



# El Colegio de la Frontera Sur

## Factores que afectan la distribución y abundancia de perros ferales y de libre rango en la Reserva de la Biosfera Tehuacán – Cuicatlán

Tesis

Presentado como requisito parcial para optar al grado de  
Maestría en Ciencias en Recursos Naturales y Desarrollo Rural  
Con orientación en Manejo y Conservación de los Recursos Naturales

Por

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# El Colegio de la Frontera Sur

San Francisco de Campeche, Campeche, 6 de septiembre de 2020.

Las personas abajo firmantes, miembros del jurado examinador de:





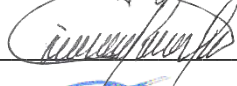
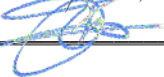
Orlando Reina Ponce

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**Factores que afectan la distribución y abundancia de perros ferales y de libre rango en la Reserva de la Biosfera Tehuacán – Cuicatlán**

para obtener el grado de: **Maestro (a) en Ciencias en Recursos Naturales y Desarrollo Rural**

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## **Dedicatoria.**

A mis padres quienes con su amor, paciencia y esfuerzo me han permitido llegar a cumplir hoy un sueño más, gracias por inculcar en mí el ejemplo de esfuerzo y valentía, de no temer las adversidades.

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## Resumen

La Reserva de la Biosfera de Tehuacán – Cuicatlán (RBTC), ubicada entre los estados de Oaxaca y Puebla, México, tiene poblaciones humanas inmersas, además de un relleno sanitario, que fungen como principal subsidio para los perros de libre rango (PLR) y perros ferales (PF) (*Canis lupus familiaris*). Esta característica tiene efectos directamente relacionados con la distribución y abundancia de PLR y PF en espacios silvestres. Para estimar los indicadores poblacionales que orienten la toma de decisiones sobre medidas de mitigación ante la presencia de PLR y PF en áreas naturales protegidas (ANP), se aplicaron cuatro métodos de monitoreo. Un método fue aplicado exclusivamente en el relleno sanitario y tres métodos diferentes en dos sitios de muestreo, en la misma escala espacial en un gradiente de subsidio humano. Se determinó el índice de abundancia relativa (IAR) de los PLR y PF, de los cuatro métodos de muestreo (captura-marca-recaptura, cámaras trampa, transectos lineales de ancho fijo y transectos de ancho variable). Los IAR de PLR y PF son mayores en el relleno sanitario ( $N= \pm 239.8$ ) y en las inmediaciones a este, demostrando que es un factor de subsidio importante. El método de captura-marca-recaptura fue el que más ventajas mostró con respecto a los otros. Lo anterior en virtud de los criterios de nivel de aplicación, tiempo de ejecución, esfuerzo de muestreo, esfuerzo de procesamiento de datos, nivel de precisión y costo. Las poblaciones humanas y el relleno sanitario al interior del ANP exacerban la presencia de los PLR y PF, teniendo un efecto negativo en la vida silvestre. Se espera que, los resultados obtenidos en el presente estudio sean la piedra angular para el establecimiento de estrategias de conservación y uso sustentable del territorio en la RBTC con respecto al control y/o erradicación de PLR y PF.

**Palabras clave:** abundancia, conservación, distribución, perros de libre rango, perros ferales.

## Introducción

Los humanos han establecido una estrecha relación con animales domesticados para su subsistencia, entre ellos el perro (*Canis lupus familiaris*), con quien la relación se remonta al menos 15,000 años de acuerdo con restos arqueológicos (Guagnin et al. 2018). Este es el canido más abundante del mundo, con una población entre los 700 y 1,000 millones de individuos aproximadamente (Hughes y Macdonald 2013; Gompper 2014). Es considerada una especie oportunista y generalista (Paschoal De Oliveira et al. 2016) subsidiada por el humano; con una amplia flexibilidad de comportamiento y altas tasas de reproducción (Gompper 2014). Además, los perros han amenazado y extinguido especies silvestres alrededor del mundo (Hughes y Macdonald 2013). Tienen una flexibilidad dietética que contribuye a su capacidad de sobrevivencia en una amplia gama de ambientes, que genera diversos conflictos con el ser humano y la vida silvestre. Por lo anterior, tienen efectos negativos, como la transmisión de enfermedades, depredación de ganado y vida silvestre, competencia e interferencia (Hughes y Macdonald 2013; Gompper 2014; Doherty et al. 2017; Young et al. 2018). Las Áreas Naturales Protegidas (ANP), no están exentas de la presencia de Perros de Libre Rango (PLR) y Perros Ferales (PF), ni de sus impactos, especialmente graves si existen especies amenazadas o en peligro de extinción (Zapata-Ríos y Branch 2018; Ellwanger y Chies 2019).

Al igual que otros depredadores, los PLR tienen roles funcionales importantes en la estructuración de las comunidades ecológicas (Vanak y Gompper 2009). La mera presencia de un depredador en un entorno natural puede afectar a las presas de forma sutil, subletal e indirecta de manera aparentemente nocivas (Preisser et al. 2005; Zanette et al. 2011). La respuesta de la vida silvestre a la presencia de un estímulo amenazante, como un perro, se conoce como interferencia (Amarasekare 2002). Estas respuestas implican la interrupción de actividades o estados normales, frecuentemente evocan comportamientos antidepredadores, que son cambios de comportamiento en presencia del estímulo amenazante (Vanak y Gompper 2009; Gompper 2014). Así mismo, implica cesar actividades normales, por ejemplo, alimentación, cuidado parental, descanso, exhibición, entre otras (Gompper 2014).

Gompper (2014) menciona que las principales fuentes de subsidio para la supervivencia de los PLR y PF, son proporcionadas por los humanos. Las cataloga de acuerdo con el nivel de asociación y dependencia con el humano en seis categorías: perros con dueño, perros urbanos libres, perros rurales libres, perros de pueblo (considerados los tres anteriores como perros de libre rango (PLR), perros ferales (perros domésticos que ya no tienen dependencia del humano) y perros salvajes (cánidos silvestres que no han pasado por un proceso de domesticación efectiva a través de la historia).

Se han documentado algunos factores que afectan la distribución y abundancia de los PLR y PF; por ejemplo, en áreas con una alta densidad de población humana, los PLR se encuentran en densidades más altas (Soto y Palomares 2015). Otro de los factores son los basureros al interior de las ANP con un mal manejo de los desechos producidos por los humanos, que brindan una fuente de alimento no intencional. Esto permite que los PLR y PF puedan subsistir, convirtiéndose en una especie omnipresente en todo el hábitat (Gompper 2014), con posibilidad de pasar por procesos de feralización y ser reclutados o ser formadores de manadas de perros ferales (Hughes y Macdonald 2013). En un estudio realizado por Morin (2018) en E.E.U.U., el 75% de las detecciones ocurrieron a <500 m de las edificaciones creadas por los humanos. Además de que existe una mayor asociación en la ocupación por PLR y PF en bosques y terrenos privados, en comparación con terrenos públicos y humedales. Demostrando que la ocupación de PLR y PF aumentó con el porcentaje de pastizales, edificaciones y propiedad privada de la tierra. En cambio, la ocupación disminuye con la lejanía a las edificaciones o poblaciones humanas (Morin et al. 2018).

En la Reserva de la Biosfera de Tehuacán Cuicatlán (RBTC) se tiene poco documentado en medios no formales (e.g. diarios locales) la presencia de PLR y/o PF. Su presencia ha provocado conflictos con los humanos por ataques al ganado doméstico y a las especies silvestres. Por lo anterior es importante conocer las características de las poblaciones de PLR y PF, dentro del área natural protegida de la RBTC, y los factores que afectan su distribución y abundancia.



## **Justificación**

La investigación se enfocó en analizar los métodos para obtener los IAR, a fin de conocer que factores afectan la distribución de los PRL y PF (*Canis lupus familiaris*) en la RBTC. Los resultados aportarán información sobre la abundancia y distribución de los PRL y PF dentro del ANP. Además, servirá como referente para la toma de decisiones, acciones de conservación y futuras investigaciones, en cuanto a los efectos de las actividades antrópicas sobre la distribución y abundancia de los PLR y PF al interior de la RBTC.

## **Pregunta de Investigación**

¿Cuál es el mejor método para obtener los índices de abundancia relativa en poblaciones abiertas de PLR y PF en la RBTC?

## **Hipótesis**

Los métodos utilizados para estimar los IAR y la distribución de los PLR y PF presentan diferencias significativas en relación con diferentes factores (e.g. como medio de subsidio, densidad de poblaciones humanas y actividades antrópicas).

## **Objetivo**

Determinar cuál o cuáles son los mejores métodos para estimar la abundancia y distribución de los PLR y PF (*Canis lupus familiaris*) en la RBTC.

## **Objetivos particulares**

- Comparar los diferentes métodos en relación con su aplicabilidad y resultados en cada sitio de muestreo.
- Identificar los factores en los diferentes sitios de muestreo que afectan la distribución y abundancia relativa de los perros ferales y de libre rango de la RBTC.

## Artículo científico

### ***Too many dogs in the park I: Comparing four methods for estimating the abundance of open populations of free-ranging dogs in the Tehuacan-Cuicatlan Biosphere Reserve, Mexico.***

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## Abstract

The Tehuacan – Cuicatlan Biosphere Reserve (TCBR), encompasses several human settlements, in addition to a garbage dump that serves as the main subsidy to maintain populations of free-ranging dogs (FRD) and feral dogs (FD), *Canis lupus familiaris*. Little is known about the best monitoring methods to estimate population indicators of FRD. Four monitoring methods were applied: one method applied exclusively for the garbage dump, and three different methods in two sampling sites, over the same spatial scale, to estimate the population indicators that guide decision-making on mitigation measures in the presence of FRD in protected natural areas (PNA). FRD monitoring is an essential activity for the management and control of their free-living populations, mainly in PNA, where they can become harmful to the environment. Through the application of four different monitoring methods to estimate FRD population indicators, we determined which of these are best adapted to varied sampling efforts. Traditional monitoring methods and, specifically, the capture-mark-recapture method applied to FRD are mainly designed for closed populations. Due to the complexity of free-living monitoring, and since FRD are reclusive in the presence of humans, we adapted a method for an open population at a landfill, mainly based on a study conducted in India. The four methods used in FRD monitoring in open populations show advantages and disadvantages, but we consider that in some situations (e.g., costs, execution time, materials, etc.) the C-M-R model based on photographs and individual identification best fits most sampling models in relation to sampling effort, analysis, and expected outcomes.

**Keywords:** Abundance, *Canis lupus familiaris*, dogs, estimation, feral, Mexico.

## Introduction

Free-ranging dogs (FRD) and feral dogs (FD) (*Canis lupus familiaris*) are considered an invasive species, since humans have established a close relationship dating back about 15,000 years according to archaeological remains (Guagnin et al. 2018). The nature of this close relationship has, in many cases, made invisible the damage that dogs can cause to the environment. Dogs are the most abundant carnivore in the world, with a population between 700 and 1,000 million worldwide (Hughes and Macdonald 2013; Gompper 2014). Dogs are opportunists and generalists (Paschoal De Oliveira et al. 2012), in addition to being an invasive species that is subsidized by humans; with wide behavioral flexibility and high reproductive rates (Gompper 2014). Dogs have threatened and extinguished various wildlife species around the world (Hughes and Macdonald 2013). Protected Natural Areas (PNAs) are not exempt from the presence of free-ranging dogs, and their impacts can be especially serious if there are threatened or endangered species in them (Zapata-Ríos and Branch 2016; Ellwanger and Chies 2019). In some PNAs, the abundance of dogs even exceeds that of native carnivores, so they have a high potential impact on these ecosystems and their native fauna (Vanak and Gompper 2009; Silva-Rodríguez and Sieving 2012; Lessa et al. 2016).

In order to estimate FRD and FD populations, statistically robust analytical methods are required. There are studies on very different FRD and FD population sizes in the same geographic areas with methods that require large sample sizes, which is complicated in open populations, therefore raising doubts about these estimates of FRD populations (Belo et al. 2017). Similarly, capture-mark-recapture methods, in their different variants, are perhaps the most widely used sampling methodology, and distance sampling is the least used (Narváez y Zapata-Ríos 2020). However, the above methods do not consider the FRD and FD populations as an open population, that is, one that is subject to population dynamics such as deaths, births, immigrations, and emigrations (Graipel et al. 2014). Other methods of estimating dog population size include extrapolation of human:dog ratios derived from household surveys and mark-and-resight methods (transects) using dog radio-collars or paint sprays (Tenzin et al. 2015).

Individual identification in the C-R-M method is resource-intensive; for example, photographic comparison (Tiwari et al. 2018), which is not well suited to operational practicalities when there is a large population of FRD and FD, or when the spatial scale is too large, or in patches (Rowcliffe

et al. 2008). In contrast, the distance sampling techniques that have been used in several studies in Asia (Sambo et al. 2018), do not require capture or marking, but are based on measuring the perpendicular distances of the animal from a randomly placed transect, resulting in a simple technique to estimate abundance when individuals cannot be identified. Several papers that use this method only report the population density, because the method is possibly better suited for these indicators than for the estimation of abundance (Rowcliffe et al. 2008; Krauze-Gryz and Gryz 2014; Hall et al. 2016; Hongo et al. 2021).

In this study, the objective was to document the distribution and abundance of FRD and FD by comparing various methods, in order to improve management decisions on population control (Suárez-Tangil and Rodríguez 2021). Many mammalian species, including FRD and FD, are inconspicuous, have elusive behaviors, and require large areas to meet their needs, making direct detection of individuals difficult (Gompper 2014). For this reason, it is important that sampling in large areas allows spatial and temporal replication, as well as short-time records of presence data, and that the methods are economically affordable and effective in detecting the target species. Thus, the comparison of different monitoring methods for open populations of FRD and FD is relevant, through criteria that explain their effectiveness and feasibility of application. Therefore, sampling effort, logistical difficulties, data processing effort, execution time, cost, among other variables are often important factors to consider the one method that best suits the needs of particular studies.

## **Materials and methods**

### **Study Area.**

The Tehuacan-Cuicatlan Biosphere Reserve (TCBR) covers an area of approximately 490,186 ha between the states of Puebla and Oaxaca, Mexico, with a total of 51 municipalities, 20 in Puebla and 31 Oaxaca (SEMARNAT-CONANP 2013). The valleys, hills, and ravines that make up the reserve are framed by the Sierras de Juarez to the south, Mixteca to the west, and Mazateca to the north, which make up a great environmental complexity in an altitudinal gradient that varies between 500 and 2,900 m. These mountain ranges form a barrier that decreases the amount of rain and humidity in the region, causing dry heat and little rain, resulting in aridity in a good part of the

TCBR, mainly in the Tehuacan Valley (Pérez-Irineo and Santos-Moreno 2013). The region's climate is warm semi-dry and warm semi-tropical in the Cañada Cuicateca region, with moderate to scarce rainfall in the summer. The predominant flora in the TCBR is xerophyte, which comprises more than a third of the species detected in the area, followed by the low deciduous forest that accounts for a fourth, and temperate forest to a lesser extent (figure 1). As it happens with the flora, the animals of the TCBR vary greatly according to the ecosystems. Among the vertebrates, it is estimated that there are 14 species of fish, 83 species of reptiles (10% of the total for Mexico), 28 species of amphibians and 102 species of mammals, of which a third are bats (almost 30% of the bats in the country) (SEMARNAT-CONANP 2013). Within the TCBR, there is a garbage dump that has been operating since 1995 and receives about 300 tons of waste per day from the city of Tehuacan and the surrounding area (OOSELITE 2018). This site has a discontinuous perimeter fence, which is non-existent in some sections, allowing FRD and FD to use it as a refuge and feeding site. We consider the three sites described below to be potential sources of subsidy at different scales for the open populations of FRD and FD in the region (Gompper, 2014) (Figure 1).

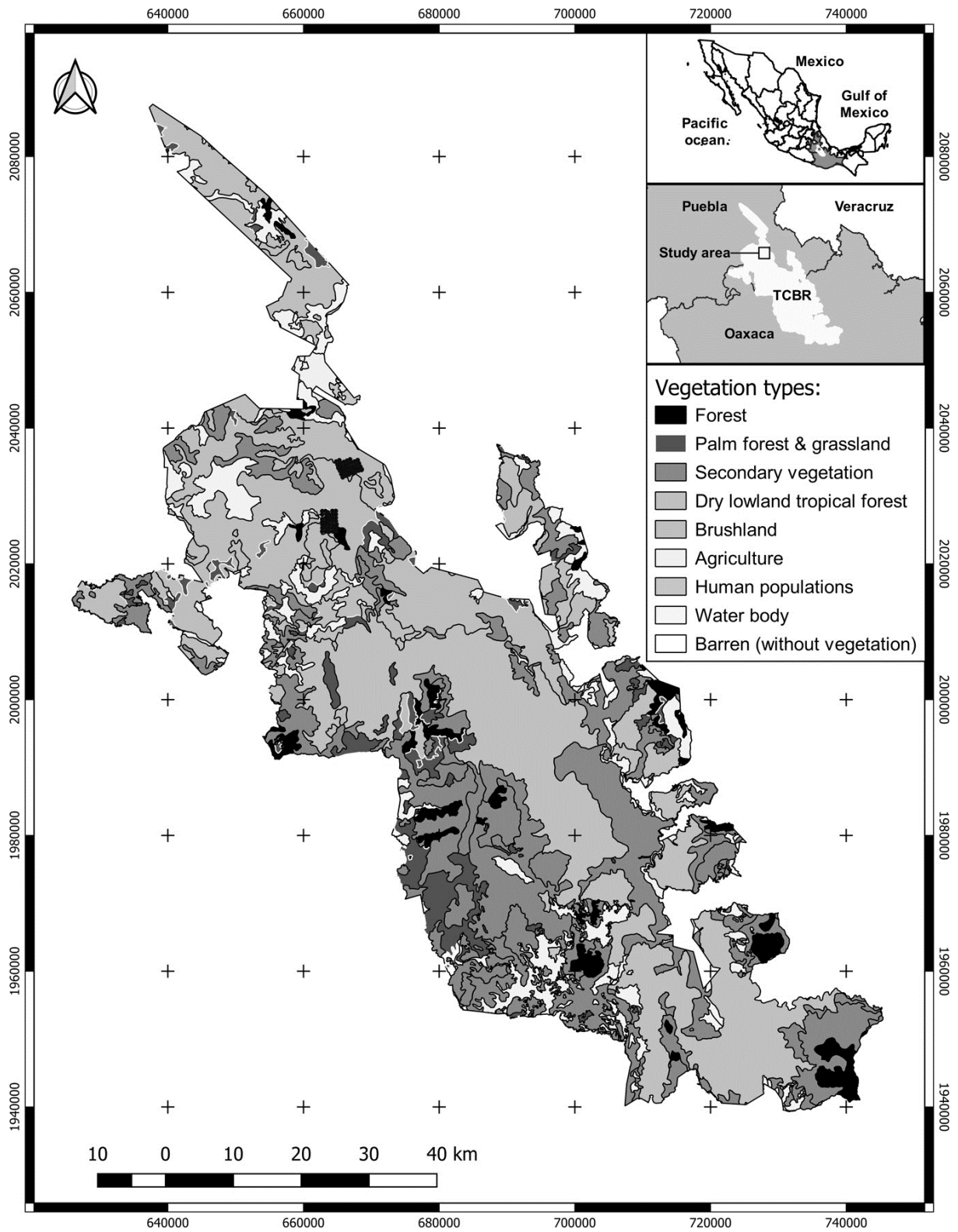


Figure 1. Map of the study area and the main types of vegetation of the TCBR.

Three sites were selected within the TCBR; the first site (Site 1) being the auxiliary board of San Antonio Texcala located in the Municipality of Zapotitlán Salinas in the State of Puebla, Mexico and is located at the coordinates longitude: -97.443611 and latitude (dec): 18.395556 at an average height of 1220 meters above sea level (masl). which is characterized by its proximity to the landfill and has a population of 1,539 inhabitants. The second site (Site 2) was the municipal seat of Zapotitlan Salinas located in the Zapotitlán Municipality of the State of Puebla, Mexico and is located at the coordinates longitude: -97.474444 and latitude: 18.331944 and is located at an average height of 1500 masl, with a population of 3,215 inhabitants. The third site that was chosen was the Tehuacan Garbage dump with approximately 12.7 ha., because it is a means of constant subsidy for the FRD and FD, due to its poor infrastructure and poor waste management.

A total of four sampling methods were applied to estimate the abundance of FRD: three methods in Sites 1 and 2 with an area of 600 ha each, and one method in the Tehuacan landfill where the presence of FRD and FD suggests a high probability of detection to estimate relative abundance.

### **Capture-Mark- Recapture (C-M-R) method based on natural marks**

In many FRD and FD populations, some individuals may have natural markings with which they can be easily visually identified (Punjabi et al. 2012). Sampling with this method was carried out between February 9 and 12, 2021. Two kilometers were sampled in five independent linear transects, georeferenced with a Garmin® GPS eTrex 20 model. Along the transects, direct FRD observations were made, and high-quality photographs were taken of each of the registered individuals, with a Reflex Nikon 5600 series professional manual camera, with a Nikon V<sub>r</sub>II 70-300 mm lens. In addition, the dogs were identified by their natural markings, such as color, size, spots or characteristic body markings, sex, injuries, physical condition (Figure 2 A). The transects were covered four times (j) every 24 h on foot with a continuous unidirectional march, to avoid re-counting the same individual (Punjabi et al. 2012). On the first walk, all dogs were marked, and, during subsequent walks, all marked and unmarked dogs were recorded. With the data obtained, an analysis was carried out with the POPAN statistical model (Schwarz, 1996) with the MARK version 6.2 software for Windows 10, considering that the population is open without demographic closure (White and Burnham 1999; Cooch and White 2019) and that it fulfills the following assumptions:

1. Each marked animal in the sampled population  $i$  has the same capture probability ( $p_i$ ).
2. Each marked animal in the population has the same probability of surviving until period  $i+1$ , that is, immediately after sampling period  $i$ .
3. The marks are not lost.
4. Sampling is instantaneous (in relation to the interval between period  $i$  and  $i+1$ ), and the release of individuals is done immediately after sampling.
5. All individuals, marked and unmarked, have the same probability of being captured.

Four Parameter Index Matrix (PIM) models were created for each parameter:  $\phi$  (apparent survival),  $p$  (probability of capture given that the animal is alive and in the study area, i.e. available for capture),  $\text{pent}$  (probability of entry into the population for this occasion) and  $N$  (size of overpopulation). For  $t$  occasions, there are estimates of  $t - 1$   $\phi$ , estimates of  $t$   $p$ , estimates of  $t - 1$   $\text{pent}$ , and estimate of  $1$   $N$ . The estimates  $t - 1$   $\text{pent}$  correspond to the probability of entry for occasions 2, 3, and 4  $t$ . The probability of being in the population on the first occasion is equal to  $\text{pent}(0) = 1 - \sum (\text{pent}_i)$ . The MLogit link function provides a constraint that makes the sum of the stored parameters  $\leq 1$ , with the probability of occurring in the population on the first occasion as  $1 - \sum (\text{pent } t)$ . Convergence of this model is problematic unless the MLogit link function is used with the contained parameters (Cooch and White 2019).

This method was applied to estimate the relative abundance of FRD and FD in the Tehuacan Landfill, based on the baseline methodology, which can be used at variable spatial scales, especially in the absence of artificial markings (Punjabi et al. 2012).

### **Camera Trap Method**

The observation of FRD and FD in natural conditions is generally difficult due to their behavioral patterns, since they tend to be elusive and avoid approaching humans. For this reason, camera traps are tools widely used worldwide to monitor wildlife populations. In PNAs, they have been used to detect the interactions of invasive alien species and native wildlife (Murphy et al. 2019).

During the study, 35 camera traps were used, 23 Cuddeback cameras model 1279 and 12 Bushnell model TrophyCam HD cameras, with a separation of 500 m between cameras, forming a grid (figure 2 B). They were placed 30 or 40 cm from the ground for 30 days and no attractant was used.



At Site 1, they were used between the months of February to April 2021; and at Site 2, from May to July 2021 (Figure 2 B). This method was not applied in the landfill for security reasons, because there is a greater presence of humans and there was a high risk that the cameras would be stolen. The cameras were active 24 hours a day and were programmed for motion detection: three consecutive photos in capture mode, a 15-second video, and a 30-second pause between each sequence. Location data was taken with a Garmin eTrex 20 model Global Positioning System, as well as the date and time at the moment of activation (González Bonilla et al. 2018). These photos were also used for individual identification and modeling of natural markings in a mark-capture-recapture model (above). In this study, only the data on the number of captures were used and the number of independent photographic records acquired per 100 nights/trap was considered to calculate the relative abundance index (O'Brien et al. 2005).

The relative abundance index (RAI) of the FRD and FD was calculated, using the following formula:  $RAI = (C/SE) \times 100$ . Where, C = number of captures or events photographed, SE = sampling effort (number of cameras per monitoring nights) per season or total, and 100 nights-trap (standard correction factor) (Jenks et al. 2011), for each of the sites, and the Wilcoxon-Mann-Whitney (WMW) non parametric test was applied to determine the difference in the mean of one group with respect to another (*p-value* = 0.05) (Sanchez 2015).

### **Fixed-Width Line Transect Method (FWLT)**

Indirect counting of wildlife species through tracks is a reliable method. It has been used in several studies to estimate an index directly related to the relative abundance of wildlife that can be compared between sites (Carrillo et al. 2000; Reyna-Hurtado and Tanner 2005; Weber 2010; Zapata-Ríos and Branch 2016).

Tracks from an animal crossing a transect were counted as a single observation. Tracks that were along a transect were taken as an observation (Carrillo et al. 2002; Zapata-Ríos and Branch 2016); dog tracks were identified by guides (De Angelo et al. 2017) and differentiated along with the excreta of other mammals using the Manual for the Tracking of Wild Mammals of Mexico (Aranda Sánchez 2016). Five GPS-georeferenced transects were established, with a fixed width of 6 meters for each study site (Site 1 and Site 2) and with a length of 100 meters each, on dirt roads or trails

(figure 2 B). These were walked on four occasions with an interval of approximately seven days between walks, between June and August 2021, and the footprints and excreta of the FRD and FD were counted and identified. In order to assess the abundance of FRD and FD. The relative abundance indices of Site 1 and Site2 were estimated based on encounter rates along transects (number of tracks and independent excreta / 10 km<sup>2</sup> of sampling area) (Figure 2 B).

### **Variable-Width Line Transect Method (VWLT)**

Distance sampling models estimate density or abundance from direct observations of individuals or groups, taking imperfect detection into account, using recorded perpendicular distances between individuals or groups and a line transect (Buckland et al. 1993; Buckland et al. 2015).

The main assumptions of this method are: 1) be sure that every animal that is close to the center of the transect will be counted with 100% certainty, 2) measure exactly the perpendicular distance of the individual, at the initial moment of its detection, and 3) do not count the same individual twice. In order to obtain reliable and robust estimates, a number of observations > 60 are required (Narváez and Zapata-Ríos 2020).

Five georeferenced transects were traced with a global positioning system, covering 500 m each in Site 1 and Site2 (figure 2 B), and the perpendicular distance of the FRD and FD was measured with a Leupold 8x30 mm laser rangefinder; as well as medium and large wild mammals that could be observed during the walk along the transect. The transects were covered four times each between June and August 2021.

In order to estimate density and relative abundance using this method, DISTANCE® software version 7.2 for Windows was used and maps made in QGIS software v.3.22.3 were coupled.

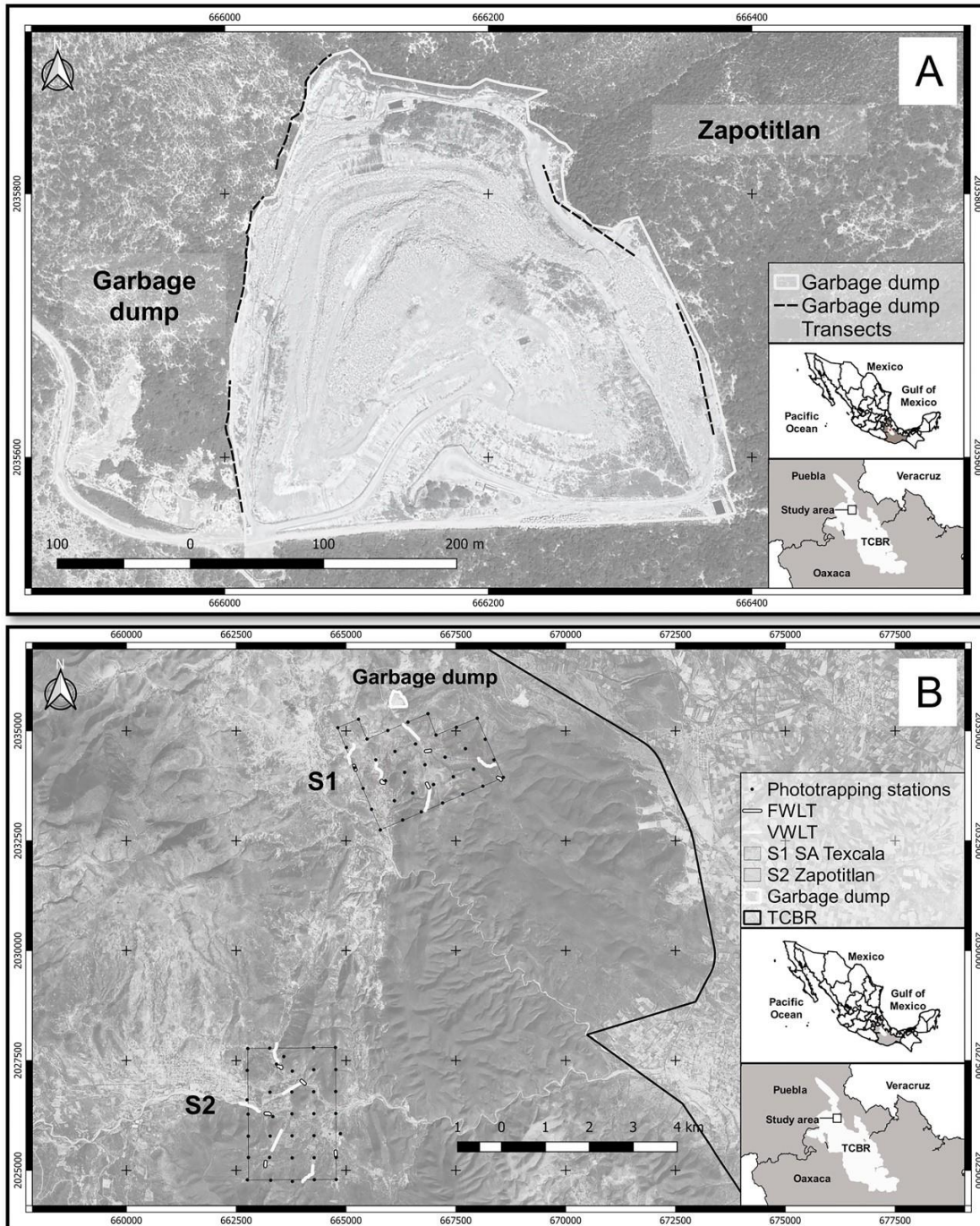


Figure 2. A) Capture-mark-recapture method used in the landfill (garbage dump), where five line transects of 100 meters each were placed, located inside and in the vicinity of the site, because it does not have a perimeter fence in good condition that allows FRD and FD to be kept out of the site. B) Sampling sites and application of three different methods (two transect methods Fixed-Width Line Transect Method (FWLT), Variable-Width Line Transect Method (VWLT) and one camera trap method) to obtain relative abundance indices.

## Results

### Capture-mark-recapture (C-M-R) method by natural markings

With a sampling effort of four days ( $j_i$ ), a total of 224 independent records were obtained at the landfill (table 1). The entry of new individuals into the population was evidenced, so the population is open, and its abundance varies over time.

<i>Occasions</i>	$j_0$	$j_1$	$j_2$	$j_3$
<i>Marked</i>	182	18	3	21
<i>Recaptured</i>	-	85	49	88

Table 1. A total of 224 FRD and FD were marked on the first occasion ( $j_0$ ), of which a total of 42 new individuals was obtained during the occasions after the first capture. Overall, 222 individuals were recaptured throughout the sampling.

To obtain the RAI, 14 models with the parameters to be studied were analyzed through the POPAN model in the MARK v. 6.2. software for Windows 10; 12 of these had convergence,  $s <$  and only two did not have convergence (both with the same Akaike Information Criterion weight). The model with the lowest standard error, best fit to the data and a 95% confidence interval (CI) was chosen, indicating that the probability of survival ( $\phi$ ) varies with time between 0.6 ( $\pm 0.06$ ; CI: 0.47-0.73) and 0.7 ( $\pm 0.08$ ; CI: 0.52-0.85); the probability of capture ( $p$ ) varies over time between 0.4 ( $\pm 0.06$ ; CI: 0.31-0.54) and 0.5 ( $\pm 0.5$ ; CI: 0.42-0.63); the probability of entry ( $p_{ent}$ ) varies over time between 0.7 ( $\pm 0.02$ ; CI: 0.04-0.12) and 0.13 ( $\pm 0.02$ ; CI: 0.08 -0.19); and overpopulation ( $N$ ) is 229.82 ( $\pm 3.34$ ; CI: 226.04-240.59) FRD. The gross estimation of the population with the same model, with a confidence interval (CI) of 95%, resulted in ( $N^*_{-hat}$ ) of 239.8 ( $\pm 4.09$ ; CI 231.95 – 248.00) FRD in the same site.

### Camera traps

A total of 47,650 photographs were obtained (Table 2), of which those that contained other mammals or other fauna and/or that did not contain FRD and FD were eliminated.

***Total number of photos and independent records of FRDs and FDs***

	Photos	Independent records	Capture effort (Camera trap nights)
<i>Site 1</i>	32,861	200	1089
<i>Site 2</i>	14,789	31	1124

Table 2. A higher number of independent records of FRD and FD were obtained at Site 1 compared to Site 2

A Whitney-Wilcoxon Man U test was applied to the data obtained, with continuity correction ( $p$ -value = 0.03487), a difference between sites. The RAI for Sites 1 and 2 resulted from the number of independent records for each monitoring site (figure 3), with a 95% confidence interval for Site 1 (CI=1.35 -1.17) and for Site 2 (CI = 1.35 -1.17). Site I is closer to the garbage dump where FRD and FD had abundant human subsidized food supply (Figure 3)

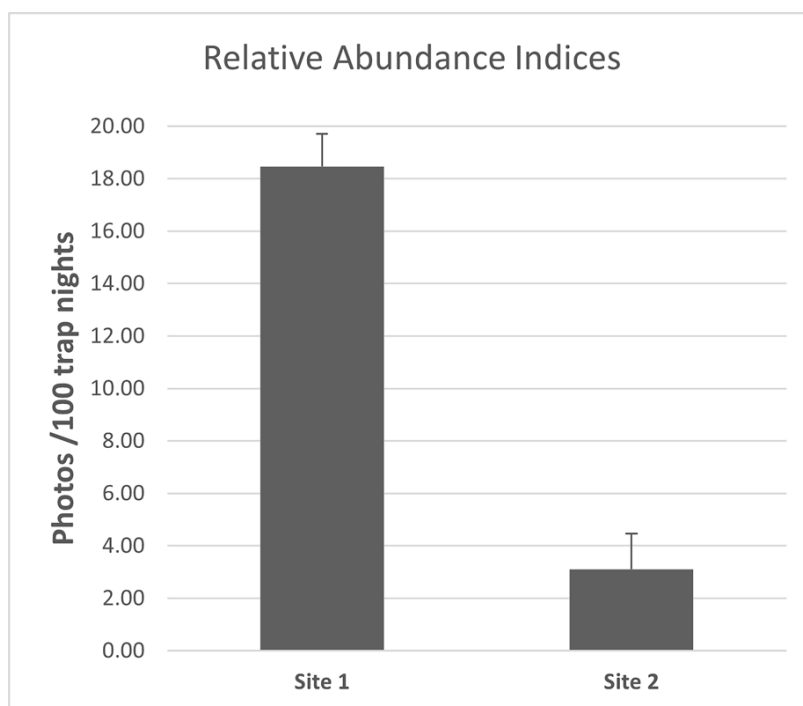


Figure 3. Relative abundance indices (RAI) for every 100 monitoring nights were higher in Site 1 (in the vicinity of the garbage dump), compared to Site 2 (in the vicinity of the town of Zapotitlan Salinas). The bars show the standard error of the sample.

### Fixed-width line transects

The total area sampled was 12 km<sup>2</sup>, counting the tracks (footprints and excreta), and individual analyses of the groups of footprints and excreta were performed, obtaining abundance through the encounter rate. The RAI of footprints was 1.25 with a confidence interval of 95% (CI= 4.89 – 0.31); and that of excreta was 2.5 (CI=6.21 – 3.79) for Site 1. In contrast, for Site 2, the RAI of footprints was 1.7 (CI= 4.33 – 2.47); and that of excreta was 1 (CI= 5.25 – -0.85) (figure 4).

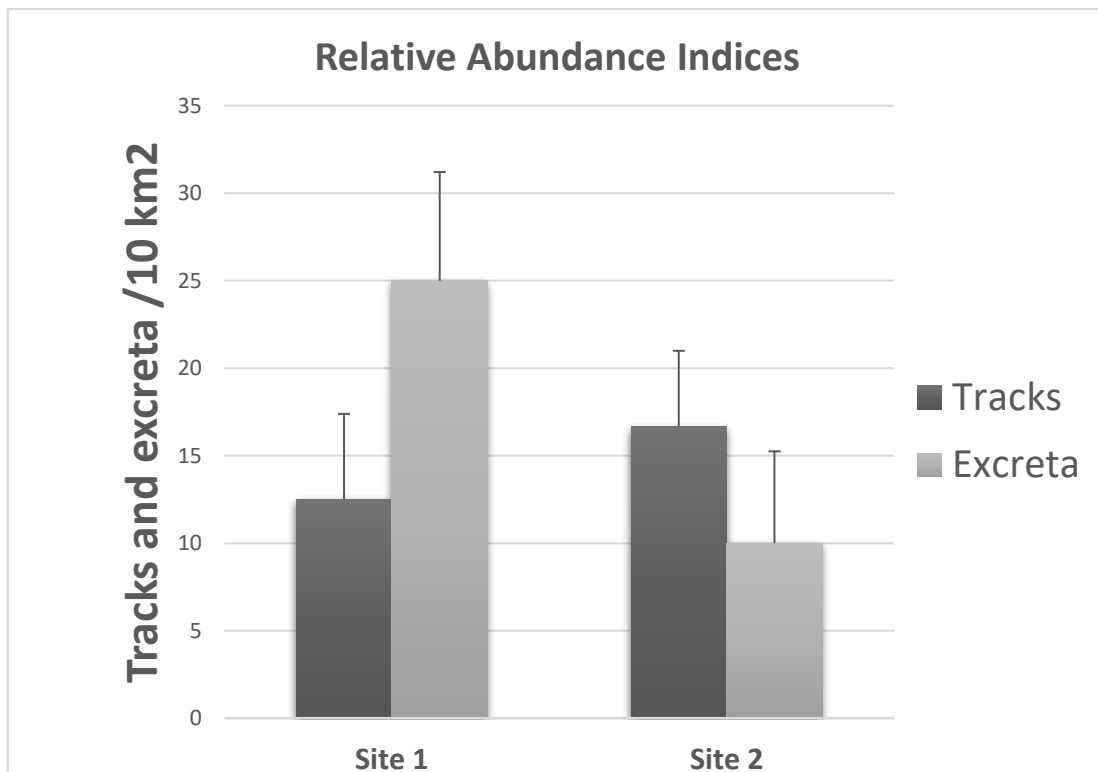


Figure 4. Number of tracks & excreta obtained per monitoring area at sampling Site 1 and 2, indicating the differences in the abundance of tracks between sites (CI=95%). The bars show the standard error of the sample.

### Variable-width line transects

Records of 18 individuals for Site 1 and five individuals for Site 2 were obtained, so it was not possible to estimate abundance through the Distance software (due to than minimum sample size required is 60 >). The RAI was obtained with the method of encounter rates / 10 km (figure 5), with a confidence interval of 95%. For Site 1, the RAI was 1.8 (CI= 4.71 – 2.49); and for Site 2, the RAI was 0.5 (CI= 0.71 – -0.23).

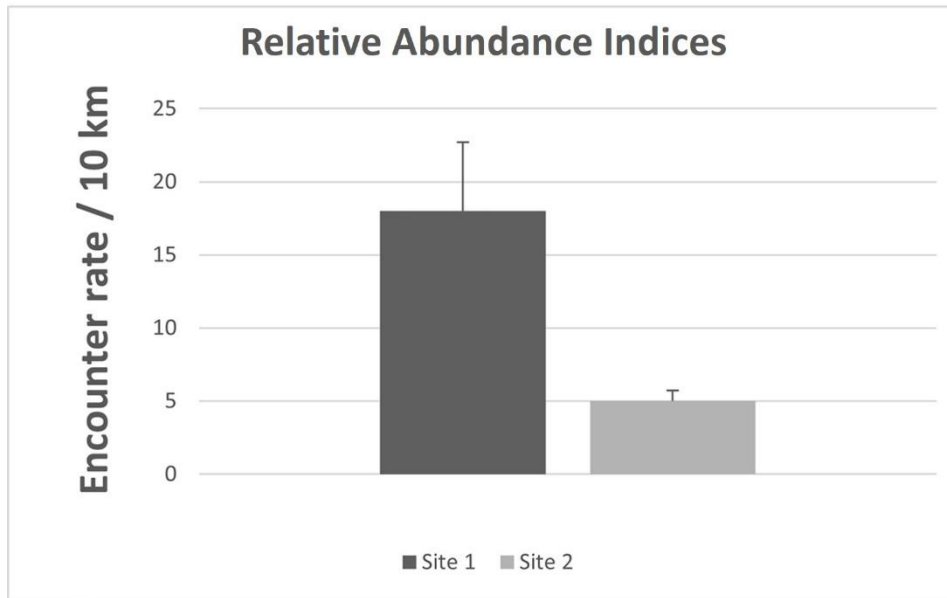


Figure 5. Encounter rate in Site 1 (in the vicinity of the Garbage dump) was higher compared to Site 2 (in the vicinity of the town of Zapotitlan Salinas).

A comparison was made between all the monitoring methods used and their characteristics according to the methodology that should be particularly considered when applying them and during the design of any given sampling of FRD and FD populations. Criteria were assigned based on the practical purposes of application, where it is shown that camera trapping is the most versatile to obtain population density, abundance and distribution indicators with respect to other methods. The natural marking method was the one that showed the most advantages with respect to all the others, considering the criteria of application level, execution time, sampling effort, data processing effort, precision level and cost. Therefore, this analysis gives us a perspective on which method may be more suitable for the execution and the effort required in other studies (Table 3).

METHODS	INDICATOR	AL	ET	SE	DPE	LK/US	LP	PEOPLE	COST
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<b>CAPTURE-MARK-RECAPTURE</b>	density abundance	easy	1 month	4 days	20 – 30 days	high	high	2	low- average
<b>CAMERA TRAPS</b>	density abundance distribution	difficult	8.5 month s	7 months	30 – 45 days	medium	high	2 - 3	high
<b>FWLT</b>	density abundance	moderate - difficult	8 months	1 month	45 – 60 days	high	medium	2	low- average
<b>VWLT</b>	density abundance	moderate - difficult	7 months	1 month	30 days	high	medium	2	low- average

Table 3. Comparison of monitoring methods used during this study to estimate population indicators of FRD and FD, through application criteria. Application level (AL), execution time (ET), sampling effort (SE), data processing effort (DPE), level of knowledge and use of software (LK/US), and level of precision (LP).

## Discussion

FRD and FD monitoring is a key component for decision-making on the management and control of invasive species populations in PNA. However, many monitoring methods are often unsound, poorly designed, and not targeted at management interventions, complicating the decision-making process. Robust estimations of population parameters are needed to manage FRD and FD, and to develop quantitative ecological, economic, and social impact assessments. In addition, strong monitoring programs may provide valuable insights that help direct efforts to prevent and contain FRD and FD. Most information on dog impacts comes from owned, FRD populations in urban and peri-urban areas (Young et al. 2011; Zapata-Ríos and Branch 2016). Unlike these, rural and wild sites belonging to the TCBR were chosen in this study, where the FRD populations are without demographic closure (open) and possibly some of these might be true feral dogs.



The capture-mark-recapture methodology conducted in the city of Mumbai, India, with a closed population of FRD, and which can be used at variable scales, especially in the absence of artificial marks, was used in the present study as a basis (Punjabi et al. 2012). A robust sample size was obtained from an open population of FRD and FD in 10 hectares of a garbage dump. However, although individual identification through photographic comparison is low cost, it takes time (Tiwari et al. 2018). An added disadvantage of this method is that it can be difficult to use in places where the vegetation is closed or abundant and does not allow individuals to be observed and photographed carefully. The POPAN model was used, which was adequately adjusted to the type of data in MARK©, as the population presented emigration and immigration, emulating the parameterization of the Jolly- Seber model, in terms of a metapopulation (N).

Camera trapping is a method widely used in the world for the monitoring of terrestrial vertebrates. Camera traps with a robust and open design of the mark-recapture model (Kendall and Bjorkland 2001; Kendall 2004) have been used to estimate the number of free-ranging dogs in protected areas located in the Atlantic Forest biome in Brazil (Paschoal De Oliveira 2016). Unlike studies using the camera trap method in urban or peri-urban areas, the RAI was obtained from an open population of FRD and FD from two sites, in a detection - non-detection model, which was found to be of less statistical difficulty and requires less time in photo processing. However, this will depend on the number of photos and analysis design. Despite this, it is still an expensive method, since camera traps in large quantities are usually expensive. Some studies recommend that, between 25 to 35 camera traps should be installed (Kays et al. 2020). In the present study, 35 camera traps were used over 30 days, obtaining precise estimates of the RAI with a robust amount of data. It is recommended to perform further studies in different seasons, since this could impact the populations and mobility of FRD and FD.

In the study of the FRD and FD populations, the method of fixed-width line transects has been used, through the identification of tracks, comparing the abundance in relation to wildlife between sites (Reyna-Hurtado and Tanner 2005; Zapata-Ríos and Branch 2016). A disadvantage of this method is that experience is needed in the use of track identification guides (footprints and excreta), which allow us to make a precise identification. Another disadvantage is the sample size, for example, the one obtained in the present study was small ( $n < 60$ ) for each group of tracks, due to

this, some statistical software (e.g. Distance v.7.2) does not allow the precise execution of ideal statistical models (Zapata-Ríos and Branch 2016).

One of the most used methods to study population densities of terrestrial mammals are variable-width transects (Atickem et al. 2010; Narváez and Zapata-Ríos 2020). However, it was the least convenient in the present study. It presented difficulties over the other methods in sample-taking, since the FRD and FD are quite elusive (Gompper 2014), overly cautious, with well-developed senses, which put the observer at a disadvantage. In addition, during this investigation there were abundant atypical rains and excessive growth of bushy vegetation, preventing the observation of FRD and FD. Therefore, this method is suggested to be applied in open and semi-open spaces where the observation of free-living individuals can be carried out properly. The RAI was estimated through the encounter rate times the distance traveled, as an alternative to using software when small samples are due to estimate the RAI.

Effective abundance estimation efforts will help inform where FRD and FD are most abundant, most likely to spread, or most likely to be contained. This study compares different criteria of four sampling methods for FRD and FD, which contributes to expanding the information regarding the methods used to determine relative abundance indices. Overall, the results of this study show that although there are different methods for monitoring FRD and FD available, they must be effective in terms of time and cost. Therefore, the use of the capture-mark-recapture (C-M-R) methodology (Punjabi et al. 2012) is recommended for abundance estimation of open populations of free-ranging and feral dogs of relatively easy individual identification.

In the study conducted in a milk-producing village in Mumbai, India through the C-M-R method based on natural marks, the abundance of a closed population of FRD was estimated with 680.64 PLR in 1280 ha (Punjabi et al. 2012). In contrast to the previous study, the abundance of an open population of FRD and FD in a landfill inside the TCBR was estimated with the same method, obtaining 239.8 FRD and FD in only 12.5 ha, with a greater number of FRD and FD in a smaller surface unit. Clearly, human subsidy provided by the daily provision of tons of human waste at the garbage dump (within the NPA), exacerbate the presence of FRD and FD. Several studies have shown that the presence of FRD and FD in NPA has negative effects on wildlife (Torres and Prado 2010), as well as affecting their activity patterns and abundance (Zapata-Ríos and Branch 2016). This study was no exception; a total of 12 terrestrial wildlife mammalian species were recorded, of

which only four were recorded at Site 1 and all species at Site 2. Therefore, this subsidy consequently puts wildlife within the TCBR at a clear disadvantage perhaps being at greater risk of predation, harassment and displacement by FRD and FD. Therefore, there should be no garbage dumps within any NPA and surveillance of clandestine rubbish dumps and proper management of human waste should be improved to prevent human subsidized invasive species.

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## **Author Contributions:**

All authors contributed to the study conception and design. Material preparation, data collection and analysis were performed by Orlando Reina-Ponce and Manuel Weber. The first draft of the manuscript was written by Orlando Reina-Ponce and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

## **Data availability:**

The datasets generated during and/or analyzed during the current study are not publicly available due institutional postgrad thesis & data privacy policy until student graduation but are available from the corresponding author on reasonable request.

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## Conclusiones

### a) Comparación de métodos.

Este estudio compara diferentes criterios con base a las finalidades prácticas de aplicación, lo cual contribuye a ampliar la información con respecto a los métodos utilizados para determinar índices de abundancia relativa de PLR y PF. Sin embargo, los resultados de este estudio reflejan que a pesar de existir diferentes métodos para el monitoreo de PLR y PF, estos métodos también deben de ser efectivos en términos de tiempo y costo. Por lo anterior se recomienda el uso de la metodología C-M-R, para poblaciones abiertas de fácil identificación individual en espacios abiertos.

### b) Índices de Abundancia Relativa (IAR)

Los IAR obtenidos en este estudio reflejan que es importante conocer cómo está estructurada la población de PLR y PF y cómo se mantiene (subsidio humano). La abundancia es un atributo de la población que varía con el tiempo y en el espacio. Asimismo, requiere de la obtención de información utilizando recursos mínimos, explotando las características del entorno, y así maximizar los recursos para la intervención. Se recomienda realizar el estudio en diferentes estaciones del año, dado que esto podría tener un impacto sobre las poblaciones y la movilidad de PLR y PF. Se recomienda realizar futuros estudios sobre la dinámica de las poblaciones de PLR y PF al interior de la RBTC, para establecer las tasas de natalidad y mortalidad, por lo anterior estudiar el tiempo de recambio poblacional. De igual manera, extrapolar este estudio a diferentes ANP del país para investigar la dinámica poblacional de PLR y PF, y poder manejar indicadores que analicen el impacto que han tenido los PLR y PF sobre la vida silvestre

### c) Medios de subsidio para PLR y PF.

Los PLR y PF tienen una flexibilidad dietética alta, demostrando que los rellenos sanitarios y tiraderos de basura al interior de la RBTC, son fuentes de subsidio para los PLR y PF. Debido a ello, son más abundantes en los alrededores de este. Sin embargo,



el gradiente de densidad poblacional humana y actividades de origen antrópico en zonas rurales demuestra que estos influyen fuertemente en la distribución de los PLR y PF. Por añadidura, son la fuente de las poblaciones de PLR y PF en vida libre. Los resultados reflejan diferencias en la abundancia y distribución de PLR y PF entre los sitios. Por lo anterior, se ha favorecido la ocupación de PLR y PF, así como el desplazamiento de las especies silvestres en estas zonas, posiblemente debido a diferencias en el hábitat y cambio de uso de suelo, principalmente para ganadería. Se recomienda la vigilancia para evitar estos sitios de subsidio, eliminación de tiraderos y rellenos sanitarios al interior de ANP, así como acciones en el control y remoción de PLR y PF.

d) Implicaciones para la conservación de la vida silvestre.

Las poblaciones de PLR y PF en vida libre pueden transmitir enfermedades a especies silvestres. Además de fungir como depredadores y competidores de varias especies. Las abundancias de PLR y PF obtenidas en este estudio, están por encima de las especies silvestres, poniéndolas en una desventaja a estas, por el subsidio humano. La riqueza de mamíferos se ve afectada por la presencia de los PLR y PF. Por ello, es importante realizar estudios sobre los impactos que generan los PLR y PF en la fauna silvestre. Además de la depredación, como las enfermedades transmitidas y que son en la mayoría de las ocasiones letales en la fauna silvestre. De igual manera, realizar estudios sobre las consecuencias que su sola presencia puede causar en la rutina de un animal silvestre.

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