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Comparing the effectiveness of tracking methods for medium to large-sized mammals of Pantanal

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ABSTRACT. Most Neotropical mammals are not easily observed in their habitats, and few studies have been conducted to compare the performance of methods designed to register their tracks. We compared the effectiveness of track registry between sand plots and two tracking methods that use artificial materials to record tracks: the sooted paper, and the plastic board methods. The latter is described here for the first time. From 2002 to 2005, we conducted two experiments in three study sites in the Pantanal region of Brazil. We compared the artificial methods with the sand plot by registering track presence/absence, the number of identifiable tracks, and the total number of tracks (identifiable and unrecognizable) in each tracking plot. Individuals avoided artificial tracking plots either by not stepping on them or by doing it fewer times than on the sand plots. The use of artificial materials to register mammal tracks resulted in underestimates that are especially relevant to short-term ecological studies. We recommend the use of the traditional sand plot method whenever possible and the development of detailed studies on the efficiency of artificial methods under a variety of environmental conditions and time lengths. Despite their relatively lower efficiency, we believe that artificial methods are useful under specific conditions and may be more efficient if used in more comprehensive sampling efforts.

KEY WORDS. Mammals; Pantanal; plastic board, scent-station; track-plates.

Many mammal species are not easily observed in their habitats. Among non-invasive methods proposed to overcome this limitation, tracking traps have been one of the most widely used. Track registry has been largely used in ecological studies of wild mammals to estimate mammal abundance, density, distribution, and richness (Justice 1961, Marten 1972, Schaller 1980, Travaini *et al.* 1996, Wilson *et al.* 1996, and others). It is one of the oldest methods for the identification of medium-sized to large mammals (Becker & Dalponte 1991).

Among the several techniques proposed to obtain tracks, the sand plot (usually called scent-station when baited) is the most used. Sand plots consist basically of a plot of fine soil to record tracks. After the development of the sand plot technique, some new tracking methods have been proposed. Kymograph paper (Sealander *et al.* 1958), toner or talcum powder applied over contact papers (Mayer 1957), and carbon-sooted aluminium plates (Barrett 1983, Raphael *et al.* 1986, Taylor & Raphael 1988) have been used to record tracks of small rodents and medium-sized carnivores. Nevertheless, little is known about their performances, and many of them are susceptible to adverse weather conditions. Smoked aluminium surfaces, kymograph paper, and sand plots are usually damaged by rain (Conner *et al.* 1983, Nottingham *et al.* 1989, Diefenbach *et al.* 1994, Mabbe 1998), and sand plots may dry during warm days before animals step on them, which may compromise track registry. In addition, track stations made of print ink board are impractical for registering tracks of large mammals (Ratz 1997, but see Palma & Gurgel-Gonçalves 2007).

Mabee (1998) described a tracking method for small mammals based on tracking tubes designed by Merriam (1990) and Van Apeldoorn *et al.* (1993) which withstand wet environmental conditions. Different kinds of covers have been proposed to protect tracking plots from precipitation, such as cages or plastic sheeting to host the tracking plots (Zielinski & Kucera 1995, Loukmass *et al.* 2002, Baldwin *et al.* 2006, Palma & Gurgel-Gonçalves 2007). However, they are overall impractical, especially if they need to be large enough to protect tracking plots designed for medium-sized to large mammals. Besides the limitations of these methods, their adequacy for use in particular habitats, weather conditions, and different taxa can only be assessed by systematic experiments that test their efficiency and point out their advantages under each condition. Nevertheless, a few studies have compared the performance of methods designed for track registry (Loukmass *et al.* 2002, Belant 2003, Sargeant *et al.* 2003, Baldwin *et al.* 2006, Gompper *et al.* 2006). Therefore, it is necessary to test the efficiency of tracking methods, and to find alternative methods to overcome their pitfalls.

We have experimentally compared the efficiency of two artificial tracking methods against sand plots: the first method is called plastic board and is described here for the first time; the second method is the sooted paper method. We compared the track presences/absences, the total number of tracks and the number of identifiable tracks between sand plot and artificial tracking plots to investigate the effectiveness of the artificial methods. We expected a greater number of total tracks and track presence on sand plots, since some species may be wary of, and avoid stepping on, plastic board or sooted paper, due to their shape or smell. We also expected the performance of each method to vary according to environmental conditions. We anticipated a greater number of identifiable tracks on the plastic board, once it is protected against rain and it is not affected by dry weather conditions. Sooted paper and sand plot methods were expected to perform in a similar way under wet conditions, as both are susceptible to damage by rain. However, sooted paper would not be affected by dry conditions and therefore, may perform between under such condition.

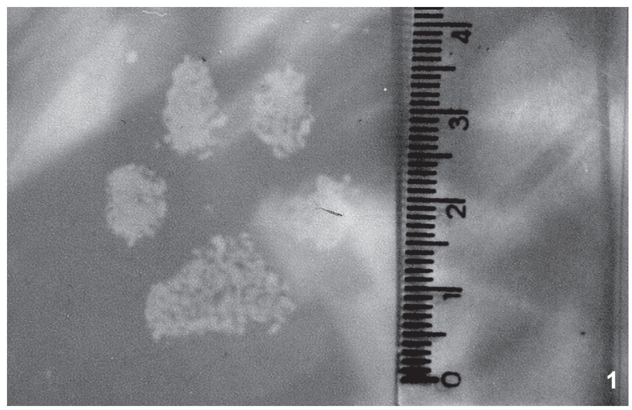
MATERIAL AND METHODS

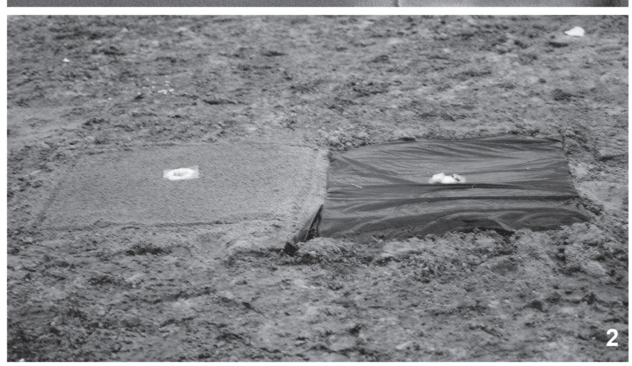
The comparison between techniques took place in three areas located in the Nhecolândia sub-region of Pantanal, Brazil: Alegria ranch (19°08'S, 56°49'W), Nhumirim ranch (18º59'S, 56º39'W), and Rio Negro ranch (19°34'S, 56°14'W). The Pantanal is the world's largest seasonal floodplain. This region is characterized by sandy soil with a mosaic vegetation of semideciduous forest, dispersed shrub vegetation, and seasonally flooded fields (Rodela 2006). Several permanent and temporary ponds and "salinas" (brackish water ponds) are present. Human population density is low (< 2 people per km^2) and the main economic activity is cattle ranching (Adamoli 1987). The Pantanal has a high diversity and density of medium-sized to large mammals (Alho *et al.* 1988, Mittermeier *et al.* 1990, Alho & Lacher 1991), which makes it an adequate area for this study.

The three study sites differ mainly in land use: the Rio Negro ranch is a preserved area located at the margin of the Negro River, where cattle ranching is absent. Alegria ranch and Nhumirim ranch are neighbour ranches located in an area without major rivers nearby. Nhumirim ranch is a research station of the Brazilian Agricultural Research Corporation (Embrapa) where there is a preserved area, but also pasture lands for cattle ranching; Alegria ranch is predominantly composed of pastures in which cattle ranching is the main economic activity.

The new tracking method consists of two overlapping plastic sheets. The plastics are commonly used together in a toy named "magic board" (we have adopted the name "plastic board" to describe this new method), and its commercial name is Lamicel© (CIPATEX). This is a synthetic laminated plastic used in baby carts, bag covering, plastic paints, toys, domestic and office utensils, and can be obtained from companies working with plastic manufactures.

The upper sheet is made of a fluorescent pink plastic, and the lower one is a white sheet. Both sheets are 0.2 mm thick. When the animal steps on the trap, the upper sheet is pressed against the lower one and the tracks become visible (Fig. 1). Track images remain visible until one separates the plastics by pulling them apart. To protect the plastic board from rainfall, we covered it with a thin light-brown plastic. The plastics were then attached with adhesive tape to a basis made of a hard material that is able to account for substrate irregularities (usually a thin light metal or hard plastic sheet).





Figures 1-2. (1) Track of *C. thous* on the plastic board plot and (2) tracking station showing a sand plot and a plastic board placed side by side in 2002 at the Rio Negro ranch, Pantanal, Brazil.

The other method tested was the sooted tracking paper. We used sheets of glossy paper coated with soot from a kerosene-camphor torch (15 g per kerosene litre; adapted from Barret, 1983, Taylor & Raphael 1988, Heske 1995).

From July, 2002 to June, 2005, we developed two experiments in the three study sites. The performance of the artificial methods was tested by comparing their effectiveness against the sand plot method in a total of six expeditions to Alegria ranch (March 2003), Nhumirim ranch (June 2005), and Rio Negro ranch (July-August 2002, February 2003, May 2003, and October 2004). Tracking stations with the plastic board were established in Rio Negro (2002 and 2003) and Alegria ranches (2003), whereas sooted paper stations were placed in the Rio Negro (2004) and Nhumirim (June 2005) ranches.

We established a total of 69 tracking stations spaced at least 200 m apart, each composed of two 0.49 m^2 tracking plots (70 x 70 cm; Fig. 2). One of the plots was either set with the plastic board, or the sooted paper, and the other was a sand plot made of sifted and moistened sand. We used several different baits, such as bacon, meat, roots, seeds, fruits, and salt placed in the centre of each tracking plot; bobcat, fawn, and rabbit urines were also used as attractants. The same type of bait or attractant was used in each pair of tracking plots. The tracking stations were checked every day and damaged plots were fixed or replaced; baits or attractants were renewed as needed. We set up to seven tracking stations a day. Each remained at the same site for a maximum of five days. By frequently moving the tracking station sites, we maximized the number of habitats sampled, avoiding occasional trap-happy resident individuals. Track stations were set in different habitats, including open areas, forest, edge of salinas, and ponds, and along dirty roads.

Tracks obtained on the plastic board were photographed and copied by overlapping and outlining the tracks on an acetate sheet; those obtained on sooted paper were photographed, cut away and laminated. Tracks were then identified by comparing them with a reference collection of footprints obtained on the plastic board from mammals of the Rio de Janeiro's Zoo (Fundação RIO-ZOO) and with track field guides for Brazilian mammals (Becker & Dalponte 1991, Borges & Tómas 2004).

To investigate the relative efficiency of the methods in recording species tracks, we counted the number of identifiable tracks (those judged as clearly recognizable) on each tracking plot and used them to compare between sand plots and artificial tracking plots (either the plastic board or the sooted paper) using Wilcoxon matched pairs test (Siegel 1977). Lost records such as plots damaged by adverse weather conditions or animal interference were included in this particular analysis because we understand that they reflect the limitations of the methods being compared. The Wilcoxon test is a non-parametric test designed for dependent samples, which are the two plots of a tracking station in this case. Therefore, variables not directly linked to the two plots being compared (*e.g.* different baits used between stations) do not represent a source of error in the comparison. The only variables that must be controlled are those directly related to the two plots being compared in a tracking station (*e.g.* plots of the same size and baited with the same bait or attractant). Moreover, given that our experiment was designed for a pair wise analysis (sand plot *vs.* plastic board and sand plot *vs.* sooted paper), a three-way comparison using a Kruskal-Wallis test, for example, would not be adequate for this dataset. To investigate whether some species avoid the plastic board or sooted paper methods, we compared the frequency of track presence/absence on pairs of tracking methods using the McNemar test (Siegell 1977), and the total number of tracks (identifiable and unrecognizable) using the Wilcoxon matched pairs test.

An additional way of evaluating the degree to which species are wary of the artificial methods is to compare the observed frequency of animals visiting the plastic board or the sooted paper with the "expected frequency" of visits, after an animal has been detected in a tracking station. Therefore, we selected the tracking stations where animals occurred and compared the observed frequency of tracks on artificial plots with the expected frequency using Chi-square tests. The presence of an animal at a station was determined by its tracks on any tracking plot of a station. The expected frequency of tracks on the artificial plots was expected to be 50%, i.e., once present at a tracking station, the probability of an individual stepping or not on an artificial plot was assumed to be the same. For those stations that remained in the field for a total of five consecutive days, we presented cumulative species richness curves for the sand plots and the alternative methods using a diversity rarefaction analysis (PAST 1.99 software, Hammer *et al.* 2001).

When more than twenty tracks were found on a tracking plot, the total number of tracks was rounded off to the nearest ten; we did this to compensate for the increased error probability when counting a large number of tracks. Whenever possible, registries of the most abundant species – the crab-eating fox, *Cerdocyon thous* Smith, 1839 and the agouti, *Dasyprocta azarae* Lichtenstein, 1823 – were analyzed separately. Data for the remaining species were clumped and analyzed together due to small sample sizes. Since the occurrence of these species on the tracking stations was somewhat rare, the effect of each on the test result was similar.

We considered $\alpha = 0.05$ for large samples sizes (*C. thous*, $N > 70$). For the remaining comparisons, $\alpha = 0.10$, given the small sample sizes ($N < 30$) and the relatively low power of the analyses (*e.g.* Power = 73% for N = 30 and medium effect size in a two-tailed comparison between number of tracks using Wilcoxon test – Faul *et al.* 2007).

RESULTS

We obtained 138 records (number of species per track station-night) of 11 medium to large-sized mammal species (Tab. I) on a total of 173 tracking station-nights. Fourteen mammal track records could not be identified at the species level but were included in the statistical analysis. Tracks of young *C. thous* are usually undistinguishable from those of the hoary fox, *Lycalopex vetulus* Lund, 1842. Nevertheless, because *L. vetulus* is rare or absent in the studied areas, we considered all tracks as being of *C. thous* in the analysis.

Table I. Mammal species registered in each tracking station-night placed in the Pantanal region, Brazil over 2002-2005. Species richness (number of species) and non-identifiable track records are also shown.

	Sand plot	Plastic board	Sand plot	Sooted paper
Carnivora				
Cerdocyon thous	82	43	7	2
Leopardus pardalis	2	2	0	0
Procyon cancrivorus	1	1	0	0
Nasua nasua	3	6	0	0
Speothos venaticus	1	0	0	0
Eira barbara	0	0	1	1
Rodentia				
Dasyprocta azarae	21	14	6	5
Hydrochoerus hydrochaeris	3	0	0	0
Perissodactyla				
Tapirus terrestris	1	0	0	0
Artiodactyla				
Tayassu pecari	0	0	7	5
Edentata				
Dasypus sp.	1	1	2	2
Unindentifiable tracks	15	12	4	2
Total	130	79	27	17
Species richness	9	6	5	5

Overall, individuals were wary of artificial tracking plots. They either did not step on them at all, or did so less often than on sand plots (Tab. II). Additionally, the number of identifiable tracks for *C. thous* was more than two times greater on sand plots than on plastic boards; the number of identifiable tracks was also greater on sand plots than on sooted papers (Tab. II). On the other hand, once an individual was present at a station, its probability of stepping on an artificial tracking plot was either higher than or equal to not doing so: about 67% of the individuals present at a station stepped on the plastic board and 74% stepped on the sooted paper (Tab. III).

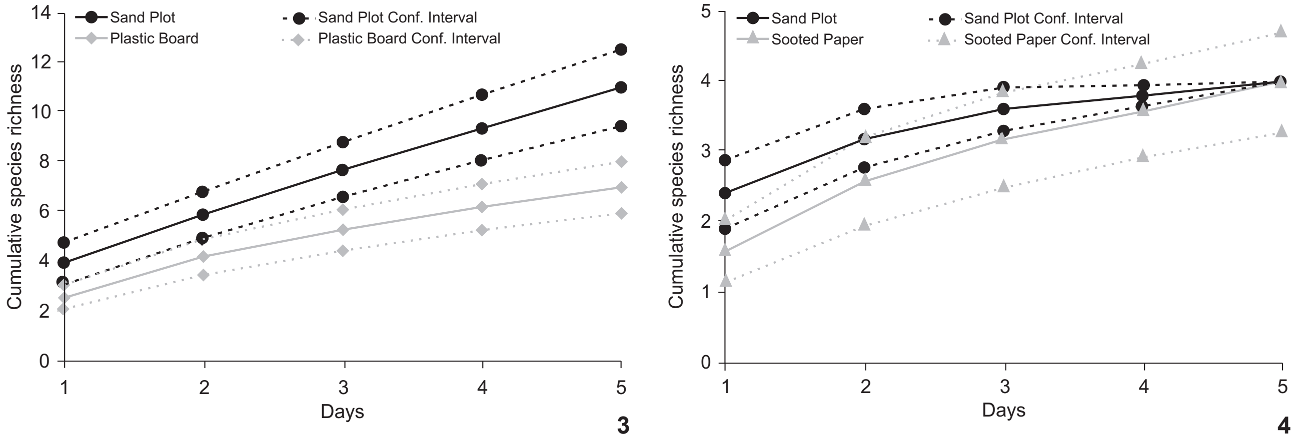
Table III. Number of track presences and absences on artificial tracking methods, given the occurrence of animals in the tracking stations. Observed track presences on artificial methods were compared with the expected frequencies (50%) using Chi-square test. The occurrence of an animal at a station was determined by the presence of tracks on any tracking plot of a station.

Comparison		Track		p
		Presences	Absences	
Plastic board	*C. thous*	49	19	< 0.001
	D. azarae	14	8	0.201
	Other species	9	8	0.809
Sooted paper	Species total	17	6	0.023

Species richness calculated over artificial methods tended to increase with time (Fig. 3), but such increase was slower than with sand plots. When comparing species richness during five days of sampling, the number of species registered on sand plots was always higher than on plastic boards, but not different from sooted paper (Fig. 4), probably due to small sample sizes in this last comparison.

Table II. Mean (SD) total and identifiable tracks obtained in sand plots and artificial plots (plastic board or sooted paper) placed in the Pantanal region, Brazil over 2002-2005. Track presence was the total number of records (number of species per track station-night) obtained on each type of tracking trap. Total number of tracks and number of identifiable tracks were compared using Wilcoxon matched pair test; track presence was compared between pairs of tracking traps using McNemar test. Whenever possible, data on *C. thous* and *D. azarae* were analyzed separately from other species listed in Table I. n = sample size.

Comparison		Total tracks	n	Identifiable tracks	n	Track presence	n
Sand plot vs. Plastic board	*C. thous*	23.3 (18.0) vs. 6.2 (9.9)	71	5.3 (9.5) vs. 2.1 (3.7)	75	73 vs. 55	74
		Z = 7.13; p < 0.001		Z = 4.54; p < 0.001		χ^2 = 14.45; p = 0.001	
	D. azarae	11.9 (10.9) vs. 7.5 (10.8)	22	4.5 (5.0) vs. 2.6 (3.7)	22	21 vs. 14	22
		Z = 2.31; p = 0.021		Z = 1.18; p = 0.237		χ^2 = 2.08; p = 0.149	
	Other species	12.3 (15.3) vs. 10.5 (20.0)	16	2.3 (2.9) vs. 2.3 (5.6)	24	16 vs. 09	16
		Z = 2.48; p = 0.013		Z = 1.48; p = 0.140		χ^2 = 3.27; p = 0.070	
Sand plot vs. Sooted paper	Species total	16.4 (17.3) vs. 8.0 (13.4)	28	4.6 (5.1) vs. 2.0 (4.2)	26	14 vs. 10	17
		Z = 1.73; p = 0.084		Z = 1.74; p = 0.082		χ^2 = 0.44; p = 0.505	



Figures 3-4. Cumulative species richness and confidence intervals obtained with tracking stations that remained in the field for 5 consecutive days over 2002-2005 in the Pantanal region, Brazil; (1) sand plot vs. plastic board (N = 15) and (2) sand plot vs. sooted paper (N = 5).

DISCUSSION

This is one of the few studies attempting to compare the relative efficiency of tracking trap methods for medium-sized to large mammals (see also FORESMAN & PEARSON 1998, HARRISON *et al.* 2002, LOUKMASS *et al.* 2002, GOMPPER *et al.* 2006, BAREA-AZCÓN *et al.* 2007). Despite the small sample sizes, the differences between tracking methods were evident: sand plots performed better than artificial methods in 2/3 of the comparisons (Tab. II). Individuals were generally reluctant to step on the plastic board or the sooted paper.

Many medium-sized to large mammals, especially carnivores, have an outstanding sense of smell or sight, which allows them to perceive artificial materials in the environment. It is not surprising, therefore, that the canid *C. thous* avoided the plastic board. The same behaviour seems to occur in other mammalian taxa; BALDWIN *et al.* (2006) have found that bobcats and coyotes avoided sites where scent stations were covered by plastic sheets. Likewise, GOMPPER *et al.* (2006) found that some grey and red fox individuals were not detected by track plates at the studied site, although they were registered by cameras traps (but see BULL *et al.* 1992), and SARGEANT *et al.* (2003) found that swift foxes visited sand stations 2.4 times more frequently than track plates.

Despite species wariness, we noticed that once an individual was detected at a tracking station, the probability of it stepping on the artificial plot was always equal to or greater than the probability of it not doing so (Tab. III). Therefore, we believe that species richness underestimates generated by the utilization of artificial methods are not too severe and may be minimized by keeping artificial track plots in the field for extended lengths of time, so that animals can get used to them. In fact, GOMPPER *et al.* (2006) noticed that each species requires an acclimation period that precedes its willingness to step on track plates; for instance, it is necessary to place track plates for about 30 days in the field before the probability of capturing raccoon and mustelid tracks reaches 100%. Furthermore, we observed that species richness obtained with artificial tracking methods tended to increase even during a short period of time (five days). However, it was not possible to provide a minimum time length necessary to attain reliable estimates of species richness.

Despite the overall species wariness with respect to artificial methods in this study, the same may not be true for studies carried out in other regions or involving other species. Species may respond differently to distinct census techniques (see BALDWIN *et al.* 2006, GOMPPER *et al.* 2006, BAREA-ASCÓN *et al.* 2007), and there is still a need for testing different tracking techniques, not only under different environmental conditions, but also for different taxa. Due to our small samples, we had to clump data on some species, which prevented us from detecting additional interespecific variation on the degree of wariness with respect to the methods employed. However, differences are obvious between crab-eating foxes and agoutis, with agoutis being apparently less wary of artificial methods than foxes (Tab. II).

In half of the comparisons, the number of identifiable tracks on artificial methods was lower than on sand plots. This is an additional indication that sand plots perform better than artificial methods, since species identification relies on identifiable tracks. However, in the remaining two comparisons, the number of identifiable tracks obtained with artificial methods was not different from sand plots (Tab. II). Moreover, one actually needs just a single or a few good tracks to identify most of the medium and large mammalian species. In this sense, artificial methods are useful, especially when utilized for extended periods of time. In addition, there were few rainy days during the sampling period, since most of the study took place during the dry season. The plastic boards might have performed better than the sand plots in wetter conditions because they were protected from rain.

The plastic board can be easily protected from precipitation and it is inexpensive, reusable, and lightweight. Moreover, the upper fluorescent plastic sheet can be obtained in different colours (*e.g.* grey or green) and thickness, which might result in better track record efficiency. Therefore, despite species wariness, the plastic board may be a good option under specific conditions, such as extremely wet weather or when track plots cannot be checked every day. Its efficiency under such conditions remains to be tested.

We have found that mammal species respond differently to the tracking method used. We believe that artificial tracking methods are disadvantageous in short-term surveys because individuals and species tend to need time to get used to them. In long-term studies, however, these methods seem to yield robust estimates of species richness. We encourage further studies comparing tracking methods under different weather conditions, biomes, time lengths, and for different taxa, so that researchers can select the best method under specific environmental conditions and for particular species. As noticed by Gompper *et al.* (2006), the use of non-invasive surveying techniques is wide and is increasing, which highlights the importance of studying their limitations and biases.

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