodiversity in the world, harboring more than a million species of animals. Today, the Atlantic Forest is reduced to less than 30% of its original area, and highly fragmented due to anthropic interventions. One of the most important causes of fragmentation is the construction of linear infrastructures like roads, pipelines, and electrical networks that generates deforestation in the continuous forest. In this dissertation, we analyzed the effects of a linear pipeline on the movements of *Marmosa paraguayana*, an arboreal marsupial of the Atlantic Forest. Movements between transects in the interior and the edge of the fragments were recorded through capture, mark, and recapture method. We monitored movements in three areas in the Biological Reserve União, and three in the APA Bacia do Rio São João, State of Rio de Janeiro, Brazil. We compared frequencies of movements within the forest and across the pipeline to evaluate if the pipeline act as a barrier for *M. paraguayana*. Also, distances of movements were measured and compared between areas and sexes. The capture success was higher on the side of the pipeline with smaller forest fragments. There were more captures of *M. paraguayana* at the interior of the forest than on the edge, suggesting an edge effect. Pipeline crossings were significantly less frequent than movements inside the forest indicating that the pipeline acts as a barrier for the movements of *M. paraguayana*. All pipeline crossings occurred during the mating season. Both sexes cross the pipelines, but males tend to do more crossings and tend to travel greater distances than females. This study revealed an underestimated effect of pipelines on forest connectivity and the urgency to elaborate measures to mitigate these impacts for the species conservation.

Keywords: Linear infrastructure, Barrier, connectivity, conservation, crossing, Fragmentation, marsupial, movements.

RESUMO

A Mata Atlântica é um dos cinco hot spot mais importantes para a conservação da biodiversidade do mundo, abrigando mais de um milhão de espécies de animais. Hoje, a Mata Atlântica está reduzida a menos de 30% de sua área original e altamente fragmentada devido às intervenções antrópicas. Uma das causas mais importantes da fragmentação é a construção de infraestruturas lineares como estradas, dutos e redes elétricas que geram desmatamento na floresta contínua. Nesta dissertação, analisamos os efeitos de uma faixa de dutos nos movimentos de *Marmosa paraguayana*, um marsupial arbóreo endêmico da Mata Atlântica. Os movimentos foram analisados através do método de captura, marcação e recaptura. Os animais foram capturados em armadilhas distribuídas em quatro transectos paralelos, dois localizados na borda do oleoduto e dois no interior da floresta, este desenho amostral foi replicado em seis áreas em duas unidades de conservação da bacia do rio São João, estado do Rio de Janeiro, Brasil. As taxas de movimento através do gasoduto e dentro da floresta foram medidas e comparadas. Além disso, distâncias de movimentos foram medidas e comparadas entre áreas e sexos. Os resultados mostraram travessias de dutos menores do que movimentos dentro da floresta, indicando que o duto atua como uma barreira para os movimentos de *M. paraguayana*. Além disso, houve mais capturas de *M. paraguayana* no interior da floresta do que na borda, indicando um efeito de borda. Observamos um melhor sucesso de captura no menor fragmento de floresta e notamos uma influência da encosta de cada área nas distâncias percorridas por *M. paraguayana*. Verificamos que os machos de *M. paraguayana* tendem a percorrer distâncias maiores que as fêmeas e que a maioria dos cruzamentos entre os fragmentos ocorre durante a época de acasalamento. Este estudo revelou os efeitos subestimado produzidos por um duto, na conectividade florestal e a urgência da elaboração de medidas para mitigar esses impactos para a conservação das espécies.

Palavras-chave: Mata Atlântica, Barreira, conectividade, conservação, travessia, Fragmentação, marsupial, movimento

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INTRODUCTION

Tropical forests are essential ecosystems for the survival of many animals, plants, and humans, hosting most of the species on the planet, and providing uncountable services for human well-being (PEARCE, 2001; WRIGHT, 2005). Therefore, the preservation of these ecosystems is important for the conservation of biodiversity and a sustainable future (RIBEIRO et al. , 2009). Nevertheless, natural ecosystems without any anthropic intervention are increasingly rare (ELLIS; RAMANKUTTY, 2008). Human population growth leads to the colonization of natural areas and requires many anthropic interventions. Nowadays, more than 60% of the earth's land surface has been modified by humans, and only 22% of ice-free land area remains as wildlands without human intervention (BARNOSKY et al. , 2012; ELLIS et al. , 2010).

A recurrent and conspicuous type of anthropic intervention is the construction of linear infrastructures, such as roads, electric transmission lines, and fuel pipelines (gas and oil), which are an integral part of the economic and social development of a country (WILKIE et al. , 2000). Although these structures have a large impact on the environment (LAURANCE; GOOSEM; LAURANCE, 2009), in many cases they are necessary for the economic, cultural, and political development of human societies. Therefore, it is imperative to find a way to optimally minimize their impacts on biodiversity, assuming that anthropic activities cannot be excluded from conservation planning (ELLIS; RAMANKUTTY, 2008).

Linear infrastructures can adversely affect forest biodiversity since they contribute to habitat loss and fragmentation, besides other direct and indirect impacts on animal mortality (LAURANCE; GOOSEM; LAURANCE, 2009). Most studies evaluating impacts of linear infrastructures are focused exclusively on roads (LACERDA; TOMAS; MARINHO-FILHO, 2009; LAMBERT et al. , 2014; ZUNINO et al. , 2007). However, roads include one important specific impact, the roadkill, which is not present in other linear infrastructures. Little is known about the impacts of narrow corridors of matrix crossing forests aside from the roadkill. The removal of the trees, for instance, may affect the movements of terrestrial and arboreal species in different ways (SAUNDERS; HOBBS; MARGULES, 1991). For terrestrial animal species, changes in vegetation structure may cause problems of orientation or higher vulnerability to predators (BROWN; KOTLER, 2004; RÍOS-UZEDA; BRIGATTI; VIEIRA, 2019). For arboreal animals, tree removal implies the loss of canopy connectivity for dispersal, therefore limiting individuals to tree-covered patches and restricting their movements (CAREY, 1996). Indirect impacts are related to edge effects that could be positive or negative, depending on the species (PARDINI, 2004). Changes in the microclimate (temperature, relative humidity, speed of wind among others) in the forest edge bordering the deforested area may affect food availability and diversity, which might alter the abundance and distribution of animal species (STEVENS; HUSBAND, 1998). Ultimately, the potential isolation could also disrupt demographic and genetic interchanges among populations that inhabit forest fragments separated by the deforested corridor, likely increasing the vulnerability of populations to local extinctions (HANSKI, 2011).

On a populational level, evidence points to several detrimental effects of fragmentation on effects size and genetic variation of populations. Almeida-Rocha et al. (2020) conducted a meta-analysis of 61 studies investigating genetic responses of species to fragmentation and concluded that habitat loss and decreasing connectivity among populations are the most detrimental factors to population size and genetic diversity of animals and plants. In this context, the restriction of animal movements due to the creation of barriers among forest fragments are limiting factors for the preservation of the ecosystem functions and even for the survival of the species (TUCKER et al. , 2018). However, species responses to habitat fragmentation may vary based on how generalist they are in terms of movement capacity among forest patches (HANSKI, 2011). When a continuous forest is fragmented, forest specialists tend to avoid crossing open areas between fragments, isolating their populations, and increasing inbreeding (PIRES et al. , 2002). In the long term, the viability of the species is compromised due to the random loss of beneficial mutations and the accumulation of harmful mutations (HANSKI, 2011). Species that can at least occasionally cross open habitats are prone to form a metapopulation dynamic in fragmented habitats, allowing genetic exchange between patches and the colonization of unoccupied patches, decreasing the likelihood of local extinctions (HANSKI, 2011).

Among the several processes involved in the creation of barriers among forest patches, the construction of underground pipelines for the transportation of raw materials, such as oil or gas, deserves special attention. It is particularly important for the energy and industrial sectors, especially in countries that are still highly dependent on fossil fuels. Brazil, for example, already has an extensive network of structures of this type and is investing in its expansion in the near future. Therefore, we may foresee further fragmentation of forests, clearings, edges, and other potential problems related to anthropogenic actions such as the presence of hunters, the passage of vehicles, and changes in the use of the matrices surrounding the forest (GRANGEIA, 2008; LAURANCE; GOOSEM; LAURANCE, 2009). Despite their potential impacts on biodiversity, gas, and oil pipelines have been poorly studied concerning their impacts on movements and population connectivity of tropical forest mammal species (LAURANCE; GOOSEM; LAURANCE, 2009; LUCAS et al. , 2019; RUIZ-MIRANDA et al. , 2018). Even though pipelines are normally buried into the ground, the arboreal vegetation is systematically removed and managed to be kept low to avoid damages to the duct (e. g. growing roots of trees), and therefore preventing oil spills or gas leaks (VASEK; JOHNSON; ESLINGER, 1975). Usually, the area where the vegetation is removed forms an open corridor of around 20-30 m wide between the two forest fragments (Figure 1).

Animal responses to these corridors may vary depending on the species traits, such as body size, locomotion habit, diet, and habitat use (FAHRIG, 2017). For example, given the absence of trees, small mammal species that use the arboreal strata may not be able to cross the open corridor. The diet and the perception of risk to predators could also be modified by the pipeline corridor, since the open area generates edges that change the availability of food resources, the structure of vegetation cover, and the micro-climatical conditions, among other factors (FAHRIG, 2007). Some arboreal species are less agile on the forest floor, which makes them easy prey to local predators in open areas (VAN HELDEN et al., 2020). Finally, some species prefer to shelter under tree cover and avoid open areas (BROWN; KOTLER, 2004). Many species depend exclusively on the existence of the canopy for foraging, reproduction, and locomotion (DUBLIN, 1903). Therefore, areas without arboreal vegetation may act as barriers to the passage of these animals. Carey

(1996) attributed the barrier effect to the level of “arboreality” of rodents, assuming that this effect is smaller in animals that constantly dwell on the ground.

Most of the studies that involve pipelines have focused on the effects of their installation and management on biodiversity (AGBAGWA; NDUKWU, 2014; GREGORY et al. , 2017; LAMBERT et al. , 2014; PACHECO et al. , 2013; RICHARDSON et al. , 2017), and only a few on the effects of forest fragmentation on small mammal populations (DARLING; LESTON; BAYNE, 2019; HANSKI, 2011; PIRES; FERNANDEZ, 1999). On a review about the impacts of pipelines and power lines on biodiversity, Richardson et al. (2017) found only 29 studies evaluating the impacts of pipelines on animal and plants populations; most studies focused on plants (35%) and medium/large mammals (32%). Although there are few studies on the responses of animals to the presence of the pipeline, Kwast-Kotlarek et al. (2019) warned about the dangers the construction of the pipeline offers for the migration of amphibians and reptiles, and about the noise generated by the machines that affect bird communities. Richardson et al. (2017) highlighted that the long-term effects after the construction of the pipeline will depend on the environmental requirements of each species. It is expected that species will avoid crossing these corridors, especially the arboreal species (FORERO MEDINA; VIEIRA, 2009).

Figure 1. Open corridor resulting from the installation of an underground pipeline.

Measuring species migration rates between and within fragments allows determining the connectivity of a landscape for a species (PARDINI et al. , 2010). Movement rates calculation includes quantifiable measures of movement between fragments through open areas and is an important tool for conservation because it allows determining how vulnerable the species are to local extinction (PIRES et al. , 2002). Besides, these rates are also useful to determine the structure of populations that interact between fragments, since species that have higher movement rates between forest fragments could be considered as a single population that shares genes, while for species with low movement rates, separated populations tend to be present in each fragment with little gene sharing (PIRES et al. , 2002). Spatially fragmented populations generate the greatest concern in terms of conservation since isolation can lead to local extinction (HANSKI, 2011).

Animal movements can determine the structure of populations and communities (MUELLER; FAGAN, 2008; NATHAN et al. , 2008). Thus, studies of how the structure of landscapes impacts animal movements are important to understand their natural history and ecology. Fahrig (2007) also pointed out that dispersal ability is not simply an attribute of a species, but it varies strongly with landscape structure. In this context, she suggests that populations that have inhabited landscapes with high habitat cover should have evolved low boundary responses and moderate to high movement probabilities. Nonetheless, these populations are expected to be highly susceptible to increased movement mortality resulting from habitat loss and reduced matrix quality. This condition is probably similar to what is expected with a pipeline corridor crossing a continuous forest.

Animal movements can also be affected by climatic factors such as the season, temperature humidity among others, and by aspects of their biology such as gender, age, body size, and trophic guild (DELCIELLOS et al. , 2019; PIRES; FERNANDEZ, 1999; SANDERSON, 2013; SWIHART; SLADE; BERGSTROM, 1988), These differences are important to determine the level of impact that the modification of the ecosystem has on the movements of the species. Swithart, Slade, & Bergstrom (1988) suggested that the area occupied by 23 species of terrestrial mammals was related directly to their body mass where bigger mammals tend to move and occupy greater areas than the smaller ones. Delciellos et al. , (2019), founded that the tortuosity of the movements of *Philander quica* was influenced by population density, sex, and climatic season.

The impact of the climatic season is usually linked to resource availability and reproductive activity. In periods of high resource abundance, it would be easier to find resources in a small geographic area, leading to shorter movements or smaller home ranges compared to seasons of reduced resource availability (Winker et al. 1995). Small mammal reproduction, especially for neotropical marsupials, tends to vary seasonally (LAFERRIERE; ATRAMENTOWICZ, 1990). Male and female behaviors are different during the reproductive season in polygynous species, which can lead to differences in movement distance. Males tend to have longer movements, attempting to mate with the largest number of females (GENTILE et al. , 2000; HINGST et al. , 1998; LORETTO; VIEIRA, 2005). The movements of *M. paraguayana*, the model species in this study, also seem to be influenced by sex and reproductive status; males tend to travel longer distances looking for a partner, while females have more territorial behavior (PIRES; FERNANDEZ, 1999).

Marmosa paraguayana is a scansorial marsupial, relatively abundant on the Atlantic Forest at Rio de Janeiro state, and has been the focus of movement studies (MORAES JUNIOR; CHIARELLO, 2005; PIRES et al. , 2002). This omnivorous species plays an important role in preserve forest functions through activities such as seed dispersal and pest control (SMITH, 2009). Thus, the main objective of this dissertation is to evaluate the impacts of the deforested corridor produced by the installation of a pipeline on the movements of *M. paraguayana.* Additionally, we are interested in assessing the effect of sex, reproduction, and environmental conditions on the movement distance.

The work hypotheses are:

1- Pipelines act as a barrier to the movement of small scansorial species. If this hypothesis holds, we expect to find higher frequency of movements of *M. paraguayana* within than between forest fragments crossed by the pipeline corridor.
2- The deforested corridor above the pipeline causes an edge effect that alters the habitat use of small scansorial species. If this is true, we expect differences in the number of captures of *M. paraguayana* between the edge and the interior of the forest.
3- Since size and habitat quality of forest fragments are positively related, larger fragments support larger populations of scansorial species. We expect that the higher densities of *M. paraguayana* result in a higher capture success in the larger fragments.
4- Since sex and reproductive stage affect displacement distances of several scansorial species, we expect that males of *M. paraguayana* will display longer distances of movements than females during the reproductive season.

MATERIALS AND METHODS

Studied species – We selected *Marmosa paraguayana* (Tate, 1931) as our study species. Several studies on this didelphid marsupial (Didelphimorphia: Didelphidae) (GARDNER, 2008) have been carried out evaluating the effects of forest fragmentation and others anthropogenic interventions on its populations in the Atlantic Forest (PIRES; FERNANDEZ, 1999; PIRES et al. , 2002; RUIZ-MIRANDA et al. , 2018). *Marmosa paraguayana* has a moderate degree of tolerance towards anthropogenic intervention (SMITH, 2009) and is abundant in our study area (RUIZ-MIRANDA et al. , 2019). Also, it is considered as “least concern” (LC) worldwide (BRITO et al. , 2018), but its population is decreasing drastically, due to habitat loss in the Atlantic Forest (HONORATO et al. , 2015). This species presents a total length that could reach 46 cm and a maximum weight of 230 g, being considered a large size when compared to other species of the Marmosini tribe (SMITH, 2009). It is distributed in Paraguay, Argentina, and eastern Brazil, from Minas Gerais to Rio Grande do Sul. The species is more abundant in dense forests, abundant in palms and vines (SMITH, 2009). The locomotor habitat is classified as scansorial and arboreal by different authors (HONORATO et al., 2015; PAGLIA et al., 2011; WILSON et al., 2015). It can breed in both primary and secondary forests (SMITH, 2009). Regarding the species diet, it is considered an opportunistic generalist species, ingesting mostly fruits and seeds (SANTORI; LESSA; ASTÚA, 2012). The reproductive cycle of *M. paraguayana* is seasonal, and the breeding season corresponds to the months with greater availability of resources (PIRES; FERNANDEZ, 1999). *Marmosa paraguayana* presented an intermediate condition, with low rates of inter-fragment movement, supposedly allowing gene flow, and thus genetic connectivity among the populations on different fragments, according to metapopulation dynamics at the Atlantic forest (PIRES et al., 2002).

Study areas- The six study areas are embedded within the São João river basin and all of them contain several types of linear structures (roads, power lines, and gas and oil pipelines) that cross and fragment the landscape (Figure 2). The regional climate is characterized by a wet season, from September to April, and the dry season, from May to August (ALVARES et al. , 2013).

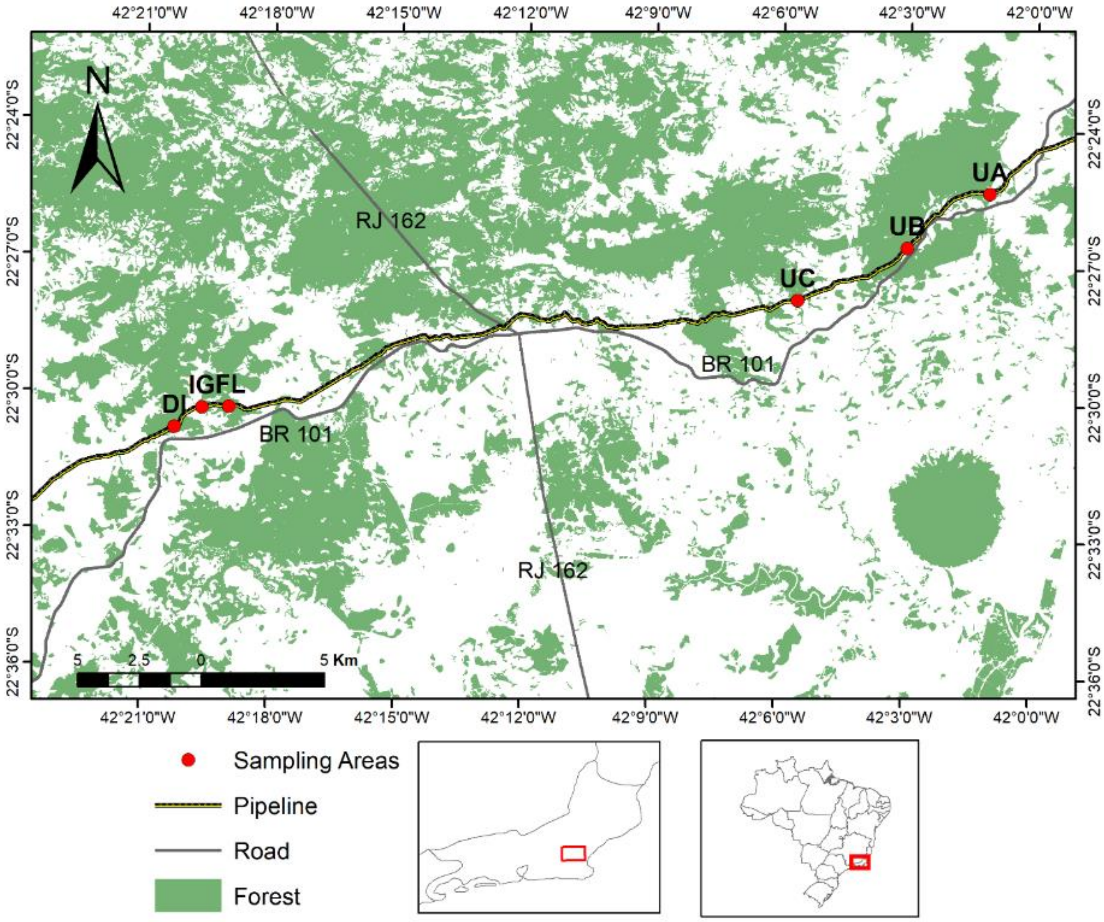

Figure 2. Sampling áreas in the São João River Basin, Rio de Janeiro, Brazil. The red points represent the sampling areas: Fazenda Dois Irmãos (DI), Fazenda Iguapê (IG), fazenda Flandria (FL), REBIO União A (UA), REBIO União B (UB), REBIO União C (UC).

The São João River basin has 3,000 km^2, encompassing eight municipalities and the main vegetation type is Submontane Rainforest (LUCAS et al., 2019). Three of the six sampling areas are located in a sustainable use protected area, the Environmental Protected Area of Rio São João (hereafter called APA): Fazenda Flandria (FL) (-22. 505250°, -42. 316260°), Fazenda Iguapê (IG) (-22. 505720°, -42. 326960°) and Fazenda Dois Irmãos (DI) (-22. 512860°, -42. 337680°) (Figure 3). The APA was instituted in 2002 and has an extension of 150,700 ha, located in the municipalities of Silva Jardim and Casimiro de Abreu. About 50% of the area is covered by altered forest, predominantly on scarp or on hilltops. The plains are more frequently used for agriculture and livestock (LUCAS et al. , 2019).

The other three areas are located within a strictly protected area, the União Biological Reserve (REBIO União): REBIO União A (UA) (-22. 422637°, -42. 018349°), REBIO União B (UB) (-22. 443004°, -42. 050378°) are natural conserved ecosystems and REBIO União C (UC) (-22. 462706°, -42. 093187°). This reserve has more than 7,000 ha of preserved forest, but it is crossed by linear infrastructures such as highways, electrical networks, and pipelines (LUCAS et al. , 2019). REBIO União C is located in an area recently added to the protected area 2017 (LUCAS et al. , 2019). Since the area near UC has been actively used for farming purposes, it has a higher degree of human intervention compared to UA and UB. The forest connectivity is higher at the north side of the duct in all three areas, and the fragments are larger (Figure 2). It is important to highlight that the north side of the areas UA and UB are in the same fragment, but they are treated as independent since the distance between UA and UB is larger than the life range size of the species.

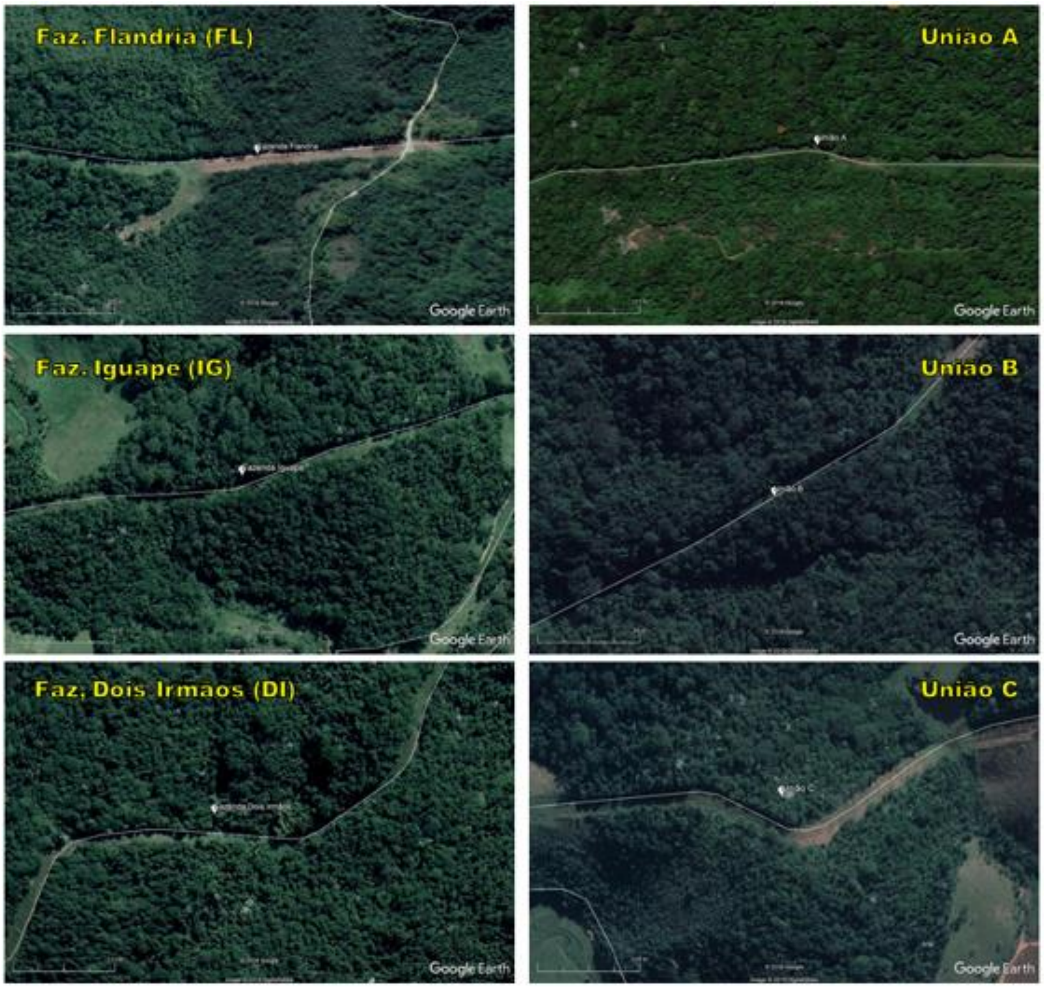

Figure 3. Sampling areas. Satellite imagery (Digital Globe®) of the six small mammal sample areas along the pipeline (GASDUC II) that section forest remnants in the São João River Environmental Protection Area and the REBIO União Biological Reserve.

Sampling design- We positioned live traps along four parallel linear transects at each area. Two transects were placed at the edge of the forest, one on each side of the pipeline and the other two were placed in the forest interior, at a distance equivalent to the width of the pipeline (20 to 30 m), parallel to the edge transects (Figure 4). Each transect was composed of 15 stations, 10 m apart from each other, with a Sherman® (30 cm x 8 cm x 9 cm) and a Tomahawk® (45 cm x 16 6 cm x 16 cm) trap (according to Voss & Emmons, 1996), totaling 120 traps per area. On each station, we installed one trap on the ground and one on the understory, 1-2 meters in height, and tied it to branches (Figure 5). We alternated the types of traps between the ground and the understory at every station. Each trap was baited with a mixture of banana (*Musa* spp.), sardine, cornmeal, and peanut powder.

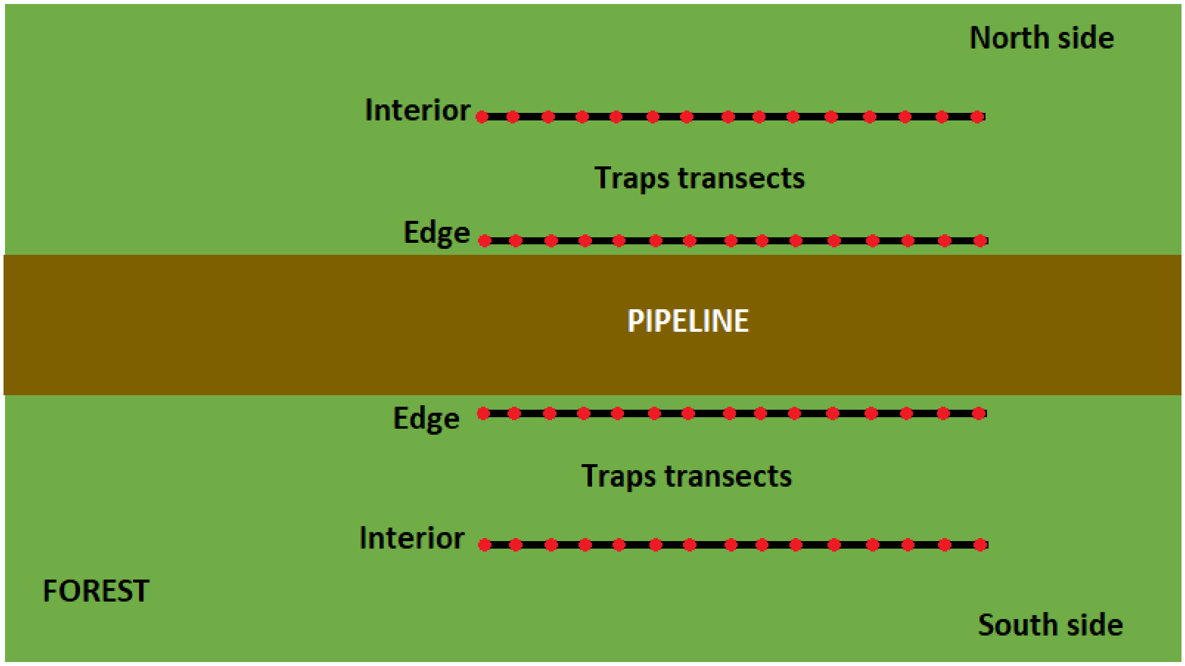

Figure 4. Scheme of transects at each sampling area. Each transect consists of 15 stations. Image obtained and modified from Ruiz-Miranda et al (2018).

Figure 5. A. Tomahawk trap set on the ground with a captured individual (*Metachirus myosurus*). **B.** Sherman trap set on the understory.

We carried out samplings quarterly, from October-2018 to December-2019. Each sampling campaign lasted seven days. REBIO União and APA areas were sampled separately. APA was sampled in October-2018, February-2019, May-2019, August-2019, and December-2019, while the REBIO União was sampled in January-2019, April-2019, July-2019, and

October-2019. The sampling effort was 840 traps/nights in each area per campaign, totaling an effort of 10,080 traps/nights for the REBIO União and 12,600 traps/nights for the APA.

Animal handling- Captured individuals were carefully immobilized and handled. For each individual, we measured the body length (BL), tail length (CL), ear length (EL), and weight, in grams. We also sexed and aged the animals according to Macedo et al. (2006), inspected signs of reproductive activity (lactating mammae or suckling young), collected samples of ear skin, dorsal hairs, and feces for molecular analyses, and then marked with numbered ear tags (Figure 6; National Band Tag Inc. number 1), following the ASM guidelines (SIKES, 2016). Immediately after marking the individual, we released it in the same station where it was captured. This method allowed us to use the capture-recapture history of an individual as information of its movements. If an animal was captured at a different station than its previous capture, the linear distance between successive capture-stations was considered as its minimum movement distance. The study was authorized by the license No. 64807-2 issued by The Institute for Conservation of Biodiversity "Chico Mendes" (ICMBio) and the Ministry of the Environment of Brazil (MMA).

Figure 6. Specimen of *Marmosa paraguayana*, captured and marked with numbered earrings. Photograph obtained from Ruiz-Miranda et al (2018).

Data analysis

Edge effect- We expect that the frequency of habitat use will vary if the edge effects caused by the pipelines changed in the important habitat variables for *M. paraguayana*. We considered the number of captures as a surrogate for the frequency of habitat use. Therefore, we built a generalized linear mixed model (GLMM) in which the number of captures was the dependent variable, the capture position (two levels: edge or interior) the explanatory variable, and “area” and “sampling session” as random effects. A Poisson distribution was assumed for the dependent variable. Then, we use Akaike’s information criteria for small sample sizes (AICc) to compare this model to a null model including only the intercept and the random effects. The candidate model with the explanatory variable was selected whenever its respective delta Akaike (ΔAICc = AICc*i* – minimum AICc), the difference between ranked models, was smaller than 2 and smaller than the null model.

Effect of fragment size - We expect that the number of captures of *M. paraguayana* will be higher in transects at the north of the pipeline compared to those at the south. This hypothesis is based on the larger size of the forest fragments north of the duct and their higher connectivity to other forest fragments (Figure 2). Therefore, we built a generalized linear mixed model (GLMM) in which the number of captures was taken as a dependent variable, the capture side (two levels: north or south) as the explanatory variable, and “area” and “sampling session” as random effects. A Poisson distribution was assumed for the dependent variable. Then, we use Akaike’s criteria to compare this model to a null model including only the random effects. The candidate model with the explanatory variable was selected whenever its respective delta Akaike (ΔAICc) was smaller than 2 and smaller than the null model.

Distances of movement and population parameters- We inferred the movements and the distances traveled by each individual based on its capture-recapture history, and the coordinates of the trap-stations where the animal was successively captured. We tested if there are differences in the distances traveled by male and female individuals of *M. payaguayana* with a t-test. For this analysis, we included only individuals in the age class 4 (adults), since they represented 65.3% of the captures, and comparisons including other age classes would include ontogenetic variation in size and behavior that could affect movement distance.

Spatial and temporal variation in the movement distances- To explore if unaccounted differences between the studied areas affect the distances traveled by *M. paraguayana*, we carried out a Kruskal-Wallis test where the distances of movements were taken as a dependent variable and the study areas as predictive variables. We determined if there were significant differences (α=0. 05) between the distances traveled in each area. Movement distances can also be affected by weather conditions. Therefore, we conducted a t-student test to evaluate the variation of movement distances in function of the climate season of the recaptures (wet, dry). For this analysis, we considered the distances of movement as a dependent variable and the seasons (two levels: wet or dry) as explanatory variables.

Movements- We classified the recorded movements into four categories:

1- Non-movement (NM): recapture in the same station as the previous capture.
2- Transect movements (TM) – recapture at a different station along the same transect of the previous capture.
3- Forest crossings (FC) – recapture on the same side of the forest, but in a different transect (edge or interior) of the previous one.
4- Pipeline crossings (PC) – recapture occurred on a different side of the pipeline than the previous capture.

To evaluate if the pipeline act as a barrier to movement for *M. paraguayana*, we compared the number of PC and FC through a Chi-square test (χ^2) (α=0. 05). PC is expected to be lower than FC if the pipeline is a barrier.

To obtain the rate of pipeline crossings (PCr), we divided the total number of pipeline crossings by the sum of forest and pipeline crossings (PCr=PC/(PC+FC)). To obtain the rate of forest crossings (FCr), the same procedure was carried out, but dividing the FC by the sum of forest and pipeline crossings (FCr=FC/(PC+FC)).

To determine whether factors such as sex or climatic season affect the number of pipeline crosses of *M. paraguayana*, we built a generalized linear mixed model (GLMM) in which the values of PC were used as the dependent variable, and the variables season, sex, and the interaction between both were the explanatory variables, the "area" was used as a random effect. The null model included only the random effect. We selected models whenever its respective delta Akaike (ΔAICc) was smaller than two and different from the null model.

RESULTS

We captured a total of 72 individuals, being 38 males and 34 females, 185 times. In terms of age, we registered 47 adults in the age class 4 (65.3%) and 25 young (34.7%) with lower age classes. In the APA Bacia do Rio São João we registered 27 individuals, 15 males (55.5%) and 12 females (44.4%), and 37 recaptures of individuals in this area. In the REBIO União, we recorded 45 individuals, 23 males (51.1%) and 22 females (48.9%) and made 76 recaptures. We also observed a variation of the number of captures of males and females throughout the year, capturing more females than males in the first semester (F=22 M= 14), with an increase in the frequency of males in the second semester (F=31, M=36) (Figure 7).

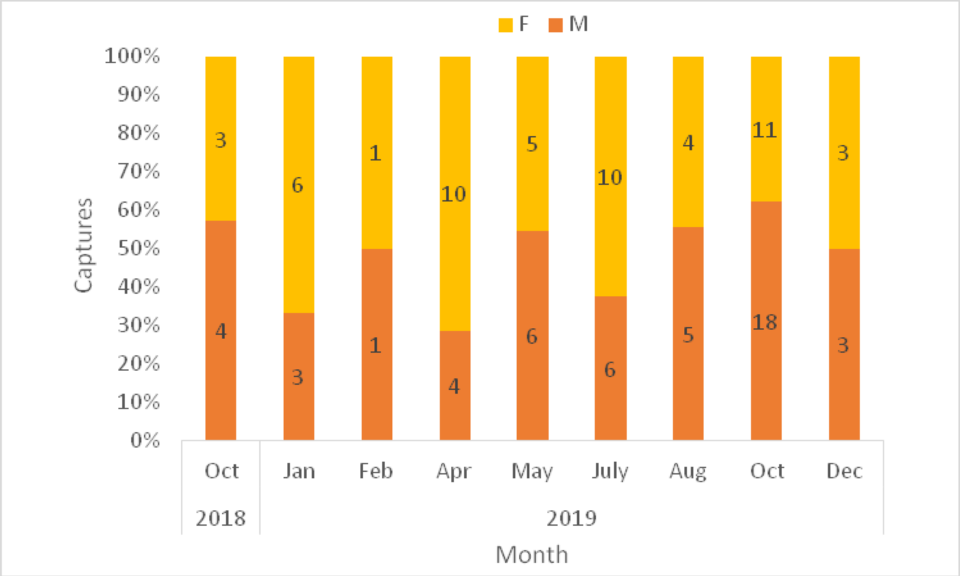

Figure 7. Percentage of captures of males (M) and females (F) of *Marmosa paraguayana* over time in the six study areas within the Rio São João basin, State of Rio de Janeiro, Brazil.

We recorded six reproductive females that were recaptured 27 times, four of those were registered at least on one occasion carrying their young while the other two were lactating. On all the females with young, we registered 11 cubs. Reproductive individuals were more frequent in the campaigns between October and December (Figure 8).

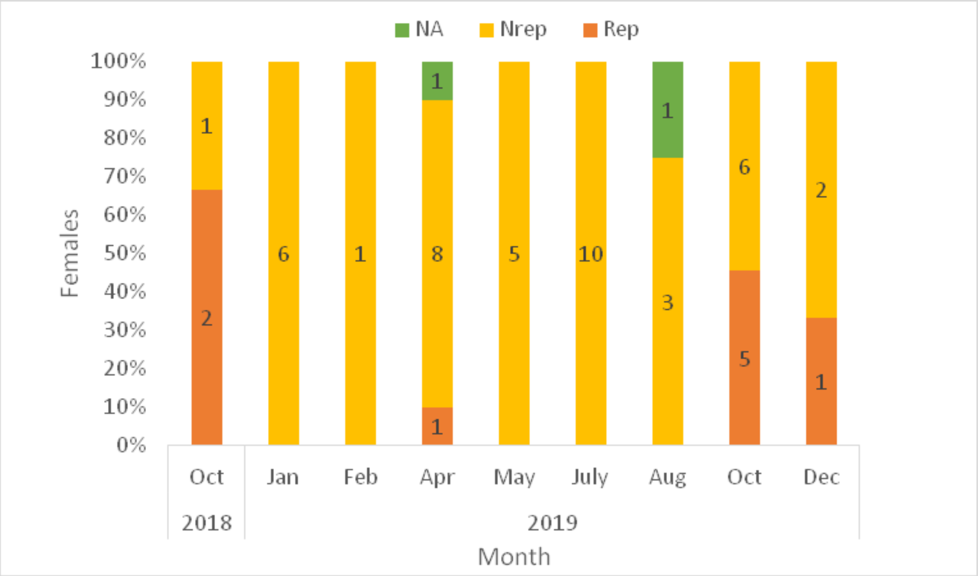

Figure 8. Number of reproductive (Rep), Non-reproductive (Nrep), and without information (NA) females captured in all areas studied at forest fragments of the Rio São João basin, State of Rio de Janeiro, Brazil.

Edge effect - The position (edge or interior) influences the number of captures of *M. paraguayana* (Table 1), as the best-fit model included this variable. A higher number of captures was found in the forest interior than in the edge (Figure 9).

Table 1. Model selection analysis of position (edge/interior), affecting the number of captures of *Marmosa paraguayana*. Pos: Position, R^2: determination coefficient r, d. f: degrees of freedom, logLik: log-likelihood value, AICc: Akaike information criterion, ΔAICc: difference of AIC compared to the best model.

	Intercept	**Pos**	**R^2**	**d. f.**	**logLik**	**AICc**	**ΔAICc**	**weight**
2	0. 78	+	0. 75	4	-113. 31	235. 44	0	0. 75
1	0. 95	NA	0. 73	3	-115. 59	237. 67	2. 23	0. 25

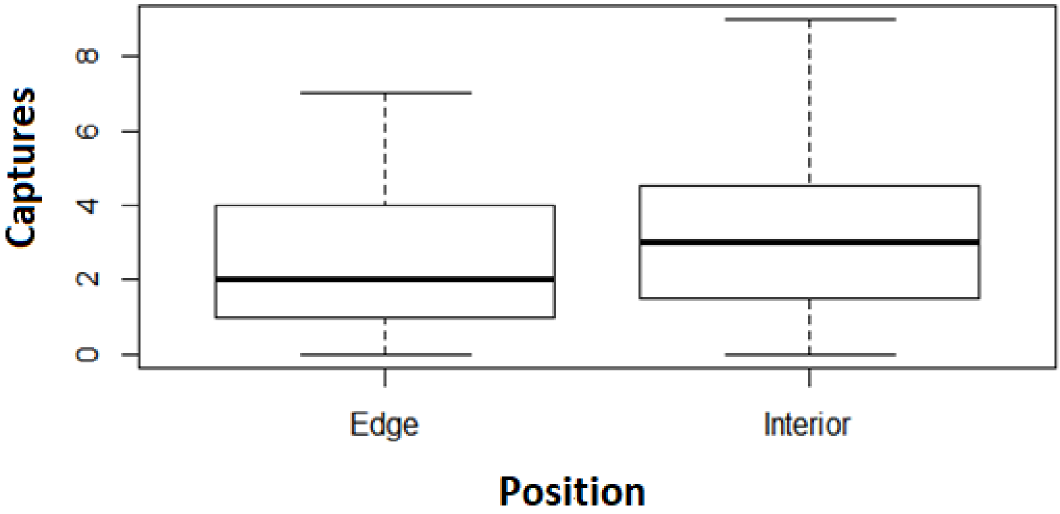

Figure 9. Variation in the number of captured individuals between the edge and interior of the forest fragment. The boxes represent the percentiles of the captures, the black lines within the box represent the median and the whiskers represent the minimum and maximum value of captures.

Effect of fragment size - We also found differences between the number of captures on each side of the pipeline. The model selection analysis indicated that the side (North or South) of the pipeline corridor significantly affected the number of captures of *M. paraguayana* (Table 2). We recorded a higher number of captures on the south side of the pipeline corridor (Figure 10).

Table 2. Model selection analysis of side (north or south) affecting the number of captures of *Marmosa paraguayana* Side: north or south, R^2: determination coefficient r, d. f: degrees of freedom, logLik: log-likelihood value, AICc: Akaike information criterion, ΔA ΔAICc: difference of AIC compared to the best model.

	(Intercept)	**Side**	**R^2**	**df**	**logLik**	**AICc**	**ΔAICc**	**weight**
2	0. 64	+	0. 78	4	-139. 18	287. 18	0	0. 99
1	0. 93	NA	0. 72	3	-144. 84	296. 15	8. 97	0. 01

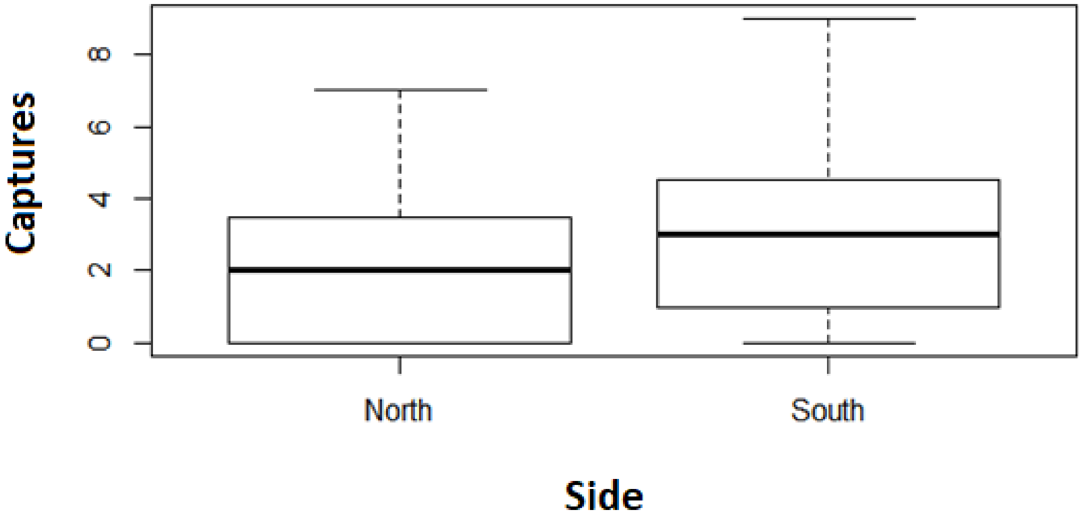

Figure 10. Variation in the number of captured individuals between the north and south side of the forest fragment relative to the pipeline. The boxes represent the percentiles of the captures, the black line within the box the median, and whiskers the minimum and maximum value of captures.

Spatial and temporal variation in the movement distances- There were differences in the movement distances among areas, with longer distances being recorded in “Fazenda Flandria” (H (5, N= 91) =15. 94; p =0. 007) (Figure 11A). There was no significant variation in movement distance between climatic seasons (F =1. 30; D. F. = 1;94; P= 0. 26) (figure 11B).

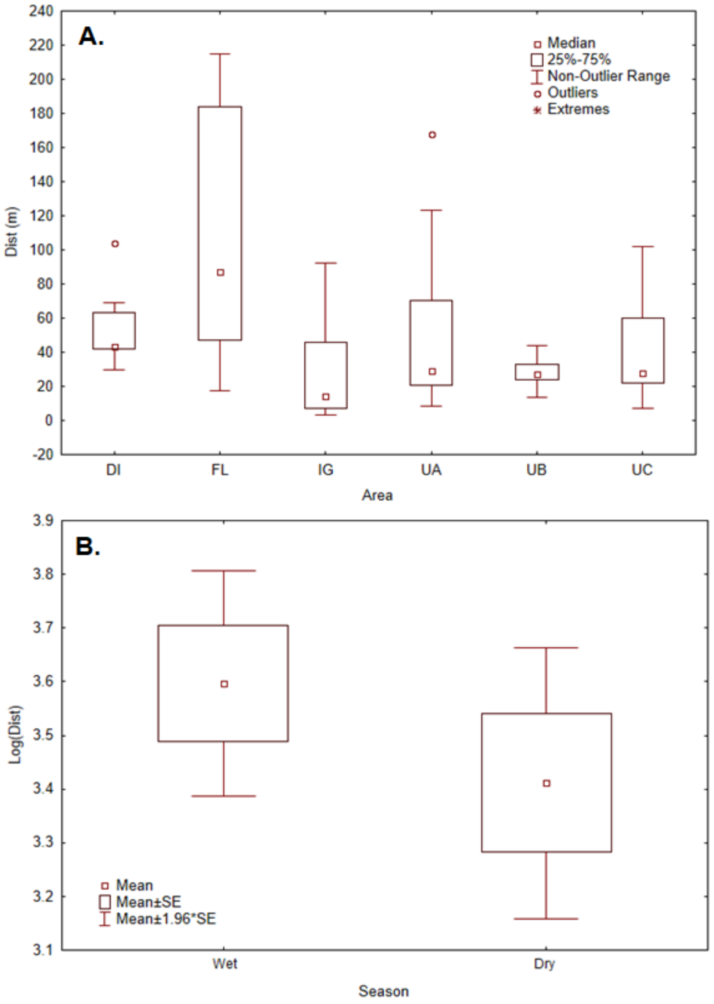

Figure 11. Variation of the movement distance by *Marmosa paraguayana* between sampling areas (**A**), and climatic seasons (**B**). The boxes represent the percentiles of the distances (25-75%), the square within the boxes the median of the distances, and the whiskers the minimum and maximum distances recorded.

Distances of movement and population parameters - There were differences in the movement distance among adult individuals according to sex, with males presenting larger movement distances than females (F =4. 70; D. F. =1;94; P=0. 033) (Figure 12).

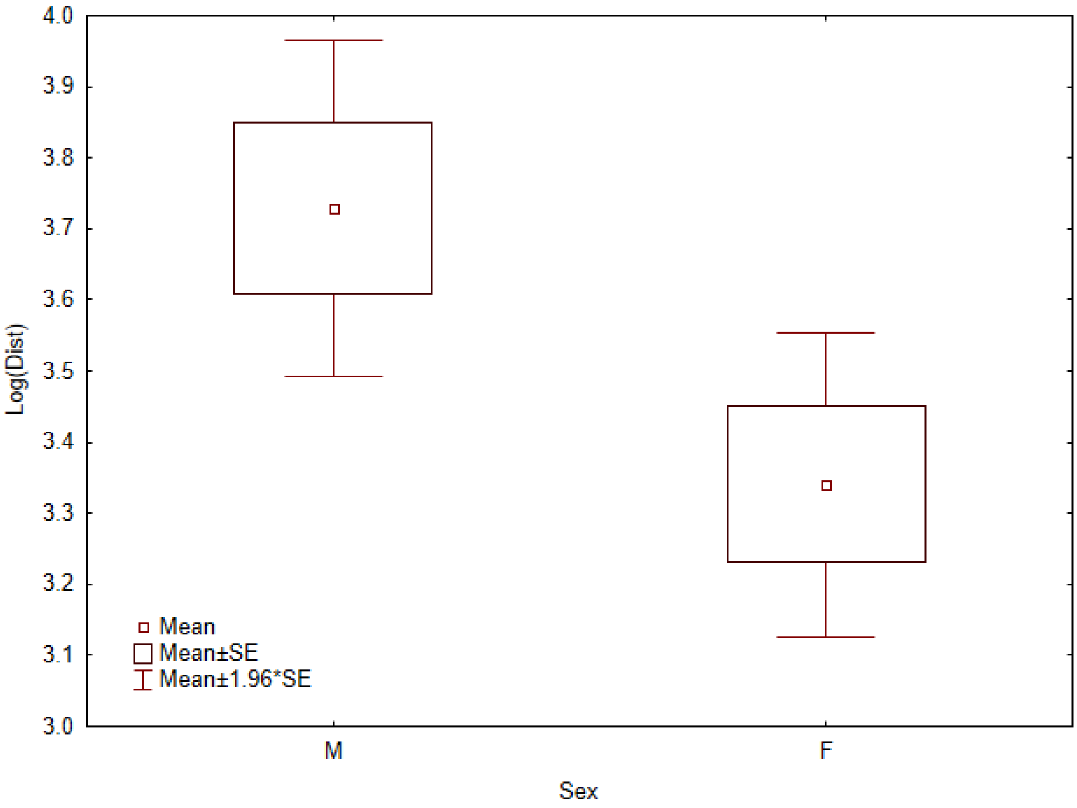

Figure 12. Variation of the distance traveled by male (M) and females (F) of *Marmosa paraguayana*. The boxes represent the values of the mean and standard error of the logarithm of distances traveled by each sex. The square within the boxes the mean of the logarithm of distances, and the whiskers the mean ±1. 96* the standard error.

Movements-We recorded 108 movements, of which only eight represented pipeline crossings (7.41% of all movements). Also, 97 movements were between traps within the forest (46 transect movements and 51 forest crossings). We registered eight recaptures in the same trap of the previous capture, which was not considered as movements (NM) (Table 3).

For the APA da Bacia do Rio São João, we registered 33 movements, of which two (6.1%) were pipeline crossings. On the REBIO União, we registered 72 movements, of which six (8.3%) were pipeline crossings (Table 3) (Figure 13). Despite the variation in the number of captures and recaptures, the pipeline crossings and forest movements rates were similar for both protected areas at the REBIO União (PCr=14.6% and FCr = 85.3%) and APA (PCr=12.5% and FCr = 87. 5%).

Table 3. Capture and recapture information for the sapling areas. (M) number of males, (F) number of females, Total mov: Total movements, Forest: FC, transect TM, Crosses: PC.

Area	Sex	Captures	Recaps	Total Mov	FC	TM	PC
APA	M	15	17	15	7	7	1
	F	12	20	18	6	11	1
	Total	27	37	33	14	18	2
REBIO União	M	23	27	26	14	9	3
	F	22	49	46	24	19	3
	Total	45	76	72	35	28	6

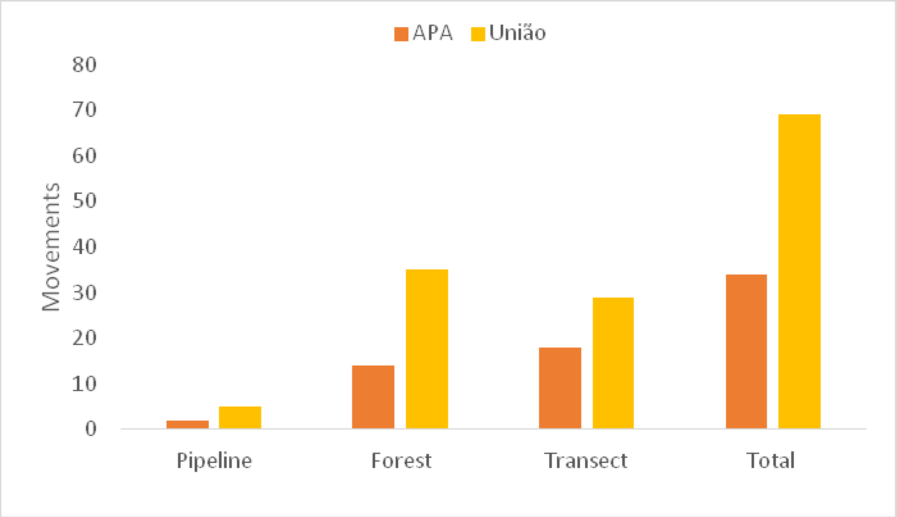

Figure 13. Movements of *M. paraguayana.* The movements are divided according to the conservation unit (APA or REBIO União) and the type of movement. Forest: FC, Transect: TM, and Pipeline: PC.

We recorded eight pipeline crossings, four by males (PCr=0.16) and four by females (PCr=0.12). Also, 30 of the 51 forest crossings were performed by females and 21 by males. Females did 30 of the 46 transect movements and the other 16 were made by males, giving a total of 64 movements for females (60.6% of the total movements) and 41 movements for males (39.4%) (Figure 14).

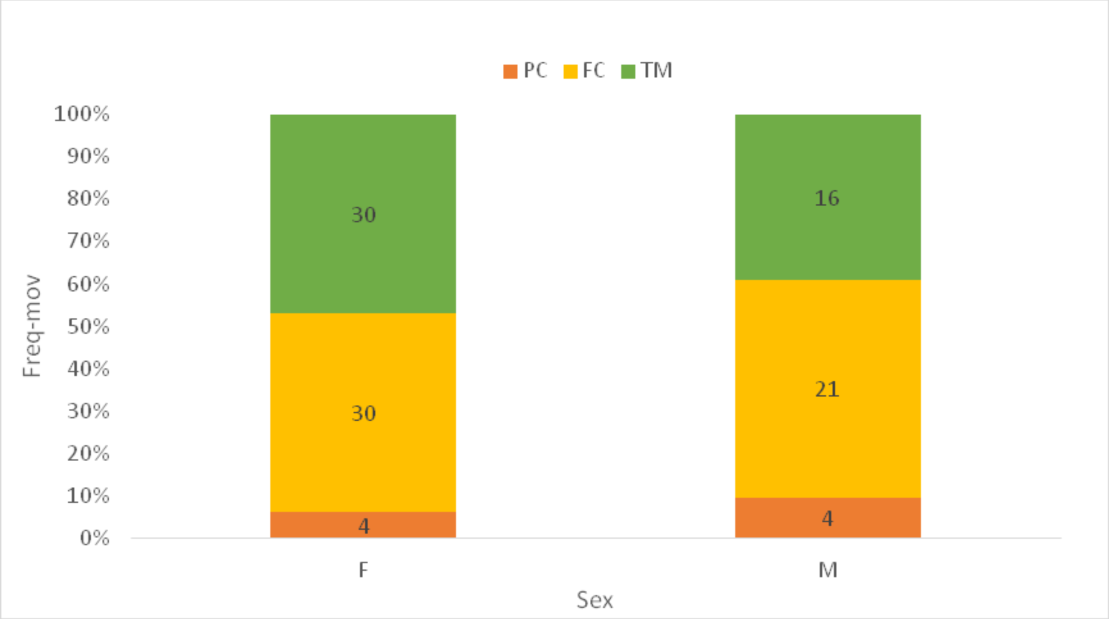

Figure 14. Frequencies of movements by sex made by *M. paraguayana*. PC: Pipeline crossings, FC: Forest Crossings, TM: transect movements.

The χ^2 test indicated that pipeline crossings are significantly less frequent than forest crossings (χ^2=29. 491; D. F. =1; p-value= <0. 001;). We also detected significant effects of sex and season on the pipeline crossings, as "season" was an explanatory variable present in the first and second best fitted models, while "sex" was present in the second best fitted model. Together, the first and second best fitted models accounted for 79% of the cumulated likelihood of all models (Table 4).

Table 4. Model selection Analysis of variables affecting crosses.

	Intercept	**Season**	**Sex**	**Season:Sex**	**R^2**	**d. f**	**logLik**	**AICc**	**ΔAICc**	**weight**
2	-1. 23	+	NA	NA	0. 14	4	-16. 56	42. 01	0. 00	0. 57
4	-1. 54	+	+	NA	0. 15	5	-16. 30	43. 96	1. 95	0. 22
1(null)	-3. 06	NA	NA	NA	0. 03	3	-19. 36	45. 24	3. 24	0. 11
8	-1. 54	+	+	+	0. 15	6	-16. 30	46. 55	4. 54	0. 06
3	-3. 47	NA	+	NA	0. 04	4	-19. 19	47. 26	5. 26	0. 04

Finally, the history of movements over time showed an increase of crossings in October 2018 and October 2019. Additionally, October 2019 was the month with higher FC and PC values. We did not record movements in February-2019 and neither PC from April to August -2019 nor in December-2019 (Figure 15).

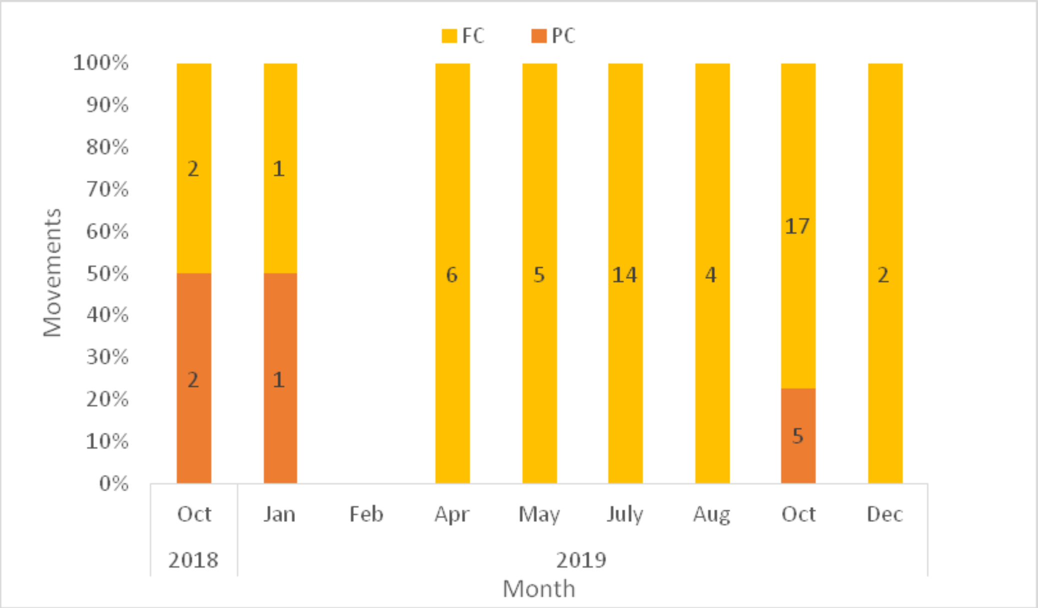

Figure 15. Pipeline crossings (PC) and forest crossings (FC) recorded for *M. paraguayana* over time.

DISCUSSION

The present study highlighted the detrimental effects of oil and gas pipelines on movements and connectivity for an arboreal Neotropical small mammal, which had not been previously evidenced. We demonstrated that the unforested corridor acts as a barrier, decreasing the rate of movements of *Marmosa paraguayana*, and leading to differences in population density between sides of the duct, and cause negative edge effects. The effect of the sex on the movement distances and pipeline crossing rates evidence that duct impacts toward the females.

The presence of the pipeline generated a negative edge effect that decreased the number of captures of *M. paraguayana* near the edge of the forest (Table 1). Considering that individuals move between the edge and interior, the number of captures is a proxy of habitat use. This result suggests that the use of habitat by this opossum is modified at the edge of the pipeline. Species response to habitat edges varies depending on the specific edge type encountered. These changes may be due to abiotic or biotic changes in the environment, changes in interspecific interactions, or a combination of these and other factors (RIES; SISK, 2004). Pardini (2004) pointed to an increase in captures of *Marmosa murina*, the most common arboreal marsupial, at the edge in a study conducted in a landscape of fragmented Atlantic Forest. Nevertheless, there was a reduction in number of species like *Marmosops incanus* and *Monodelphis americana*, suggesting that the fragmentation could be positive for the conservation of some species and negative for others. Donovan et al. (1997) pointed that a possible consequence of forest edges is the increase in predation by middle and big size mammals, therefore, animals that nest on the trees would avoid the edges. Another possible explanation is the human presence since the pipeline demands frequent maintenance to keep it free from arboreal plant material, and since it is used by hunters to access good hunting areas (RUIZ-MIRANDA et al. , 2018). Human presence and activities such as hunting, which are generally associated with the use of domestic dogs, have been classified as a generator of edge effect for small mammals (LACERDA; TOMAS; MARINHO-FILHO, 2009; VILLASEÑOR et al. , 2014). Our results also could indicate a geometric edge effect (GEE), in which it is explained that the animals are not avoiding the edge because of the changes in quality of the edge, but due to purely geometric processes. Thereby, due to the polygonal form of the fragment, points closer to the center can receive individuals of any direction at the interior of the forest, while the points at the edge cannot receive individuals from the outside of the fragment, and for that reason, the abundances tend to be lower at the edge (PREVEDELLO et al. , 2013). In this study, we did not measure the GEE. Nevertheless, due to the narrowness of the matrix (<100 m) that separates the two fragments, the presence of GEE would indicate that there is a very intense barrier effect that does not allow the passage of individuals between the fragments. We think that these results could be due to a combination between edge effect and GEE.

The number of captures of *M. paraguayana* was also influenced by the side of the pipeline (north or south). The south side tended to present more captures than the north side (Figure 10), contradicting our hypothesis. We also noticed that the south side has the smallest fragments of forest in the study areas. For that reason, we inferred that our number of captures was influenced by the size of the fragment rather than by the abundance. Pardini (2004) also pointed that the small mammal communities were richer and more abundant on small forest fragments in Atlantic Forest. Nevertheless, the effects of the edge on the abundance and richness were positive. For that reason, she inferred that in small forest fragments, the edge effects were more intense, and it contributed to the increase in richness and abundance. Genua et al. (2017) suggested that the fragmentation impacts are heavier on predators, and for that, the densities of predators are lower on small fragments and the densities of herbivores increased. Our data do not allow us to make inferences about these results, but there is the possibility that due to the limited space, the number of captures is greater on the smaller side of the pipeline.

The significant difference between the pipeline crossings (N = 8) and the forest crossings (N = 49), and the smaller crossing rates (PCr=12. 5%), indicated that the presence of the pipeline acts as a barrier to the passage of individuals, decreasing the connectivity between fragments for the wooly opossum. These results agree with those proposed by Pires et al. (2002) who, despite having found small crossing rates, determined that the populations of *M. paraguayana* could keep gene flux and therefore a metapopulation dynamic. In the present study, the matrix is narrower, and the fragments are larger but the results seem to persist. Our results are also in contrast with those of Lucas et al. (2019), who studied Golden Lion Tamarin (*Leontopithecus rosalia*, Linnaeus 1766), and did not find barrier effects of the pipeline on animal movements. This difference could be due to the larger size of *L. rosalia* compared to *M. paraguayana*, and therefore able to move greater distances (BAKER; DIETZ, 1996).

Concerning the spatial variation in crossing rates, we observed more captures, movements, and crossings at the three areas of the REBIO União than on the areas of the APA. The areas selected in the APA present higher anthropogenic intervention due to activities such as cattle, farming, and construction (CARVALHO et al. , 2004; LUCAS et al. , 2019). The anthropogenic uses of the matrix around the forest could be a determining factor of the abundance and behavior of the species modifying its perception of risk, orientation, and foraging behavior (BROWN; KOTLER, 2004; FAHRIG, 2017; VIEIRA et al. , 2009), and these could act synergically with the pipeline decreasing the species movement and abundance. Vieira et al. (2009) determined that economic activities in the matrix, like agriculture or urban settlements, influenced the local composition of small mammals inhabiting the Atlantic Forest fragments. Those authors observed that forest fragments near urban areas surrounded by highly disturbed matrices had a lower abundance of *M. paraguayana* compared to fragments near small rural properties. Our results suggest that, in addition to the lower abundance of *M. paraguayana*, fragments surrounded by matrices with higher anthropogenic intervention may also offer a higher resistance to the movements of this species.

Despite the barrier effect of the pipeline corridor on *Marmosa paraguayana*, we observed few crossings, and their rate varied throughout the year. We observed an increase in the number of crosses in October and January (figure 15), synchronized with our first records of reproductive females (female with newborn pups attached) (Figures 7 and 8). These results suggest that the crossings of *M. paraguayana* might be related to mating season. Additionally, males traveled longer distances than females, although females capture frequency was higher, agreeing with the results of Pires and Fernandez (1999). Male home ranges tend to be larger than female home ranges, as they travel higher distances searching for partners and females present territorial behavior, presumably due to parental care (Pires and Fernandez 1999). Several other studies also described relationship between movement distance and sex, or mating season (e.g., DAHLE; SWENSON, 2003; GLAUDAS; RODRÍGUEZ-ROBLES, 2011; KJELLANDER et al. , 2004; MADSEN, 1984). Dahle and Swenson (2003), working with brown bears (*Ursus arctos*), inferred that the shorter distances of movements presented by females were due to a protectorial behavior against infanticidal males, while for grass snakes (*Natrix natrix*), smaller home ranges of females were due to the availability of food to offspring development (MADSEN, 1984). In a study with quolls (*Dasyurus maculatus maculatus*), Belcher and Darrant (2004) determined that greater distances traveled by males were associated with the competition between males, which would be more intense in denser populations.

We detected differences in the distance of the movements in each area. The individuals of *M. paraguayana* in the Fazenda Flandria, moved longer distances than those on the other areas, even considering the number of captures. The Fazenda Flandria is the only sampling area completely flat. In all the other areas at least part of the transect presents some level of slope. Then, it is probably less costly to move longer at the Fazenda Flandria than at the other areas. Since we did not take into account the slope of the areas on the calculation of the movement distances, the above statement should be taken with care. JEANSON et al. , (2003) pointed that small animals tend to be more susceptible to this heterogeneity on the landscape. Nevertheless, we did not find any bibliography comparing movements for arboreal animals in different degrees of slope. We also noticed that the distance of the movements of *M. paraguayana* were not influenced significantly by the climatic season. Delciellos et al. (2019) determined that the movements were related to the density of the populations and with the climatic seasons (humid and super-humid) for the marsupial *Philander frenatus*. This difference could be related to differences in diet and locomotor habit. *Philander frenatus* is scansorial and move long distances on the ground. The species diet varies seasonally, being prone to eat insects and small vertebrates in the dry season and becoming more frugivorous in the wet season when it does not need to travel long distances to obtain food. (CEOTTO et al. , 2009; PREVEDELLO; DELCIELLOS; VIEIRA, 2009). Since the diet of *M. paraguayana* is mainly composed of fruits and seeds with no indication of seasonal variation (SANTORI; LESSA; ASTÚA, 2012), their distances of movement would not be influenced by the climatic season.

CONCLUSIONS

The fragmentation produced by the pipeline act as an impediment to the movements of *M. paraguayana* and implies in reductions of crossing movements between forest fragments. Nevertheless, generally males can eventually cross the pipeline, especially during the mating season. Our results support the hypothesis that the pipeline generates a negative edge effect for *M. paraguayana* populations. The distances of movements of this opossum seem to be affected by the slope of the area but are unaffected by the climatic season. The results highlight the importance of the conservation of forest remnants and incite to find ways to improve the connectivity between fragments to guarantee gene flow and survival of *Marmosa paraguayana* and other arboreal species that are intolerant to open corridors since the construction of linear structures is increasing in Brazil and the world.

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