RESEARCH ARTICLE

Behavioral Indicators of Ovarian Phase in White-Faced Capuchins (*Cebus capucinus*)

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In many primate species, conspicuous behavioral and/or morphological changes are indicators of the fertile phase of the female cycle. However, several primate species, such as the white-faced capuchin, lack these cues. This is referred to as "concealed ovulation," and is argued to be a reproductive strategy that confuses paternity and lowers the risk of infanticide. We studied 10 adult female white-faced capuchins in Santa Rosa National Park, Costa Rica, from January to June 2002. We determined their ovarian cycling patterns by analyzing fecal ovarian hormones, and compared simultaneously collected behavioral data to determine which, if any, cues females use to signal their fertile phases. We found that four females cycled during the study period but ceased to cycle without becoming pregnant. We considered several explanations for the lack of conception during our study, including reproductive seasonality. We found that female *C. capucinus* showed only small increases in rates of affiliative/proceptive behaviors directed toward adult males during their periovulatory phases. The best indicator of cycle phase was a significant increase in male affiliative behaviors (e.g., following and grooming bouts) and sexual behaviors (e.g., copulations and courtship displays) directed toward females during the periovulatory phase compared to the nonovulatory phase. Our finding that females exhibit little proceptive behavior, but that copulations and male courtship are nonetheless concentrated in periovulatory phases suggests that even though females do not provide behavioral and morphological cues to ovulation, males are still able to detect it. Infanticide occurs with some frequency in these monkeys, and there is evidence for postconceptive mating as a female strategy to lower risk of infanticide via paternity

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confusion. However, despite this occurrence of nonconceptive mating and the absence of female cues to ovulation, truly concealed ovulation does not appear to be characteristic of this study population of white-faced capuchins. Am. J. Primatol. 67:51–68, 2005.   © 2005 Wiley-Liss, Inc.

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INTRODUCTION

In many primate species, conspicuous behavioral and morphological changes are indications of an alteration in the female's reproductive state (e.g., *Macaca fuscata* [Enomoto et al., 1979], *Cercocebus torquatus atys* [Gordon et al., 1991], and *Cebus apella* [Carosi & Visalberghi, 2002; Linn et al., 1995; reviewed by Hrdy & Whitten, 1987]). However, the females of several primate species lack physical cues as to their reproductive state, and are also cryptic in displaying behavioral indicators of that state (e.g., *Cercopithecus aethiops* [Andelman, 1987], *Presbytis entellus* [Hrdy, 1977], and *Saguinus oedipus* [Ziegler et al., 1993a]). This lack of female cues has been referred to as ''concealed ovulation'' because the optimum conceptive stage of the ovarian cycle (i.e., ovulation) is not made obvious to the male [Dixson, 1998; Nunn, 1999; Sillén-Tulberg & Møller, 1993]. Concealed ovulation has been argued to be a reproductive strategy that has evolved in some multimale species to promote paternal care [Alexander & Noonan, 1979] or to lower male–male competition and hence reduce the likelihood of infanticide [Hrdy, 1979]. Female white-faced capuchins (*Cebus capucinus*) do not exhibit any external morphological signs of ovulation, and are not known to display obvious behavioral cues to indicate they are within the periovulatory phase of their cycles [Manson et al., 1997].

For some species (e.g., *Cebus apella* [Linn et al., 1995; Carosi et al., 1999], *Cercocebus torquatus atys* [Gordon et al., 1991], *Saguinus oedipus* [Ziegler et al., 1993b], *Pan paniscus* [Heistermann et al., 1996]), laboratory studies have provided essential information about changes in behavior across the ovarian cycle by providing controlled plasma, urinary, and fecal hormonal data correlated with simultaneously collected behavioral data. However, in the past it was difficult for field researchers to infer ovulation phases and estimate gestation lengths in primate species that exhibit inconspicuous ovulation and nonseasonal mating patterns (e.g., *Cercopithecus aethiops* [Andelman, 1987]). Fortunately, improved techniques now allow field researchers to analyze ovarian hormones in feces collected from wild primates, and to look for a relationship between hormonal patterns and behavior in species that offer no morphological cues to ovulation. In this study we examined ovarian hormone data from wild *Cebus capucinus,* collected concurrently with behavioral data, to investigate whether there are behavioral cues that may be reliable indicators of the periovulatory phase of female cycles.

Cebus apella is the most studied species in the genus *Cebus*, and the females of this species exhibit strikingly obvious proceptive patterns. For instance, *C. apella* females actively solicit copulations with the alpha male via such behaviors as ''eyebrow raising with vocalizations,'' ''touching and running,'' ''nuzzling,'' and ''head cocking'' [Carosi et al., 1999; Carosi & Visalberghi, 2002; Linn et al., 1995; Philips et al., 1994]. These persistent proceptive behaviors are thought to be the driving force behind mating in *Cebus apella*, since females

exhibit them during the peak of the periovulatory period (within the first few days of proceptivity) and direct them only toward the alpha male. Female *C. apella* also mate with subordinate males, but only near the end of their proceptive phase, when the presumed optimum conceptive period has passed [Janson, 1984].

In contrast, male white-faced capuchins are almost exclusively responsible for initiating the courtship dance toward females, who may or may not respond with a similar display of behavior (see Perry [1997] and Manson et al. [1997] for descriptions of courtship displays). Furthermore, females of *C. capucinus* mate with all adult males in the group (although it has not yet been determined whether they restrict their copulations to the alpha male during their most fertile days [Fedigan, 1993; Manson et al., 1997]). There may be subtle behaviors that females use to signal their sexual readiness to males, which are not directly and noticeably part of copulatory patterns. Following Dixson [1998], we refer to actions that serve to bring the sexes into proximity and to maintain them in close physical contact as ''pericopulatory behaviors.'' We divide these pericopulatory behaviors into proceptive and attractive indicators, such that behaviors directed by the cycling female toward a male are considered proceptive, and behaviors directed toward the cycling female from an adult male are considered attractive.

In *Cebus apella*, grooming tends to increase between males and females during the periovulatory period [Di Bitetti, 1997; Linn et al., 1995]. White-faced capuchins are very social during rest periods, and grooming is a common social activity; therefore, grooming may be one behavioral variable that is used to indicate either proceptivity or attractivity, or both. Other behaviors that we investigated as possible pericopulatory behaviors included follows, approaches, and maintenance of spatial proximity (e.g., during feeding or resting). We predicted that females would increase the rate of proceptive behaviors (e.g., *directed* grooming, approaches, and follows) toward males during the periovulatory phase compared to the nonovulatory phase in an effort to advertise their conceptive state. Additionally, we predicted that during the periovulatory phase, adult males would increase the frequency of behaviors that indicate attraction to the females (e.g., grooming, approaching, and following females). We also predicted that males and females would maintain proximity to each other more during the periovulatory phase compared to the nonovulatory phase in order to facilitate copulation.

Manson et al. [1997] suggested that mating in capuchins serves functions in addition to procreation, since white-faced capuchins perform courtship displays and copulate throughout the year at times when the female is in various reproductive states (i.e., pregnant, lactating, or cycling). Because white-faced capuchins mate year-round and exhibit no morphological signs of ovulation, it is difficult to determine whether courtship and copulations actually indicate that the female is conceptive (i.e., in the periovulatory phase). However, Carosi and Visalberghi [2002] found that in *Cebus apella,* male sexual mounting of females occurs at a significantly higher rate during the periovulatory phase compared to the nonovulatory phase (also see Linn et al. [1995]). Additionally, sexual behavior in many species of Old and New World primates increases during their conceptive phases. Therefore, we predicted that during their periovulatory periods, the rate of copulations and courtship displays would increase in our study subjects.

Urine washing may also qualify as a proceptive behavior and vary with respect to cycle phase. This behavior is common in all species of capuchins, and consists of rubbing urine onto the hands and feet. Some researchers have proposed that urine washing functions in thermoregulation [e.g., Robinson, 1979; Roeder & Anderson, 1991], whereas others have suggested that it communicates

reproductive status [Boinski, 1992; Perret, 1992] or territoriality (mainly in prosimians [Schilling, 1979]). In white-faced capuchins, all age/sex classes exhibit urine washing, which suggests that its function is common to all individuals. However, male white-faced capuchins are occasionally seen smelling and licking the branch where a female has performed a urine wash (unpublished data). There may be multiple functions to this behavior, including communication of reproductive status; therefore, we predicted that the rate of urine washing would be higher during the periovulatory phases of female cycles.

In this study we compared the occurrence of sexual and pericopulatory behaviors between the periovulatory and nonovulatory phases of female white-faced capuchin ovarian cycles. Our purpose was to determine whether female white-faced capuchins exhibit behavioral cues that signal their fertile phase to adult males. This study is the first to relate behavioral data to hormonal cycles in free-ranging *Cebus capucinus*.

MATERIALS AND METHODS

Study Site and Subjects

This study took place in Santa Rosa National Park (SNRP), the original sector of the Area de Conservacion Guanacaste (ACG). SNRP is located in northwestern Costa Rica, close to the Nicaraguan border, and encompasses approximately 108 ha of dry deciduous forest fragments, semi-evergreen and riparian forests, and regenerating pasture land. The park is home to three primate species (*Cebus capucinus, Ateles geoffroyi,* and *Alouatta palliata*) that have been the subject of many behavioral ecology studies [Fedigan & Jack, 2001].

The subjects for this study were 10 white-faced capuchins that constituted all the adult female members of two habituated groups: CP and LV (five per group). They ranged in age from 6 to ~24 years, and varied in rank within each group. Eight females were multiparous, and two were nulliparous at the start of the study. We inferred that our study females began cycling by 5.5 years of age since 6 years is the earliest recorded age at first birth in this population [Fedigan & Rose, 1995]. During this study there were six adult males in the two groups (one alpha male and one subordinate male in CP, and one alpha male and three subordinate males in LV). Individuals were identified by natural markings such as broken digits, scars, fur coloring, blotches on the face, and the brow and peak shape. Both groups live in similar types of habitat and are exposed to similar environmental pressures, including water scarcity, predator pressures, food availability, and level of human exposure, and both groups of capuchins have been part of L.M.F.'s research since 1983. Capuchins are female bonded [Fragaszy et al., 2004], and the kinship relations among the females have been known for the last 15–20 years.

Behavioral Data Collection

Between January and June 2002 we collected 443 hr of focal animal data (ranging from 39 to 47 hr per female) between 6 A.M. and 6 P.M. Focal sessions lasted for 15 min each, during which time all behaviors and interactions were recorded continuously [Altmann, 1974]. We attempted to sample different individuals sequentially throughout the day so that each female would be equally represented. However, if we could not find the target female within 15 min, we would move on to the next on the list. We allowed a 90-sec "out-of-sight" period (i.e., the time when the monkey is not in view but its approximate location is

known, and it is unlikely that confusion with another individual will occur). If the subject was out of sight for longer than 90 sec, that session was discarded.

We used hand-held computers (PSION Workabout MX) and entered data into a software program entitled ''Behavior,'' which was designed by Syscan International Inc. (Montreal, Quebec) for researchers at the University of Alberta. This program records the time each behavior code is entered so that durations of behavior can be accurately and easily calculated. We employed an exhaustive ethogram developed over the years by previous *C. capucinus* researchers to identify and code behaviors. When fast-moving events occurred, such as fights or sexual displays, we dictated the behavioral sequences into a microcassette recorder (Sony M-430). We later transcribed those sequences and integrated them into the behavioral data set for that day. To ensure interobserver reliability for the data collected, the first author and her field assistant spent several weeks working together with one group, identifying individuals and practicing data collection before the assistant began to collect data on her own. Once per month, the first author and the field assistant collected data together on one set of females, and compared notes to remain consistent over the course of the study.

Fecal Collection and Field Extraction

We collected fecal samples from each female on each of the same days that we obtained behavioral data. A minimum of two to three samples per week per female is considered adequate to assess ovarian patterns in females [Hodges & Heistermann, 2003], but we collected additional samples in case the females could not be located in subsequent days. We collected feces within 10 min of defecation, placed them in plastic vials, and stored them in a cold pack until the end of the day, taking note of the individual and date [Strier et al., 2003]. In total, we collected over 600 samples from all 10 subject females, but we analyzed only 400 samples because once we discovered the reproductive state of the pregnant and noncycling females, not all of their samples required analysis. For initial field extraction of the steroids, we followed the techniques described in Strier and Ziegler [1997]: 2.5 ml of distilled water and 2.5 ml of ethanol were added to 0.1 g of wet weight fecal matter in a 15-ml centrifuge tube. The tube was vortexed for 10 min and then centrifuged for 10 min at $2,000 \times g$. The resulting fecal pellet was discarded and the ethanol/water/steroid mixture was refrigerated until it was transported to the National Primate Research Center (NPRC) at the University of Wisconsin in Madison for the laboratory analysis.

Laboratory Extraction and Analysis

We carried out hormone analyses at the NPRC in July and August 2002. Earlier (in February 2001), 10 fecal samples had been collected and field-extracted by the methods described above, and sent to the NPRC in Wisconsin for validation of the steroid hormones. The 10 preliminary samples were pooled and used to validate the progesterone (P) and estradiol (E2) assays that were used. We evaluated the accuracy with which the assay measures the steroid in each sample by adding small volumes of the fecal pool to each point of the standard curve. Parallelism was determined through serial dilutions of the fecal pool along the standard curve. Intra- and interassay coefficients of variation (CVs) were determined by running duplicate samples within and between each assay. The preliminary samples from 2001 were used only for the validation tests, and

the samples indicated that this extraction technique is reliable for measuring these ovarian steroids in capuchin feces.

Analysis of the preliminary fecal samples also indicated that high concentrations of steroids were excreted in a conjugated form. Before the assays were performed, solvolysis was used to break the conjugated steroids into an unconjugated form that would ensure an accurate estimation of the hormone levels. The solvolysis procedure we used is described in Ziegler et al. [1996]. Once the samples had undergone solvolysis, we measured progesterone using enzyme immunoassays [Ziegler et al., 1996] and estradiol using radioimmunoassays [Strier & Ziegler, 1994].

Interpretation of Hormonal Data

Nagle et al. [1979] found in *Cebus apella* that ovulation occurs at approximately the same time as the rise in excreted urinary progesterone. Carosi et al. [1999] further found in *C. apella* that there was only a very short (0–1 day) lag time in the excretion rate between urinary and fecal measurements of P and E2. Therefore, because of the minimal lag time in excreted progesterone from feces compared to urine, we assumed that ovulation occurred on the first day of the rise in fecal P in our capuchins. Following Linn et al. [1995] and Carosi et al. [1999], and to account for any error or variability, we conservatively defined the periovulatory phase as being 5 days long (3 days before the rise in P, the day of the rise, and 1 day after the rise).

We used the results of the fecal assays of the 400 analyzed samples to create a hormone profile for each female that allowed us to infer the female's reproductive state during the course of the study. Of the 10 female subjects, only four were cycling. New World monkeys typically have very short follicular phases and luteal phases that present simultaneously elevated levels of estrogens and progestins over a 10–15-day period [Dixson, 1998; Ziegler et al., 1987, 1993b]. It was difficult to distinguish the three phases of the ovarian cycle (follicular, periovulatory, and luteal) for the cycling females from our hormone profiles. Therefore, we categorized the three phases into two and looked for behavioral differences simply between periovulatory and nonovulatory phases. This grouping into two phases is also consistent with Carosi and Visalberghi's [2002] study of ovarian cycles in *C. apella*. All four cycling females ceased to cycle approximately midway through the study, and three did not start cycling until 1–2 months into the study. Consequently, we used only behavioral data collected while they were cycling (periovulatory: 16.75 hr; nonovulatory: 38.5 hr; total=55.25 hr).

Behavioral Analysis

The behavioral variables analyzed for phase variation included copulations and courtship displays, and the hypothesized behavioral indicators of attractivity and proceptivity. Copulations were defined as male-on-female mounts with thrusting. Behaviors that we considered as indicating proceptivity were approaches, follows, and grooms directed by the subject female toward adult males. We measured the frequency of grooming solicitations as well as grooming durations. We considered these same behaviors as indicators of attractive behavior when they were received from adult males. We also calculated a measure of which sex maintained proximity and compared these across phases.

We compared each of the behavioral frequencies across cycle phases by calculating a mean rate (frequency per hour) for each of the subject females for

each phase. We determined the mean grooming duration (directed and received) by tallying the total amount of time females were involved in grooming bouts, and calculating the proportion of that time spent grooming (or being groomed by) adult males.

We used Hinde's index [Hinde & Atkinson, 1970] to decide which member of the male–female dyad was responsible for maintaining proximity. Hinde's index is the proportion of all of the dyad's *approaches* directed by the subject female, minus the proportion of all of the dyad's *leaves* directed by the female. A negative index indicates that the male was responsible for maintaining proximity; a positive index suggests the female was responsible. *Approaches* to within 3 m, and *leaves* beyond 3 m were used in this analysis.

Since our sample sizes were small, parametric tests were not appropriate and the use of nonparametric tests (i.e., Wilcoxon signed-ranks test) would not provide enough statistical power. Therefore, we used a randomization technique [Manly, 1997] that creates a new distribution of the test statistic from the original data. For this analysis, we randomly scored our original data 10,000 times to create the new distribution that we used to compare our observed test statistic. This is a one-tailed test and significance is reached when probability is < 0.05.

RESULTS

Hormone Validations and Profiles

The mean steroid recoveries were found to be within acceptable limits for P (90.7%) and E2 (64.5%). The mean percent accuracy was high for both the P and E2 assays (> 100% accuracy). Percent bound values from the pooled samples paralleled the percent bound values of the standard curves for both hormones. The mean intra- and interassay CV values were within acceptable limits for both hormones (Table I).

Results from the hormone profiles revealed that three of the 10 females failed to cycle for the entire duration of the study (that is, they showed an absence of hormonal fluctuations and were categorized as ''noncycling'' from January to June 2002); three females were pregnant at the start of the study (and subsequently gave birth during the study); and four females displayed ovarian cycling for a period of time and then stopped cycling even though they had not become pregnant.

Of the four cycling females, three did not exhibit cycling until 14, 41, and 43 days, respectively, after the first sample was collected. Observations for the fourth female appeared to start right at the beginning of her luteal phase (indicated by elevated P). All four females stopped cycling midway through the study for

TABLE I. Summary of Results From Fecal Assay Validations

	Progesterone	Estradiol
Recoveries	90.70%	64.50%
Accuracy	107.25$\pm$2.11%	101.28$\pm$2.29%
Parallelism[a]	t=0.15, df=24, p$>$0.05	t=$-$1.74, df=42, p$>$0.05
CV[b]		
Intra	6.89% n=8	4.73% n=8
Inter	8.56% n=8	8.20% n=8

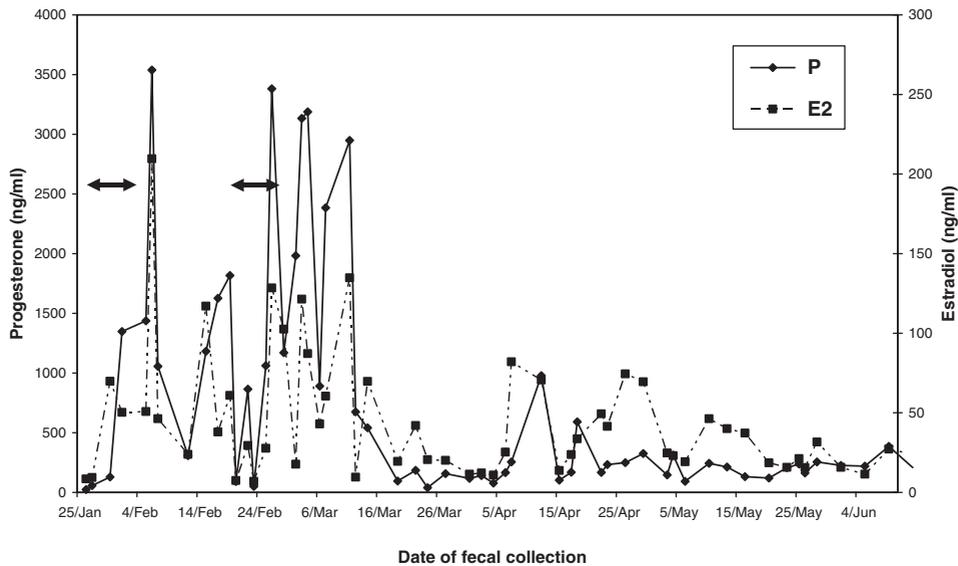[a]Slopes do not differ.
[b]Coefficient of variation.

Fig. 1. Representative hormone profile for a female white-faced capuchin (TT). The two periovulatory phases are indicated by horizontal black arrows. A periovulatory phase covers a span of 5 days (the day of the P surge, 3 days before, and 1 day after). There was only one complete ovarian cycle, calculated from the first progesterone surge to the next (January 31 to February 25). This female stopped cycling around 17 March, after which her hormone levels fluctuated slightly but never reached previous levels.

unknown reasons. We determined the end of cycling for each female as the point at which the P levels dropped to baseline levels but did not elevate again to the previous (ovulatory) levels. We calculated one to three ovarian cycles for each of the cycling females for a total of nine cycles, but only five of those were considered complete. A complete cycle was one that could be measured from one progesterone surge to the next [Strier et al., 2003]. The average length of a complete ovarian cycle was 20 ± 6.1 days (n=5; range=14–26 days). Complete cycle lengths did not differ significantly among females (χ^2=3.8, df=3, P=0.28). Figure 1 depicts one representative hormone profile for one of the subject females. Table II summarizes the ovarian cycle patterns for each female subject, including individual cycle lengths and the lengths of their cycling and noncycling periods during the course of the study.

Behavioral Differences Between Periovulatory and Nonovulatory Phases

Copulations occurred significantly more often in the periovulatory phases compared to the nonovulatory phases (mean paired difference (MPD)=0.247, P<0.001). Although we observed only three copulations among the cycling females, 100% of them occurred during the periovulatory phase of the cycle. Two of these three copulations involved alpha males and alpha females, and the third involved a subordinate male and a subordinate female (Fig. 2). Adult males also directed significantly more courtship displays toward cycling females during the periovulatory phases compared to the nonovulatory phases (MPD=0.475, P<0.001). A total of six courtship displays were observed during the

TABLE II. Summary of Female Cycling Patterns: January to June, 2002

Female	Group	Date of first analyzed samples	Date of 1st P surge	Date of 2nd (3rd if applicable)[a] P Surge	Length of complete[b] cycle 1/ cycle 2 (days)	Length of total cycling period (days)[c]	Date of cycling cessation	Length of non-cycling periods (days)[d]	Length of time since last infant born (mths)[e]
TI	CP	Jan 19	Feb 1	Feb 26/-	25/-	47	Mar 19	86	0
LI	CP	Jan 21	Mar 2	Mar 22/-	22/-	41	Apr 12	62	12
KL	LV	Jan 30	Jan 29[f]	Feb 26/-	26/-	48	Mar 16	73	12
BL	LV	Jan 29	Mar 11	Mar 25/ Apr 6	14/13	38	Apr 18	40	9

[a]Only BL had two complete cycles.
[b]A complete cycle was calculated from one progesterone (P) surge to the next.
[c]Length of cycling period was calculated from the first day of the P surge to the end of cycling when P levels decreased and did not rise again.
[d]Length of non-cycling period was calculated from the date of cycling cessation to the date the last sample was collected.
[e]This population of capuchins have a 2.5 yr interbirth interval [Fedigan, 2003]; gestation lengths are approximately 5.5 mths [Nagle and Denari, 1982].
[f]We have assumed that KL's first P surge occurred one day before the first sample was collected as she appeared to have started cycling at that time.
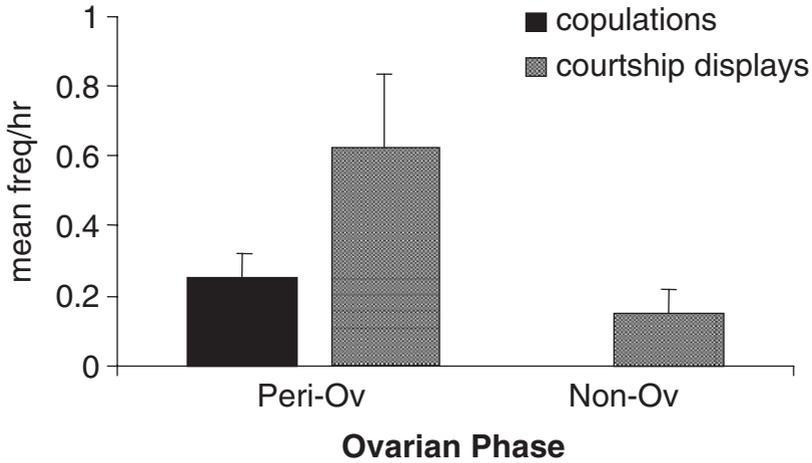
Fig. 2. Mean rate ($\pm$SE) of sexual behavior (copulations and courtship displays) during the periovulatory (Peri-Ov) and nonovulatory (Non-Ov) phases of the ovarian cycle.
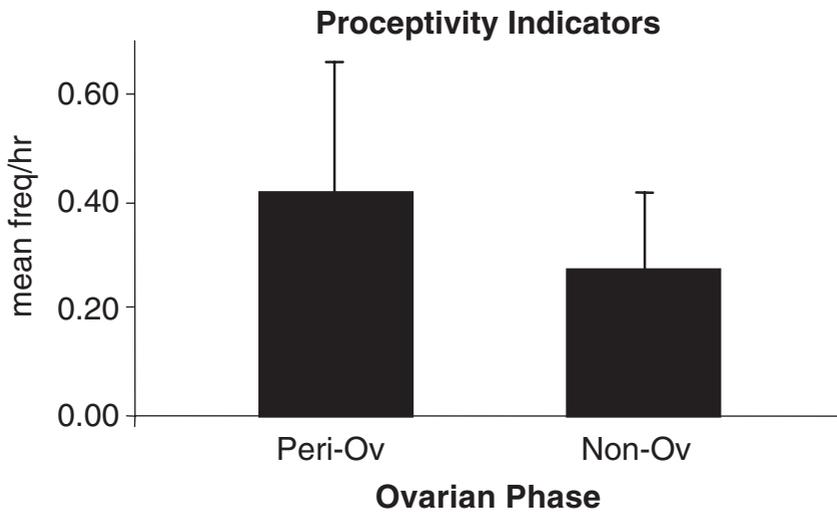

Fig. 3. Mean rate ($\pm$SE) of all proceptivity indicators (all directed approaches, follows, and grooms) from all subject females toward all adult males.

periovulatory phases. The alpha males were responsible for four of them (67%), and three of these were directed toward the alpha females.

Contrary to what we had predicted, none of the proceptivity indicators showed significant phase-related changes between the periovulatory and nonovulatory phases (Fig. 3). Adult females directly approached adult males at almost two times the rate during the periovulatory phases as compared to the nonovulatory phases (periovulatory: 1.03/hr; nonovulatory: 0.59/hr), but this difference was not significant. We never observed cycling females to either follow or direct a groom solicitation toward adult males in any phase of the cycle. We found that urine washing occurred at a significantly higher rate during the
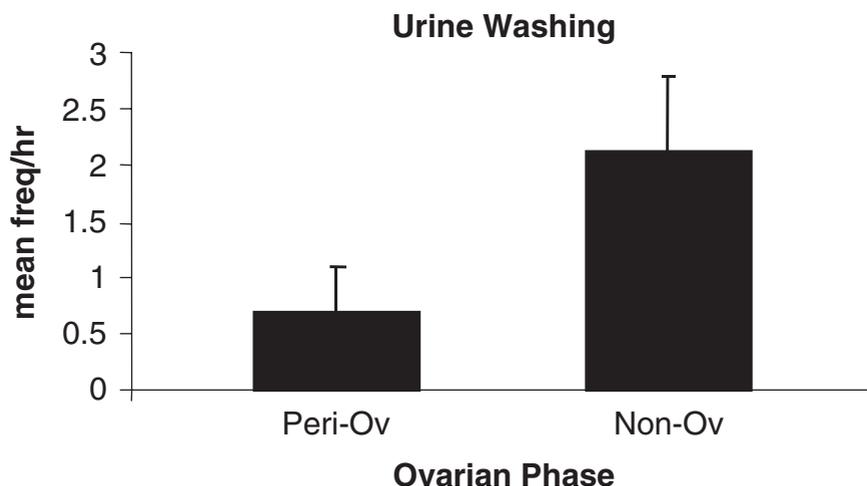
Fig. 4. Mean rate ($\pm$SE) of urine washing in the periovulatory and nonovulatory phases.

nonovulatory phases compared to the periovulatory phases (MPD$=-1.43$, $P<0.001$), which is the opposite of what we had predicted (Fig. 4).

Two of the behavioral attractivity indicators showed significant phase-related changes. The rate of received grooming bouts (the male grooms the female) and the rate of received follows (the male follows the female) occurred significantly more often during the periovulatory compared to the nonovulatory phases (grooming bouts: MPD$=0.377$, $P<0.003$; follows: MPD$=1.512$, $P<0.001$; see Fig. 5a and b). During both phases of the cycle, the alpha male was responsible for all grooming bouts directed toward all adult females.

We calculated Hinde's indices to determine which sex was responsible for maintaining proximity in each phase. A negative index suggests the male was responsible for maintaining proximity, and a positive index suggests the female was responsible. Indices could be calculated for six of the 12 possible male–female dyads for the periovulatory phases, and for seven of the male–female dyads in the nonovulatory phases. For some dyads, the male was never seen within the proximity of a subject female; therefore, indices could not be calculated for those dyads. In both phases of the cycle, adult males were responsible for maintaining proximity in the majority of the dyads (periovulatory: 67%; nonovulatory: 71%; see Table III). The remaining dyads were positive, which implies that the females were responsible for proximity in those cases. During the periovulatory phases, the alpha males were responsible for maintaining proximity in all cases, whereas they were only responsible for maintaining proximity in half of the cases during the nonovulatory phases. In contrast, when periovulatory females were responsible for maintaining proximity, they did so toward subordinate males; however, this could be measured in only two dyads.

DISCUSSION

Fecal Hormone Analysis and Interpretation of Hormone Profiles

The results from the hormone assays revealed that the collection and extraction techniques used in the field and laboratory were successful methods for
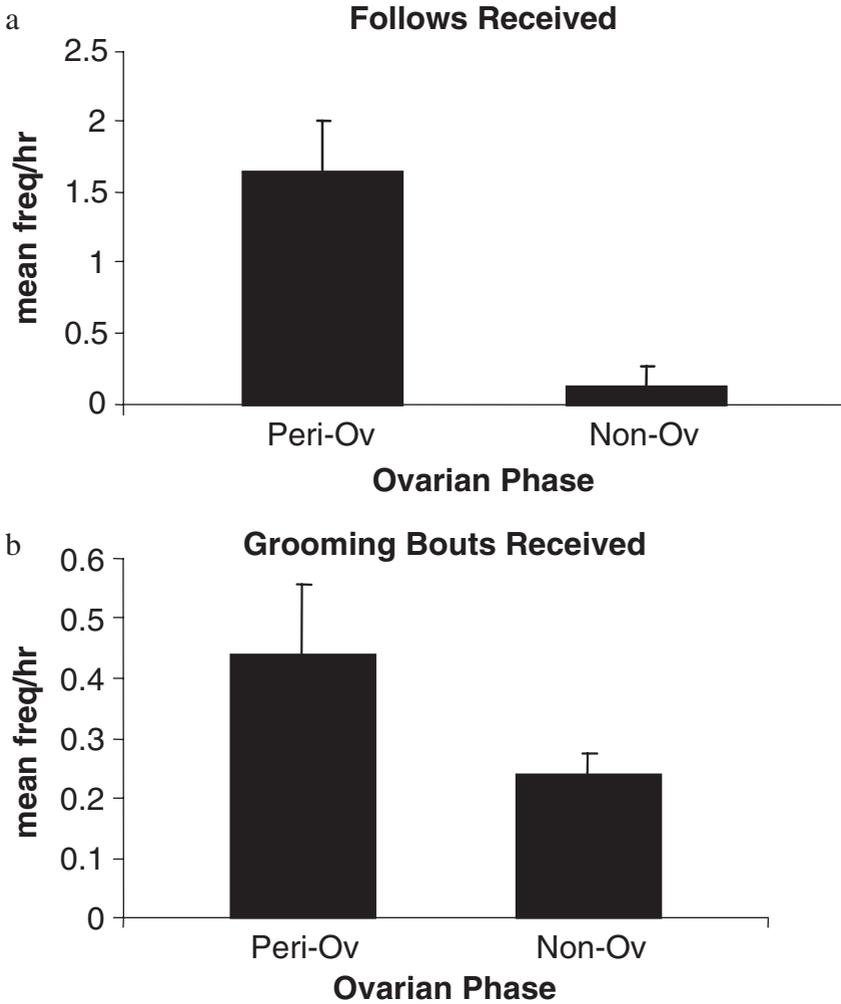
a
Follows Received

b
Grooming Bouts Received

Fig. 5. **a:** Mean rate ($\pm$SE) of follows received from adult males by subject females. **b:** Mean rate ($\pm$SE) of grooming bouts received from adult males.

studying ovarian steroids obtained from white-faced capuchin feces. The reproductive states of the female subjects were easily recognized from the hormone profiles (cycling, noncycling, or pregnant); however, among the cycling females the follicular and luteal phases of the ovarian cycle were not clearly distinguished. Therefore, we compared the periovulatory phases with the nonovulatory phases, a procedure also used by Carosi and Visalberghi [2002] in their study of capuchin sexual behavior across the ovarian cycle.

The four cycling females displayed one to three ovarian cycles before they stopped cycling. After their cycles stopped, their hormone levels fluctuated slightly but stayed low and never reached or sustained baseline levels compared to the profiles of the noncycling females. It was surprising to find that these females stopped cycling midway through the study, and that they did not start cycling again without becoming pregnant. These four females are likely to have

TABLE III. Hinde's Indices for Maintenance of Proximity*

	Periovulatory	Non-ovulatory
CP Alpha F[a]		
Alpha M[a]	−0.63	+0.25
Sub M	+0.25	−0.66
LV Alpha F		
Alpha M[a]	−0.30	−0.26
Sub M	−	−
Sub M	−	−0.40
Sub M	−	−
CP Sub F		
Alpha M[a]	−0.67	−0.75
Sub M	+0.43	−
LV Sub F		
Alpha M[a]	−0.05	+0.08
Sub M	−	−
Sub M	−	−0.08
Sub M	−	−

*A negative index implies the male is responsible for maintaining proximity; a positive index implies the female is responsible.
F, adult female; M, adult male; Sub, subordinate.
[a]denotes alpha individuals of CP and LV groups; other males listed are subordinate to the alpha male in each group.
−, Hinde's Index could not be properly calculated due to lack of data (i.e., some males were never involved in any approaches or leaves).

experienced ovulatory cycles again, later in 2002 during November and December, because they gave birth the following year between April and May 2003. The gestation length in our study species is assumed to be similar to that of *C. apella*–approximately 155 days, or 5.2 months [Nagle & Denari, 1982].

There are at least three possible explanations as to why these females ceased to cycle without becoming pregnant during our 2002 study period. First, Recabarren et al. [2000] described a period of time called ''lactational residual infertility'' in *C. apella*, which occurs subsequently to lactational amenorrhea. The latter occurs immediately after parturition when the ovary ceases to produce ovarian hormones. According to Recabarren et al. [2000], lactational residual infertility is a period of time when females may still be nursing and begin to cycle, but their hormone patterns are abnormal and most likely anovulatory. The suggested function of this extended infertile period is to increase the interval between pregnancies, as previously documented in apes (chimpanzees [Tutin & McGrew, 1973]), Old World monkeys (baboons [Altmann et al., 1978]), and some New World monkeys (howler and squirrel monkeys [McNeilly, 1994]). Recabarren et al. [2000] stopped measuring hormone levels after 150 days, so it is unclear whether their female capuchins also stopped cycling before conceptive cycles started. One finding from our study that does not support this explanation is that the female Timone was nulliparous and thus could not have been experiencing residual infertility.

A second explanation is that female white-faced capuchins may mate year-round but conceive seasonally. Twenty years of birth data and nearly 100 recorded births show that conceptions peak in October through December, with a secondary peak in July, and that significantly more infants are born in the dry than the wet season [Fedigan, 2003] (Fedigan unpublished results). Our study

site has distinct dry and wet seasons, and it is possible that conceptions are more likely to occur at the end of the wet season, when mothers have had access to abundant resources and are physically healthy enough to carry a developing fetus. This pattern would then permit infants to be weaned when resources are replenished in the next wet season [Di Bitetti & Janson, 2000]. The seasonality hypothesis may explain why cycling females did not conceive during our dry season study period, but it does not entirely explain why they started to cycle and then stopped. Even though there is a birth peak in this population of capuchins, females still conceive and give birth in all months of the year, so we cannot conclude that they exhibit true reproductive seasonality.

A third (and related) explanation is that the females may have been physiologically stressed during the dry season study period, which disrupted their normal ovarian function. A component of the stress response is the release of glucocorticoids (i.e., cortisol), and when cortisol remains in the blood at chronically high levels it may have detrimental effects on many physiological systems, including reproduction [Sapolsky, 2002]. The dry season is an extremely stressful time for capuchins because they must drink water every day. By the end of April, most ground and tree waterholes have dried up and the midday temperature can soar to 40°C. Moreover, since there are few water holes available, there is a tendency for more intergroup encounters between capuchin groups to occur, and these encounters can facilitate group takeovers by extragroup males. Aggressive takeovers almost always occur during the dry season and often lead to the death or disappearance of the youngest infants, including incidents in which males have been seen to kill infants [Fedigan, 2003, Manson et al., 2004]. Social conflicts may lead to stress responses; however, very little is known about how social stress affects females, and consequently the ''stress'' explanation is presently under investigation in this population of capuchins.

Sexual Behavior and Mate Choice

Our behavioral analyses suggest that dominance plays a major role in terms of mate choice for both males and females in this study population. Alpha males more often choose the alpha females to solicit, and alpha females more often choose the alpha male as a mating partner. Female brown capuchins (*C. apella*) are also known to preferentially choose the alpha male as a mate [Janson, 1984]. However, *Cebus apella* females are reported to mate with subordinate males after their most fertile days have passed, near the end of their periovulatory period [Carosi et al., 1999; Janson, 1984; Linn et al., 1995]. Unfortunately, we are not able to suggest that the same behavior occurs in *C. capucinus*. Given that we observed copulations only rarely among the cycling females, and only one female (a subordinate) was observed mating with a subordinate male, it would be premature to suggest that mating with the alpha male was restricted to the females' most fertile days. In fact, subordinate males directed very few sexual solicitations toward any adult female during their periovulatory phases, and neither of the alpha females mated with them. This suggests that the alpha males may have exclusive mating rights to conceptive females, and that alpha females preferentially choose the alpha males as mates. In further support of this suggestion, recent paternity studies on this population show that alpha males sire most of the infants within their groups [Jack & Fedigan, 2003, in press]. Subordinate males do mate with adult females, but they do so more often with pregnant or ''postconceptive'' females [Carnegie et al., in press].

Proceptivity and Attractivity: Indicators of Ovarian Phase

The increase in sexual behavior during the periovulatory phases suggests that adult males are at least somewhat aware of the onset of the females' fertile phases. Given that white-faced capuchins fail to display any obvious morphological cues to their conceptive phase, we predicted that adult males would respond to an increased rate of female proceptive behavior during their periovulatory periods. However, we found that proceptive behavioral indicators occurred at only slightly higher rates during the periovulatory compared to the nonovulatory phases. Females did tend to approach males at higher rates in the periovulatory phase, but this was not a significant trend and cannot be used as a reliable indicator of proceptivity. In comparison with many primate species that display overt proceptive behaviors around the time of ovulation (e.g., *Cebus apella* [Carosi et al., 1999; Linn et al., 1995], *Macaca fuscata* [Enomoto et al., 1979], and *Cercocebus torquatus atys* [Gordon et al., 1991]), female white-faced capuchins are very inconspicuous in this respect.

The rate of urine washing could also function as an indicator of the ovulatory period, and therefore we predicted that this behavior would occur at a higher rate during the periovulatory phase. However, we found the converse–that it occurred significantly more often during the nonovulatory phases. This suggests that urine washing may have functions other than communicating reproductive state in this species. Alternatively, it is possible that periovulatory females reduce their rates of urine washing in order to lower the potential for communicating their fertile status to males via chemical compounds in their urine.

Attractivity indicators appeared to provide the best estimate of the female's ovarian state, and during the periovulatory periods these behaviors were almost exclusively displayed by the alpha males. Specifically, the rate at which the alpha males followed the females was the best behavioral indicator of the periovulatory phase. Alpha males also directed more grooming bouts toward females when they were periovulatory, which is noteworthy because in our study animals, adult males very rarely groom females (Fedigan, unpublished results). This initiation of sexual behaviors by males in *C. capucinus* contrasts greatly with the pattern in *C. apella* in which males become sexually attracted to females only after the females have intensely solicited them for mating. In white-faced capuchins, the high rate of male following and the increased rate of male grooming toward periovulatory females does not appear to be preceded by any conspicuous female behavioral signals.

Males maintained proximity to females (rather than vice versa) in the majority of cases during both the periovulatory and nonovulatory phases. During the periovulatory phase, the alpha male was responsible for maintaining proximity to all females (all dyads), but only for half of the dyads during the nonovulatory phases. These findings lend support to what we also found for pericopulatory behaviors–that alpha males mated with, followed, and initiated grooming bouts with females more often when the females were periovulatory than when they were not. The fact that the alpha males were not entirely responsible for maintaining proximity to females during their nonovulatory periods suggests that they can distinguish between periods when the female is fertile and when she is not. We also found that females were 100% responsible for maintaining proximity to subordinate males during their periovulatory phases. This could only be measured in two of the male–female dyads, but suggests the possibility of female mate choice and a strategy to limit alpha male monopolization.

Overall, our study suggests that while female white-faced capuchins may slightly increase their frequency of affiliative proceptive behaviors toward males during their periovulatory phase, this is not done to any significant extent. Instead, it was the male's persistent and overt behaviors toward the female that best indicated she was in the periovulatory phase of the cycle. We cannot rule out the possibility that even very subtle increases in proceptive behaviors may act as the cue for the males, but to support this notion more data must be collected from a larger sample size of cycling females and over a longer amount of time (including phases in which conceptions occur). It is also possible that males detect the periovulatory phase from olfactory cues (pheromones released by an ovulating female). Finally, there is the possibility that females emit specific (but undetectable to the human ear) vocalizations as a reproductive cue. All of these possibilities are under further investigation. At this point, we conclude that the female white-faced capuchins in our study groups do not provide any obvious behavioral cues to the males and thus may be ''concealing'' their ovulatory periods (or at least rendering them less noticeable), but that the males are detecting their periovulatory phases nonetheless.

van Schaik et al. [2000] argued that female primates face a dilemma as to how to confuse paternity in order to lower infanticide risk while simultaneously giving certain males a high probability of paternity, thus ensuring that the ''best males'' father their infants. Perhaps female white-faced capuchins fail to broadcast widely detectable visual cues in order to confuse paternity and lower infanticide risk, but manage to convey subtle cues to selected males. Burt [1992] and van Schaik et al. [2000] argued that truly concealed ovulation does not exist in primate species. Instead, selection may have operated on females to make their ovulations unpredictable. This means that there is a large degree of variance in the duration of the follicular phase, and a good (but not perfect) correlation between proceptive behaviors and ovulation. Like concealed ovulation, unpredictable ovulation may function to confuse paternity in multimale groups [Nunn, 1999; van Schaik et al., 2000]. Our study suggests that white-faced capuchin females do not successfully conceal their ovulation, but neither do they behaviorally advertise it. We do not yet have sample sizes large enough to determine the degree of variation in the duration of follicular phases and to assess the hypothesis of ''unpredictable ovulation.'' However, we do know that pregnant females copulate with males at higher rates than cycling females [Carnegie et al., in press], and that postconceptive mating is likely to be another counterstrategy against infanticide [Manson et al., 1997]. Further studies of white-faced capuchin reproductive biology are ongoing to better elucidate what females communicate and what males know about ovulation, and whether there are seasonal effects on reproduction, and/or periods of residual infertility in this study population.

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