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in Animal Care

Methodological study on different recording rules applied to the behavior of a male captive jaguar (*Panthera onca*)

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Abstract

This study on a seven-year-old male jaguar, at the Ambue Ari Sanctuary in Bolivia, was performed to compare different recording rules. Starting from videos recorded by camera traps over the period of a month, a much smaller sample of these was selected. Later the animal behavior was analyzed with both continuous and instantaneous sampling (at 5-second intervals). The aim was to compare the differences these two methods can produce in the overall results. The average correlation obtained for all the categories was 0.941 (P-value<0.001; two-tailed), and considering all the single behaviors, it was 0.932 (P-value<0.001; two-tailed). This brings us to the conclusion that the two used methods are highly correlated, and they can subsequently be interchanged. An analysis was carried out also considering the presence and absence of people, and it highlighted into which data collection method (camera traps or direct recordings) is more appropriate for the kind of behaviors one wants to investigate.

Introduction

1. Jaguar's biology

The largest cat in the Americas is the jaguar (*Panthera onca*), and the third largest in the world. Its range extends from the southern part of the USA to Argentina *(Dobbins et al., 2017)*, and it is the only living member of the genus *Panthera* native to the Americas. Supposedly, the progenitors of the American *Panthera* species migrated from Asia into America 10.8 million years ago *(Johnson et al., 2006)*. It is characterized by a pale yellow to tan colored fur, covered by spots that transition to rosettes on its side. The jaguar is particularly active during twilight hours, but it has seen outside this timespan not engaging in hunting activities *(Maffei et al., 2002)*. Its body length can reach 1.85 m and it can weigh up to 158 kg, but it is the power of its bite – 681.56 Newton - that makes it remarkable *(Del Moral Sachetti et al., 2011)*. Its predation style is opportunistic, but its preferred feed is medium to large mammalian prey species, such as peccaries, deer, armadillos, as well as reptiles *(González et al, 2002)*. Rural human populations close to their natural range cause the depletion of their prey base, ultimately causing to the decline of the local population *(Cuyckens, Falke and Petracca, 2014)*. In fact, the jaguar's population has experienced a 20-25% decline over the past three generations, and for this reason, it is classified as Near Threatened on the IUCN Red List *(Quigley et al. 2017)*. The direct conflict with humans for wild meat and livestock, habitat loss and fragmentation, and commercial hunting, result in isolation of the populations and lower survival rates *(Quigley et al. 2017)*. Jaguars hold an extremely important ecological role, as they are considered an umbrella species, whose protection indirectly influences others species' protection *(Cuyckens et al., 2014)*. Studies regarding the jaguar as a species are mostly related to its biology and ecology *(Fontes et al., 2021)*, and behavioral ethograms are scarce among the most known academic databases *(Stanton et al., 2015)*.

2. Instantaneous and continuous sampling

In order to record a behavior, a recording rule has to be decided. To *Martin and Bateson (2007)*, there are two basic types of rules, each of which has a different aim: time-sampling and continuous recording. Time-sampling, which is further divided into instantaneous sampling and one-zero sampling *(Martin and Bateson, 2007)*, was used extensively in ethology, as it is a simple observational technique that can be used in a wide range of settings *(Tyler, 1979)*. The observer is asked to determine the presence or absence of a behavior within brief and regular intervals, without considering the duration and frequency of it *(Mann et al., 1991)*. On the other hand, continuous sampling consists in the recording of different behaviors, together with their frequency, percentage of time spent, bout length and eventually information on sequences of behavior *(Tyler, 1979)*. This type of sampling aims at giving an instance of events - behaviors occurring singularly - and states - behaviors which are determined by a start and an end - *(Martin and Bateson, 2007)*. The downside of it is that, although continuous recording can provide much information about the behaviors, it is extremely demanding for the observer *(Martin and Bateson, 2007)*. Some studies have suggested that the implementation of instantaneous sampling is valid, taking into consideration certain variables, such as the sample intervals *(Chen et al., 2016)*. In others, continuous sampling is considered a useful tool for captive animals *(Munita et al., 2016)*. The implementation of either method is eventually influenced by the species, the number of individuals and the focus behaviors, but a standardized method would help the enlargement of sample sizes thanks to cross-institutional collaborations. This would ultimately lead to a better recognition of the factors influencing a certain behavior and the implementation of ways to increase the welfare of captive animals *(Bernstein-Kurtycz et al., 2022)*.

Aim

Similar studies investigating the difference and the correlation between sampling methods have been published, but they lack in quantity. In particular, *Munita et al. (2016)* compared the same two sampling methods in a captive lactating female cheetah (*Acinonyx jubatus*), highlighting the presence of newly described behaviors, but no correlation between the methods was found. *Quirke et al. (2012)* studied the time necessary for the proper evaluation of the effects of environmental enrichment on eight cheetahs, concluding that 30-45 minutes of daily data collection with the instantaneous sampling was sufficient to get an insight into the effectiveness. *Chen et al. (2016)* worked on dairy cattle with a very narrow ethogram, validating the use of sample intervals ≤5 minutes for the determination of the total amount of time spent exhibiting each questioned behavior. More recently, *Bernstein-Kurtycz et al. (2022)* investigated how varying methods affect the quantification of pacing in bears, inferring that instantaneous sampling could capture the right data.

Drawing on these studies, our aim is to compare the results obtained with two different recording rules - continuous and instantaneous sampling with 5-second samples - when assessing the behavior of a jaguar in a controlled environment.

Materials and methods

1. Study area

The study was conducted in the Ambue Ari park (-15.66443865602221, -63.497799281872496), one of three NGOs run by CIWY (Comunidad Inti Wara Yassi). The sanctuary is located in Bolivia, near the village of Guarayos, about 350km north from Santa Cruz de la Sierra, and 200km south from Trinidad. It is a private natural reserve of 1,000 hectares, managed by twelve employees, whose mission is the rehabilitation and care of wild animals rescued from illegal trafficking.

2. Subject of the study

The animal that I worked with is a of 7-year-old male jaguar. He arrived initially at Macia, one of the three centers owned by CIWY. He arrived on September 13th 2015, but he was later moved to Ambue Ari three months later (13/12/2015). Due to poor health and lack of his mother's care, he suffered from a variety of medical problems: a skin infection, which made him lose his fur on his tail, intestinal parasites, and an eye trauma. He was operated on promptly, and after recovery, he was transported to Ambue Ari, his final destination. He was introduced to exercise and various forms of enrichment from a young age, and for this reason keeping up with his wide knowledge of all of them is a key point to properly take care of him.

3. Exhibit/experimental setting

The enclosure chosen for the present study is set deep in the jungle, about 1.2 km away from the volunteers' camp, at the limit of the reserve. It consists of a main enclosure and a management cage, which are connected by a double door tunnel, used in case medications must be provided (*see Fig. 1*). The main enclosure is hexagonal in shape, of about 200 m in perimeter, with two platforms and one shelter, not completely visible from outside the cage as they are mostly covered by dense vegetation. Only a small portion of his cage is completely cleared, and I referred to it as the "play area" through the study. That is where most of the new enrichment is put, and it is a precise part of his routine to go there, explore, and be challenged with the new toys. The management cage is smaller and includes three platforms, positioned at different levels. There is little natural vegetation there, and it is the only area artificially covered by a metal roof. Moreover, here stands his favorite of his two hay beds, and it is an area that he uses daily.

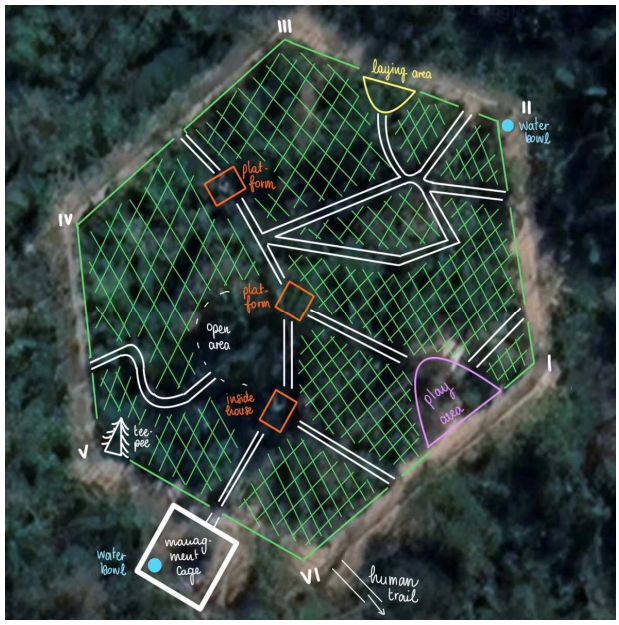


Figure 1: Picture taken from the GPS with a stylized map of the enclosure of the study subject.

4. Routine

Working with the study-subject involved sticking to a planned routine, which took place from about 10am to 12pm, every day of the week for the three months of my staying. The experience can be roughly summed up with the two most important parts: enrichment and feeding.

Past Cat Coordinators carefully planned the following routine with the president of the organization. It consisted of:

- Preparation of his meat wrapped in leaves, in a dedicated area 100 meters before the enclosure;
- Arrival at the cage, greeting and doing laps with the jaguar;
- Alternating enrichment moments with laps around the cage;
- Towards the end of the session, cleaning the main enclosure, placing the meat packages inside, doing more enrichment and opening the cage;
- Locking the jaguar in the main enclosure, clean the management cage;
- Giving access to both cages by leaving the double doors open;
- Leave.

Particular attention was paid to the proper time of food delivery. Since Kusiy is food orientated, implementing additional elements besides food was crucial for his mental, sensory and physical stimulations. For the same reason, as soon as I let him into the main cage with the food, I was neither allowed to follow him, nor interact with him as he could have become aggressive and protective over food.

The amount of food was calculated based on the body weight of Kusiy (about 80 kg), and therefore he was fed 1.5 kg of meat, six days a week with a fasting day. For enrichment and nutritional requirements, different kinds of meat on different days were provided: chicken, beef, liver, heart, kidneys. The way to deliver the food was either by wrapping it in leaves, and occasionally in a bowl with some water, to encourage the consumption of water. Twice a week, and especially the days prior to the fasting day, he received bones together with the meat. Every week, the study subject received a chicken egg. Once every two months he received some cow parts (head, skin or leg) mainly as enrichment.

When approaching his enclosure it was important to say hello, as he was able to hear me through the vegetation around his cage. Sometimes, especially after he became familiar with me, he would answer me back with some welcoming growls. As soon as I arrived there, I had to walk closer to the cage, and greet him with affection. This step was performed through the fence, and he was allowed to lick my arm and get scratched on the back. Safety is extremely important, especially with the jaguar I worked with, as his temperament is extremely excitable, which might result in a mis-reading of the behavior. During the time at the cage, I was in charge of running or walking with Kusiy around the cage. He was allowed to ask for affection during the walks, and he would express that by approaching the fence and leaning

against it with his body. Moreover, enrichment was placed in the main cage with leaves, branches, vines and perfumes, as long as it was naturally found in the environment.

5. Technical support

- Camera Nikon Coolprix B500;
- Two camera traps (brands: Browning and Bushnell);
- PC with BORIS extension.

6. Ethogram

As stated by *Stanton et al. (2015)* ethograms allow for a standardized collection of data, and their designing is crucial to any ethological study. The one developed for this study (*see Tables 1-2*) was produced by the direct observation of the subject, as well as the indirect observation of the recordings. Starting from the "Standardized ethogram for the felidae" *(Stanton et al., 2015)*, further literature review was issured. In particular, it was important to provide coherency with similar studies on felids with cheetahs *(Munita et al., 2016; Quirke et al., 2011; Skibiel et al., 2007)*, cougars, jaguars, lions, ocelots and tigers *(Skibiel et al., 2007)*. A total of thirty behaviors was described, and subdivided in eight behavioral categories.

Table 1: Ethogram produced by the observation and registered behaviors in a captive male jaguar, over a 30-day period.

Category	**Behavior**	**Definition**
Affiliative	Body rub	Cat rubs any part or entire length of body against (modifier). It usually happens when the animal is asking for attention and affection.
Exploratory	Clawing	Cat uses its paws with extended claws to move, pick or interact with (modifier). Also, claws are dragged along (modifier), likely leaving visual marks behind.
	Explore	Cat moves around attentively while sniffing, pawing or digging the ground and/or (modifiers). The aims are various: foraging, playing, curiosity for an additional element in the environment.
	Flehmen	Cat makes a grimaced facial expression, where the mouth is open, upper lip is elevated, and tongue may protrude out of the mouth.
	Lick	Cats tongue protrudes from mouth and strokes (modifier).
	Paw	Cat pats (modifier) with its forepaw(s). Usually claws are retracted.
	Play	Cat interacts with something in a "non-serious" manner (i.e. where there is no intention to harm).
	Rake	Cat makes kicking movements with one or both hind legs against (modifier).

	Roll	While lying on the ground, cat rotates body from one side to another. During the roll, the back is rubbed against ground, the belly is exposed and all paws are in the air. Cat may continue rolling repeatedly from side to side.
Locomotion	Carry	Cat picks (modifier) up and moves it to another location.
	Climb	Cat ascends and/or descends an object (resting platforms) or structure (trees).
	Jumping	Cat leaps from one point to another, either vertically or horizontally.
	Running	Forward locomotion in a rapid gait, which is faster than walking or trotting. This behavior includes also running away from and running towards something. Cat can keep watching it, but not necessarily.
	Walking	Forward locomotion at a slow gait.
Maintenance	Bite	Cat closes its teeth at and is successful in biting or chewing (modifier).
	Defecate	Cat releases faeces on the ground while in a squatting position.
	Drink	Cat ingests water (or other liquids) by lapping up with the tongue.
	Eating	Cat ingests food (or other edible substances) by means of chewing with the teeth and swallowing.
	Groom	Cat cleans itself by licking, scratching, biting or chewing the fur on its body. May also include the licking of a front paw and wiping it over one's head.
	Stretching	Cat extends its forelegs while curving its back inwards, usually followed by the arching of the back.
	Urinate	Cat releases urine, either in a squatting position, while walking or with the tail raised.
Resting	Lying	Cat's body is on the ground in a horizontal position, including on its side, back, belly, or curled in a circular formation.
	Sitting	Cat is in an upright position, with the hind legs flexed and resting on the ground, while front legs are extended and straight.
	Sleeping	Cat is lying on the ground with its head down and eyes closed, performing minimal head or leg movement, and is not easily disturbed.
	Standing	Cat is in an upright position and immobile, with all four paws on the ground and legs extended, supporting the body.
	Watching	Cat observes a specific stimulus, without getting easily distracted or with the intention to interact with the stimulus.

	Yawn	Cat opens its mouth widely while inhaling, then closes mouth while exhaling deeply.
Stereotypic	Fur-plucking	Cat excessively grooms a specific area of its body. This can include any tail- and paw-sucking actions. May result in the removal and visible loss of fur, as well as skin irritation.
Vocalization	Vocalize	Cat produces sounds or calls, originating from the throat and mouth.
Other	Out-of-sight	The cat is not in the visible area.

Table 2: Modifiers

Ground and surroundings
Food or edible sources
Bowl with/for enrichment
Balls made out of vines or hay, of different dimensions
Fence or artificial structures of the enclosure
Coconut
Logs, branches, wooden objects
Wild animals (opossum)
Other types of enrichment non defined

7. Data collection

The recordings were conducted for twenty-eight days from the 23rd June 2022 to the 22nd July 2022, using two camera traps that would activate when sensing movement or heat sources, and a camera to capture videos during the sessions, after the opening of the double door for the enrichment session.

The first camera trap was placed at the management cage, although I changed its position through the study in order to have a better angle. The other camera trap changed position every seven days, including the fasting day, during which the subject was not visited by anybody as it corresponded to the day off of the volunteer. The total time of the observations is 10,111s (168.516min), subdivided in the different shooting areas as seen in *Table S1*.The enrichment during the session was varied, but the key point was that it should not involve food, with some exceptions, and it should have been made from natural materials. It spanned from enrichment (essential oils, deodorants, cinnamon powder, catnip, blood, urine, hay from other animals, objects from the jungle), exercise (walking or running around the enclosure, piñata with logs or pieces of food, obstacles in the path), environmental enrichment (changing the position, addition or removal of the objects from the play area, hiding or making things harder to reach), and ultimately food enrichment, which occurred mainly at the end of the sessions.

7.1. Selection

Before analysing the videos, a selection was made. This was performed by implementing the following criteria:

- Presence of the study subject in the footage;
- Length, as videos longer than 1 minute were preferred over shorter ones;
- Variety of behaviors performed, as the study aimed to represent as many behaviors as possible.

The main differences in number of videos used and length of the videos is due to two main factors: placement and human ability. At the beginning, assessing the best way to place the camera traps was not easy, considering that some areas were free from visual obstacles, and therefore it was easier to collect videos in which the subject appeared for longer time. Additionally, the staff set a limitation, as it was mandatory to place them outside the enclosure, to avoid the study subject to break them and harm himself.

7.2. Reliability trial

The intra-observer reliability test was done using a total of thirty-eight videos, half of which were analysed with a continuous methodology, and the other half using the instantaneous methodology. The results showed a high degree of agreement between the first and the second reading, with a mean value of Kc=0.976 in the continuous sampling, and of Ki=0.981 for the instantaneous sampling.

The Spearman's rank correlation (P-value<0.001; two-tailed, *see Table S2*) was calculated for all behaviors. In the continuous sampling, the average was 0.997 (P-value<0.001; two-tailed). In particular, the values in all behaviors were higher than 0.999, with the exception of *body rub*, *explore*, *flehman*, *paw*, *walking* and *watching*, in which they were higher than 0.984 (P-value<0.001; two-tailed). In the correlation test for the instantaneous methodology, for all behaviors the value was 1.000 (P-value<0.001; two-tailed), with the exception of *standing* and *vocalize*, resulting still higher than 0.998 (P-value<0.001; two-tailed).

The outlier results can be attributed to the nature of the input data, as we took into consideration timeframes with milliseconds, and, for example, a behavior that was performed for 7.907s instead of 8.306s could have caused the statistics to detect it as a mismatch.

7.3. Video analysis

Out of the one-hundred-two videos selected, sixty-four were used for the final analysis. It consisted of the reading of the recordings with the two different methodologies. In the continuous sampling, the beginning and end of each behavior exhibited by the focal animal were recorded. Some overlaps occurred, as walking and exploring might have happened at the same time. For the instantaneous sampling, on the other hand, the behavioral data were recorded with a 5-second interval. This was a premade choice taken on the basis of the collected recordings, as they were not very long in length, ranging between 15 seconds and 7 minutes.

7.4. Data collection

BORIS (https://www.boris.unito.it/) is a free piece of software developed by Oliver Friard and Marco Gamba. It is the main tool used for the collection of the data in this study. It allows the observer to pause, slow down, and speed up the uploaded videos, and it permits to associate a key of the keyboard to a behavior, for a faster analysis. Lastly, it allows statistical tests to be performed and the calculation of the time budget for the selected observations. Therefore, the useful data were extrapolated from the software and analyzed via excel spreadsheets.

7.5. Data treatment and statistical analysis

The software output were tables subdivided per observation, in which every behavior (with the modifiers, if needed) was described by different variables (total number of occurrences, total duration, duration mean, duration sd dev, inter-event intervals mean, inter-event intervals std dev, % of total length). In order to compare the results obtained, we considered the percentage of execution for each behavior. O*ut of sight* was not analyzed, as it is not a behavior *per se*. For the data taken during the continuous sampling, the focus was on the total duration of the behavior performed during the observation. Therefore, the percentage was calculated for every video for every behavior, with the formula:

$$\%CR = \frac{duration\ of\ the\ behavior\ x\ (s)}{total\ lenght\ of\ the\ observation\ (s)}\%.$$

For the instantaneous recording, due to the nature of the sampling, the focal data was the number of sample points in which the behavior was recorded, and to compute the percentage the formula used was:

$$\%IR = \frac{number\ of\ sample\ points\ of\ the\ behavior\ x}{total\ number\ of\ sample\ points\ in\ the\ observation}\ \%.$$

This allowed us to run a Spearman's rank analysis to compare the results for each behavior in every observation.

Results and discussions

1. Descriptive analysis

1.1. Comparing the percentage of time for each category with the two methodologies

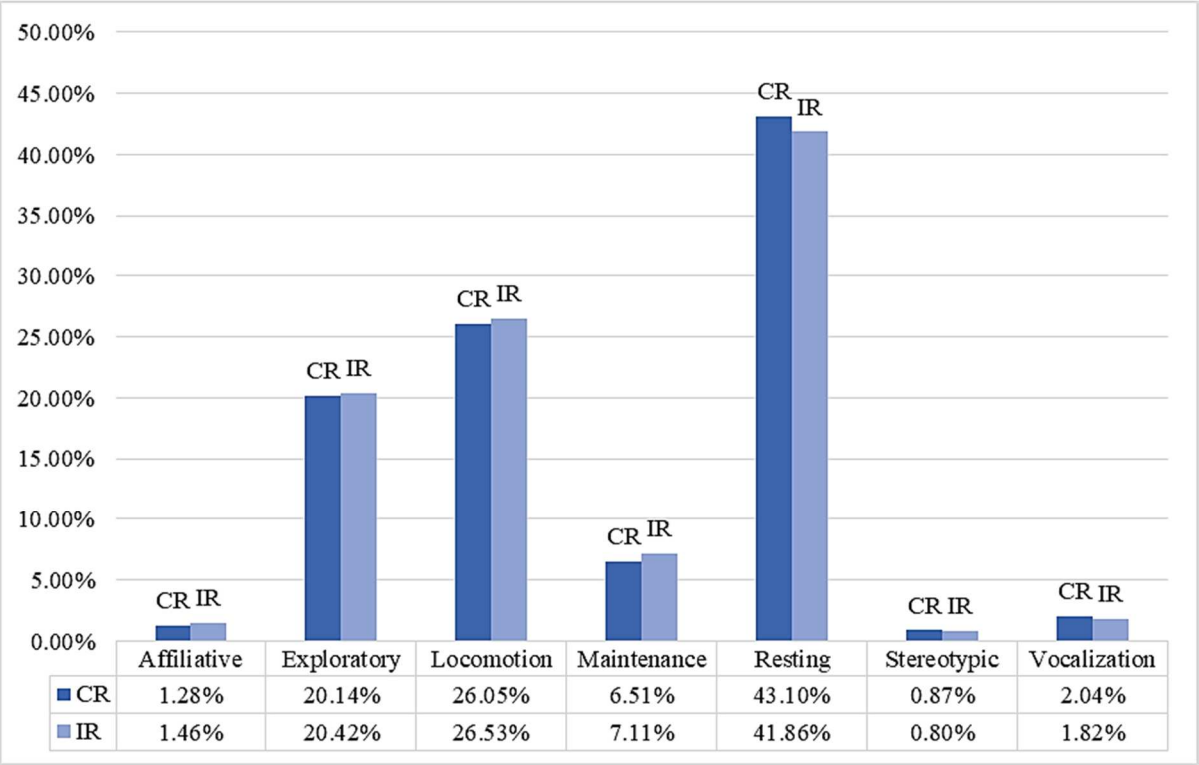
The activities mostly performed by the study subject fall under the categories of *resting* (43.1% in CR and 41.86% in IR), *locomotion* (26.05% in CR and 26.53% in IR) and *exploratory* (20.14% in CR and 20.42% in IR; see *Table S3*).

For the first category, the behavior *standing* was the most frequent, followed by *lying*. Even in the time budget described by *Munita et al. (2016)* for cheetahs, resting was the most frequent category, but contrastingly, *lying* was performed more than *standing*.

The second category is *locomotion*, and specifically the behavior *walking* was most frequently recorded by both methods. This can be influenced by the fact that my recordings were not done casually, but at

times during which the study subject was used to walking and doing physical activity encouraged by the volunteer. Moreover, while walking, the jaguar would often stop to explore and observe the surroundings, and therefore this might explain the high frequency of the behavior *standing*.

The third category *exploratory* is characterized by a high percentage of the behaviors *explore* and *play*. We can assume both of those are highly related to stimuli that volunteers placed in the cage of the animal (e.g. odor enrichment, toys, food) and to the natural location, which allows wild animals to be heard and/or interact with the subject.



	Affiliative	Exploratory	Locomotion	Maintenance	Resting	Stereotypic	Vocalization
CR	1.28%	20.14%	26.05%	6.51%	43.10%	0.87%	2.04%
IR	1.46%	20.42%	26.53%	7.11%	41.86%	0.80%	1.82%

Graph 1: Time allocation of the 7-years-old jaguar, for the two behavioral recording methods applied (continuous recording [CR] and interval recording [IR]).

1.2. Bias due to the presence of people

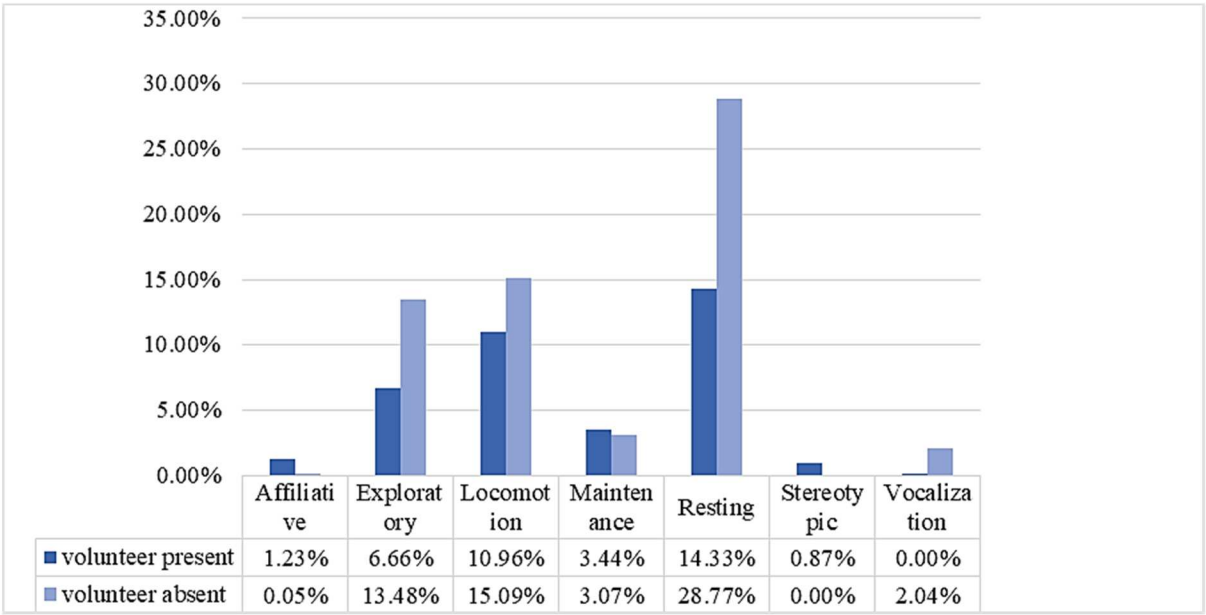
As mentioned before, a video camera and two camera traps were used to collect the observations (videos) for the study. The moments recorded by the video camera were not random, as they were taken right after the first enrichment sessions. Moreover, for the period of the study, the jaguar was not visited just by me, but also by another volunteer and veterinarians. This meant that the behaviors recorded with the camera and during visits were more varried and influenced by the presence of people.

The category *affiliative* results to be almost completely absent when the study subject was alone. The only behavior of the category, *body rub*, was mainly performed on objects with odor enrichment, and against the fence, usually near the volunteer. The people who worked with the study subject have always

described him as “friendly”. The graph clearly shows this temperament, probably resulting from the captive upbringing together with the positive experiences he had with volunteers.

People relatively influenced the time allocation: with the volunteer present, the study subject spent 17.76% of its time *exploring* (see *Table S4*), while without the volunteer, the jaguar spent 21.57% performing the same behavior. Under this category, there are different broad behaviors. As an example, the behavior *explore* could have different aims, some of which may be influenced by the presence of people (enrichment), while others may not (foraging). The presence of food in the enclosure, after the volunteer left, could explain the result, as also the behavior *eating* increased (from 0.00% to 0.78%, see *Table S4*). This kind of result tells us how, with different stimuli, the jaguar performed the same amount of behaviors in the category *exploring*.

Moreover, we have to take into account how the equipment used works: the camera traps activate through the movements and variations in heat, therefore it is obvious that behaviors associated with movement are highly represented by the observations of the camera traps. The same line of reasoning can be applied to the increase in the category *locomotion*, as the presence of the volunteer caused an absolute increase of 4.13%.



	Affiliative	Exploratory	Locomotion	Maintenance	Resting	Stereotypic	Vocalization
volunteer present	1.23%	6.66%	10.96%	3.44%	14.33%	0.87%	0.00%
volunteer absent	0.05%	13.48%	15.09%	3.07%	28.77%	0.00%	2.04%

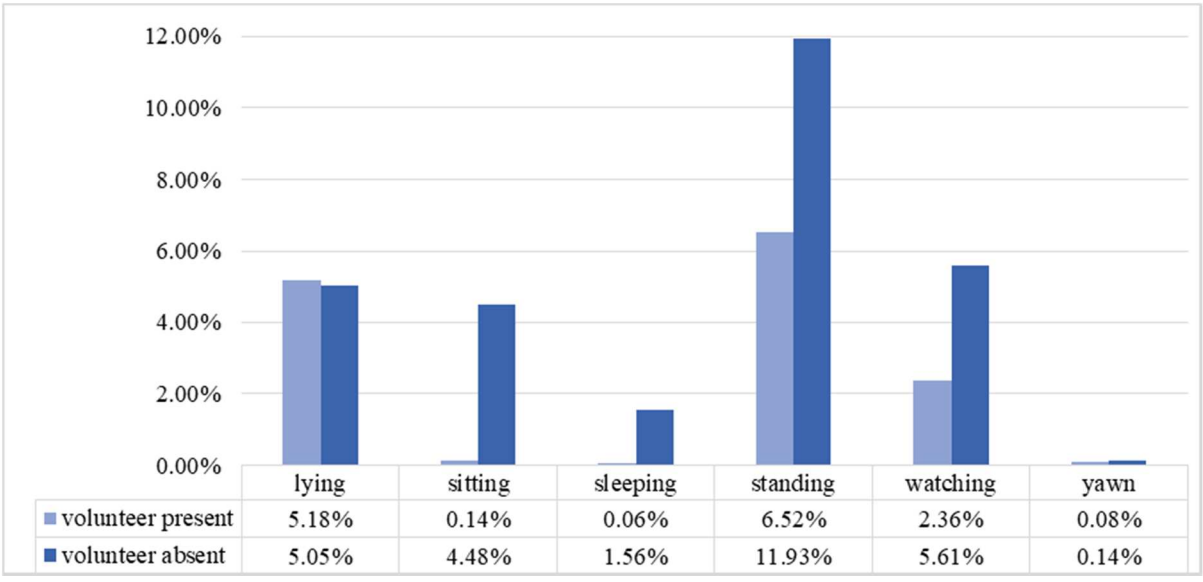
Graph 2: Absolute distribution of the behaviors during the sessions and alone time (see Table S4 for the data).

On the other hand, the increase for the category *resting* has a different nature (*see Graph 3*). It was extremely rare that the study subject was performing inactive behaviors such as *sleeping* (0.06%) during the sessions with people. Instead, more active behaviors (*standing, watching*) were performed, and it is interesting to note that they all allow the jaguar to gain information from its environment. That is more important when alone rather than in company, taking into account also the fact that other animals the

subject might have been interested to watch, hear or smell could have been scared away by the presence of humans.

In addition, the results from the category *vocalization* match with this hypothesis, as vocalizations were performed without the presence of people, mostly during the night. A wild male jaguar was captured on video by some camera traps, interacting with the caged study subject. These visits happened at night or dawn, long time after the last person visited the cage. The data do not prove that the vocalization episodes were directed to the wild jaguar, but if we assume that as the reason, it also explains why they were performed during natural times of activity for the species.

Lastly, the category *stereotypic* was only recorded while the subject was not alone. It is a self-traumatic behavior associated with boredom and stress, lack of enrichment and adequate spaces and areas to hide *(Hope et al. 2006)*. This behavior increased significantly once traces and feces from the wild jaguar started to appear around the enclosure. The assumption made with the cat coordinators and veterinarians was that, with the higher activity of the study subject during the night, he was prone to boredom and even stress during the day with the volunteers around. Even if there is no data to prove it, the behavior decreased significantly on the days when no new traces left by the wild jaguar were found anymore. We also have to consider that *tail-sucking* is a behavior was more easily recorded by the camera, or by the camera traps due to the movement of the volunteer activating them.



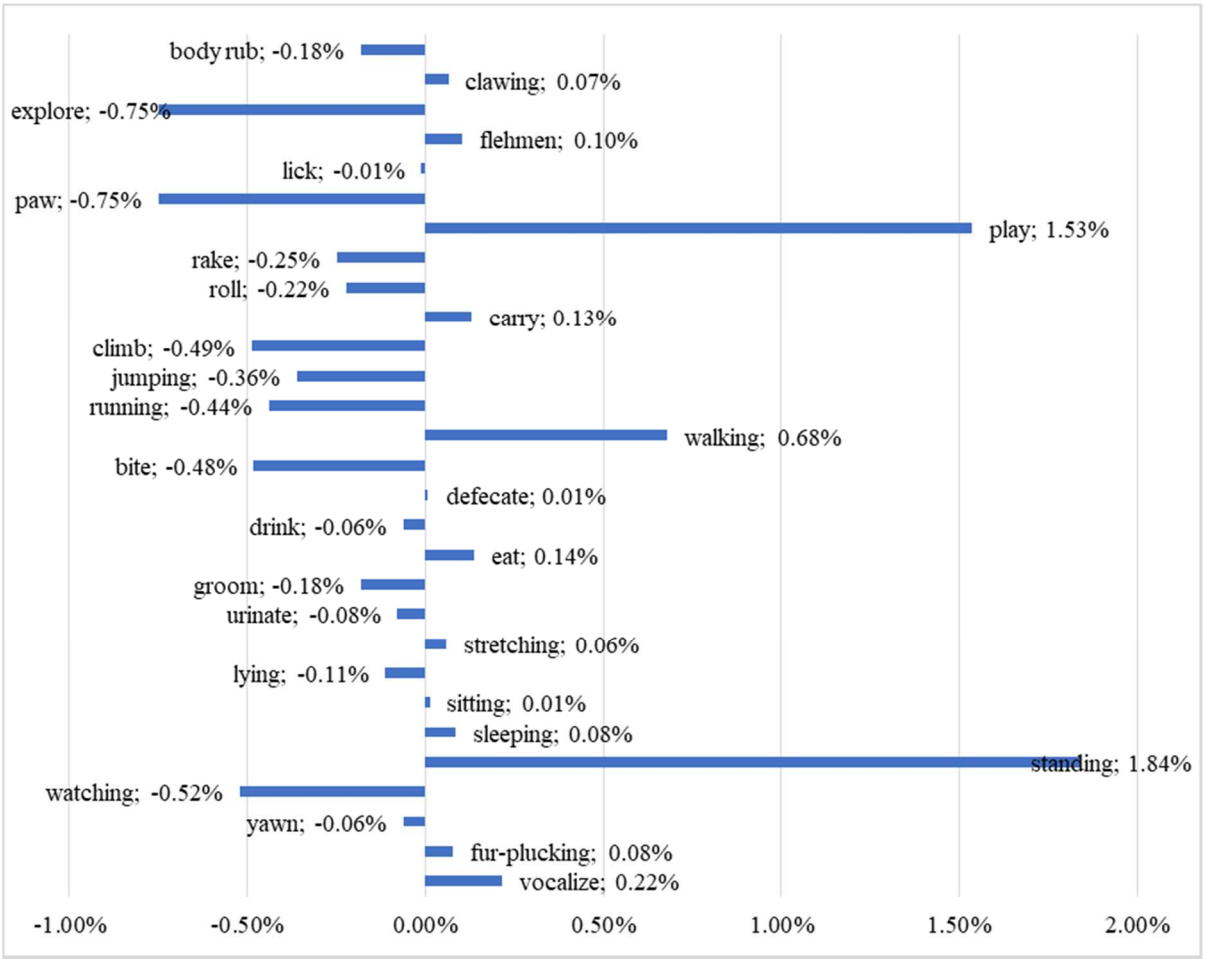
	lying	sitting	sleeping	standing	watching	yawn
volunteer present	5.18%	0.14%	0.06%	6.52%	2.36%	0.08%
volunteer absent	5.05%	4.48%	1.56%	11.93%	5.61%	0.14%

Graph 3: Distribution of the behaviors under the category "resting" during the sessions and alone time.

1.3. Over and under estimation of time by IR

The instantaneous recording method, with the sample interval of 5 seconds, allowed the recording of all behaviors. Assuming that the continuous sampling represents the reality of how the subject spent its time during the time he was video-recorded, we have run this analysis in order to understand which behaviors were under or overestimated by the instantaneous method.

The cases in which the difference is more evident (estimation higher than 0.5% or lower than -0.5%) are with the behaviors *explore*, *paw*, *play*, *walking*, *standing* and *watching*. The short duration of some behaviors might cause a wrong estimation of the percentage of time spent for each behavior, assigning a higher percentage of time (overestimating) for those ones that happened at the recording instant, and a lower percentage for those which happened between one and the other recording (underestimating). The higher values (*play, walking, and standing*) are for behaviors which have been underestimated. Regarding the results obtained in this study, the differences are still low; however, depending on the aims of the study in which instantaneous recording will be used, these not 100% accurate estimations may influence the results.



Graph 4: Over and underestimation of time by IR, considering CR as the real amount of time spent for every behavior. The negative values indicate an overestimation (%IR>%CR), while the positive values indicate an underestimation (%IR<%CR). The reference datasheet is at Table S5.

2. Inferential analysis

2.1. Spearman's for the categories

In order to assess the correlation between the two behavioral methods applied in the study, the Spearman's rank was calculated for all the categories *(see Table S6)*. The mean is 0.941, and the lowest value results to be for the category *affiliative*, still with a high correlation (0.867, P-value<0.001).

Stereotypic is the only category in which the Spearman's rank correlation coefficient was 1.000 (P-value<0.001; two-tailed), and this might have been caused by the restricted number of recordings for that behavior, as well as the long duration of it, up to some minutes. Still, all the other categories had a corresponding correlation coefficient higher than 0.900 (P-value<0.001; two-tailed), which was expected with such a sample interval.

This tells us that the use of instantaneous sampling (with 5 seconds intervals), even for most of the short duration behaviors, was able to replicate the continuous sampling data with a fairly high degree of accuracy, as demonstrated by the correlation coefficients.

2.2. Spearman's for the behaviors

The results obtained with the statistical analysis for all the single behaviors were extremely positive (see *Table 3)*. For readability, we classified them in ranges. As shown in *Table 3*, they distributed fairly well, as seven behaviors had a Spearman lower than 0.900, nine were in the range 0.900-0.950, six in the range 0.950-1.000, and the remaining six had a value of 1.000.

The behaviors in the lowest range share the characteristic that they are all short in duration, therefore the low results were probably due to this factor. *Jumping* was the lowest, with the value of 0.773 (P-value<0.001), entailing the instantaneous sampling a bad choice to study this behavior.

As we can see behaviors with longer duration, such as *flehmen*, *watch*, *walking*, *lying*, *eating* show a higher Spearman's rank correlation between CR and IR data.

The behavior *stretching* was not observed with the instantaneous sampling, therefore did not produce any statistical data. Excluding this behavior, the mean between all the results was 0.932 (P-value<0.001), which reflects a very strong correlation between the two methods.

The outliers behaviors in the over/under estimation analysis (see *Table S5*) still resulted to have a high correlation (*explore* 0.885, *paw* 0.933, *play* 0.970, *walking* 0.972, *standing* 0.897, *watching* 0.906; P-value<0.001, two-tailed), implying as a consequence that the differences detected in that analysis are irrelevant to the method used.

Range (P-value<0.001; two-tailed)	**Behaviors**
<0.900	Jumping (0.773), yawn (0.848), body rub (0.867), sleeping (0.873), running (0.884), explore (0.885), standing (0.897)
0.900<x<0.950	Rake (0.901), flehmen (0.902), watching (0.906), roll (0.907), groom (0.907), drink (0.920), vocalize (0.920), paw (0.933), bite (0.933)
0.950<x<1.000	Clawing (0.952), climb (0.956), play (0.970), walking (0.972), lying (0.998), carry (0.999)
1.000	Lick, defecate, eating, urinate, sitting, fur-plucking
n/d	Stretching

Table 3: The results obtained using Spearman correlation test for each behavior (all P-value <0.001; two-tailed), divided into ranges. N/d stands for "non-defined".

Conclusion

The aim of the study was to investigate the correlation between the two methods used, and the results were satisfactory and relatively high. Nevertheless, the sample interval was quite low (5-seconds) due to the length of the videos taken by the camera traps, and that the results were to be expected. Still, the behaviors *jumping* and *yawn* showed to have a correlation value that was lower than 0.850. For future studies, it is important to consider these results when deciding which sampling method to use, as the instantaneous recording was not satisfactory for the above-mentioned behaviors, even with a 5-seconds interval.

An interesting further study in this field would be to quantify the effect that increasing the time interval (in the instantaneous sampling) has on results compared to the 5-second interval. To find a method that produces data very correlated to the continuous sampling results with large time intervals would bring many benefits to the research.

The collection method represents the main limitation to this study, as a longer video length from the camera traps, as well as better positioning, would have gave us a better insight on the matter. Moreover, a single study subject is not enough to create a universal time budget for captive jaguars, but it sets the first milestone for the creation of one.

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Appendix A – Extra Tables

Table S1: Subdivision of the recording areas and total time samples for each of them.

Area	**Number of videos selected for the area**	**Total time in seconds of the area**
0. Camera	27	6126 s
1. Management cage	44	2285 s
2. Play area	16	800 s
3. Corner 3	4	240 s
4. Corner 4	4	240 s
5. Corner 5	7	420 s
Total	102	10111 s (168.5167 min)

Table S2: Results of Spearman's correlations ($p<0.001$; two-tailed) between the two behavioral recording methods applied (continuous recording [CR] and interval recording [IR]), for the reliability trial.

Behavior	**CR Spearman**	**Behavior**	**IR Spearman**
body rub	0.997	body rub	1.000
bite	1.000	bite	1.000
eating	1.000	carry	1.000
explore	0.994	climb	1.000
flehmen	0.997	eating	1.000
groom	1.000	explore	1.000
lick	1.000	flehmen	1.000
lying	1.000	fur-plucking	1.000
o/o/s	1.000	groom	1.000
paw	0.995	jumping	1.000
play	1.000	lying	1.000
rake	1.000	o/o/s	1.000
roll	1.000	paw	1.000
sitting	1.000	play	1.000
sleeping	1.000	rake	1.000
standing	1.000	sitting	1.000
stretching	1.000	standing	0.999

urinate	1.000	stretching	1.000
vocalize	1.000	vocalize	0.998
walking	0.984	walking	1.000
watching	0.988	watching	1.000
		yawn	1.000

Table S3: Time allocation percentages for each of the two behavioral recording methods applied (continuous recording [CR] and interval recording [IR]), over a 30-day period of observations.

Category	**Behavior**	**% CR**	**%CR by category**	**%IR**	**%IR by category**
Affiliative	body rub	1.28%	1.28%	1.46%	1.46%
Exploratory	clawing	1.11%	20.14%	1.04%	20.42%
	explore	6.28%		7.03%	
	flehmen	0.88%		0.78%	
	lick	0.63%		0.64%	
	paw	3.85%		4.60%	
	play	6.71%		5.18%	
	rake	0.49%		0.74%	
	roll	0.18%		0.41%	
Locomotion	carry	0.70%	26.05%	0.57%	26.53%
	climb	0.72%		1.21%	
	jumping	0.39%		0.75%	
	running	1.65%		2.09%	
	walking	22.60%		21.92%	
Maintenance	bite	1.20%	6.51%	1.68%	7.11%
	defecate	0.34%		0.33%	
	drink	1.67%		1.73%	
	eating	0.78%		0.64%	
	groom	2.07%		2.25%	
	urinate	0.41%		0.48%	
	stretching	0.06%		0.00%	
Resting	lying	10.23%	43.10%	10.34%	41.86%
	sitting	4.62%		4.61%	
	sleeping	1.62%		1.54%	
	standing	18.45%		16.61%	
	watch	7.96%		8.48%	
	yawn	0.22%		0.28%	

Stereotypic	fur-plucking	0.87%	0.87%	0.80%	0.80%
Vocalization	vocalize	2.04%	2.04%	1.82%	1.82%

Table S4: Distribution of the behaviors during the sessions with the volunteers and the time in which the jaguar was alone. The first four columns are the absolute values of the percentages; the four last columns are the values relative to the percentage of time spent with and without the volunteer.

	Behavior	**Absolute %**				**Relative % (value/sum)**			
	volunteer present	yes	no	yes	no	yes	no	yes	no
Affiliative	body rub	1.23%	0.05%	1.23%	0.05%	3.28%	0.08%	3.28%	0.08%
Exploratory	clawing	0.36%	0.75%	6.66%	13.48%	0.97%	1.19%	17.76%	21.57%
	explore	2.10%	4.18%			5.60%	6.69%		
	flehmen	0.13%	0.76%			0.34%	1.21%		
	lick	0.63%	0.00%			1.67%	0.00%		
	paw	1.46%	2.39%			3.89%	3.83%		
	play	1.76%	4.95%			4.69%	7.92%		
	rake	0.17%	0.32%			0.46%	0.51%		
	roll	0.05%	0.13%			0.14%	0.21%		
Locomotion	carry	0.04%	0.66%	10.96%	15.09%	0.11%	1.05%	29.23%	24.15%
	climb	0.05%	0.67%			0.12%	1.08%		
	jumping	0.11%	0.28%			0.28%	0.45%		
	running	0.33%	1.32%			0.89%	2.11%		
	walking	10.43%	12.17%			27.82%	19.47%		
Maintenance	bite	0.45%	0.74%	3.44%	3.07%	1.20%	1.19%	9.17%	4.92%
	defecate	0.34%	0.00%			0.90%	0.00%		
	drink	1.67%	0.00%			4.45%	0.00%		
	eating	0.00%	0.78%			0.00%	1.25%		
	groom	0.52%	1.55%			1.38%	2.48%		
	urinate	0.41%	0.00%			1.08%	0.00%		
	stretching	0.06%	0.00%			0.16%	0.00%		
Resting	lying	5.18%	5.05%	14.33%	28.77%	13.81%	8.07%	38.22%	46.03%
	sitting	0.14%	4.48%			0.38%	7.17%		
	sleeping	0.06%	1.56%			0.15%	2.50%		
	standing	6.52%	11.93%			17.37%	19.09%		
	watching	2.36%	5.61%			6.28%	8.97%		
	yawn	0.08%	0.14%			0.22%	0.22%		
Stereotypic	fur-plucking	0.87%	0.00%	0.87%	0.00%	2.33%	0.00%	2.33%	0.00%
Vocalization	vocalize	0.00%	2.04%	0.00%	2.04%	0.01%	3.26%	0.01%	3.26%
Sum				37.50%	62.50%				

Table S5: Over and under estimation of the IR of the percentage of time spent, in comparison with the CR.

Behavior	**ΔIR**	**Behavior**	**ΔIR**
body rub	-0.18%	defecate	0.01%
clawing	-0.22%	drink	-0.06%

explore	-0.75%	eating	0.14%
flehmen	0.10%	groom	-0.18%
lick	-0.01%	urinate	-0.08%
paw	-0.75%	stretching	0.06%
play	1.53%	lying	-0.11%
rake	-0.25%	sitting	0.01%
roll	-0.22%	sleeping	0.08%
carry	0.13%	standing	1.84%
climb	-0.49%	watching	-0.52%
jumping	-0.36%	yawn	-0.06%
running	-0.44%	fur-plucking	0.08%
walking	0.68%	vocalize	0.22%
bite	-0.48%		

Table S6: The results obtained by running the statistical analysis for each category (P-value <0.001; two-tailed).

Category	**Spearman**	**P-value; two-tailed**
Affiliative	0.867	<0.001
Exploratory	0.939	<0.001
Locomotion	0.974	<0.001
Maintenance	0.971	<0.001
Resting	0.913	<0.001
Stereotypic	1.000	<0.001
Vocalization	0.920	<0.001

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