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Research Article

Variation on Breeding Pattern of Rhesus Monkeys (*Macaca mulatta*) in Captivity

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Abstract

Rhesus monkey (*Macaca mulatta*) display a distinct seasonal pattern of breeding activity which restricts birth to specific times of the year, complete cessation of ovulation and decrease in male sexual activity which leads to marked seasonal variations. The study aimed to develop a novel method to control seasonal breeding among rhesus macaques which is eco-friendly, sustainable, cost-effective and ethically acceptable. The observational cross-sectional study was conducted where a total number of 44, 68, 123, 175, 225 rhesus macaques were studied in the year 2006, 2007, 2008, 2009, 2010 respectively. Macaques were fortified with a standard balanced diet in the form of pellet and supplemented with 100gms of pumpkins and seasonal fruits and all together 14 hours (natural and artificial) of light per day was provided. The result of this study indicates that if these macaques are exposed to ambient photo period and given proper nutritional diet, then parturition can occur throughout the year. In 2006, no distinct variation in the breeding pattern was observed but as the year passes by from 2006 to 2010, parturition were found occur almost in every month of the year even during their breeding months (October, November and December) and the total number of off-spring born was increased gradually from 2 to 45. Hence the present data support the hypothesis that exposure to a non-seasonal environment, in terms of photoperiod and nutritional diet, can advance the parturition and can control the seasonal breeding among rhesus macaques. However, future work must proceed to observe the efficiency of light and diet along with other factors like ambient temperature, rainfall, humidity and vegetation on parturition, tempo of maturation and ovulatory menstrual cycle among rhesus macaques.

Keywords: *Rhesus macaques; Seasonal Breeding; Photoperiod; Nutritional Diet; Parturition*

Abbreviations

eCG: Equine Chorionic Gonadotropin; TSH: Thyroid Stimulating Hormone; DIO3: Type III Iodothyronine Deiodinase; DIO2: Type II Iodothyronine Deiodinase; T3: Triiodothyronine; MBH: Medio-basal Hypothalamus; $CaCO_3$: Calcium Carbonate; NBRC: Nepal Biomedical Research Centre; AI: Artificial Insemination

Introduction

Rhesus monkey (*Macaca mulatta*) is the most common and least concerned non-human primate also one of the best-Known Simian species belonging to the family Cercopithecidae. They are widely distributed in South East Asia from northern Afghanistan in the east and

south to the Godavari River in India, Thailand, Laos, Cambodia, Vietnam, Nepal, Bangladesh, Tibet and China in the west. In Nepal they are found in tropical and sub-tropical forests ranging from Terai and mid- hills [13] and are capable to adapt wide varieties of habitats and elevation ranging from high heat to snow fields to cities [12]. Flexibility in diet patterns plays a pivotal role in their survival which includes fruits, seeds, flowers, buds, leaves, barks, roots, angiosperms fungi, insects, grubs and molluscs [9]. An adult rhesus has a stoutly built body having medium size tail with large cheek pouches, prominent ischial callosities, variable sex swelling, and marked sexual dimorphism male weighing 6 - 12 kg and female weighing 4 - 9 kg. Rhesus monkeys are characterized by a high degree of social flexibility having four types of social groups ranging from 8 - 180 individual of both sexes [7].

Rhesus macaques (*Macaca mulatta*) share so many biologic characteristics with humans which makes them uniquely valuable animal models for studying human health and disease. Particularly, the reproductive anatomy and menstrual cycles of female rhesus macaques are similar to those of women [6]. The onset of puberty in female rhesus monkeys is modulated by both internal and external factors whereas the male's testosterone levels increase at the onset of the breeding season in response to some stimulus provided by the ovulating females. The heightened testosterone levels, in turn, stimulate males to become fertile and sexually active, and breeding ensues [8] hence they display a distinct seasonal pattern of breeding activity in outdoor environments [10], with ovulation restricted to the fall and winter and births to the spring and summer. During this period, skin of the perineum and caudal aspect of the hind legs become edematous-swelling subsides- affected skin takes red coloration [16]. Seasonal breeding in rhesus monkeys restricts birth to specific times of the year, complete cessation of ovulation and decrease in male sexual activity which leads to marked seasonal variations.

Current breeding scheme rely on the use of exogenous hormones mainly the Equine Chorionic Gonadotropin (eCG), which is now considered hazardous to the people and to the environment. From a technical standpoint, repeated administration of eCG leads to the production of anti-eCG antibodies, thus reducing treatment efficacy and leading to a decrease in fertility. Thus the development of novel methods to control seasonal breeding has become urgent which is eco-friendly, sustainable, cost-effective and ethically acceptable [3,4].

Little is known about environmental factors regulating seasonal breeding of primates, but several factors such as photoperiod, dietary food supply, temperature, and rainfall, have been implied [10]. However, it has been reported that when these rhesus macaques are kept under controlled environmental condition, they can ovulate and conceive in every month of the year [11]. The photoperiod or light treatment along with proper supply of dietary food plays an important role in the control of the seasonal breeding cycle of primates. During this process, melatonin acts within the pars tuberalis to activate TSH production which decreases Type III iodothyronine deiodinase (DIO3) and increases Type II iodothyronine deiodinase (DIO2), leading to a local increase of T3 production within the medio-basal hypothalamus (MBH). This mechanism brings together the long-recognized roles of TH and melatonin in the control of seasonal functions by light treatment [4].

The proper dietary food which contains standard concentration of soya meal, sugar, salt, wheat, soya bean oil, skimmed milk, wheat bran, ground corn, digestible crude protein, vitamins, minerals, $CaCO_3$ collectively called as pellet food supplemented with sweet potatoes, pumpkins or fruits and meager amount of 2, 3, 7, 8-tetrachlorodibenzo-p-dioxin (TCDD) when given to female rhesus monkey, their occurs hormonal alteration and are able to conceive, experienced an uneventful gestation and give birth to a well developed offspring throughout the year [2]. These activation of reproductive axis outside of this breeding season offer many advantages which includes (i) synchronization of births over a short time period, scrutiny at birth to reduce mortality, (ii) reduction of unproductive periods (puberty, seasonal anestrus, postpartum anestrus), (iii) ability to program births at specific times of year to adjust to seasonal variations [4].

Aim of the Study

The study aimed to examine how the controlled light and standard dietary food supplement influenced the timing of menarche and ovulation on rhesus macaques and how these factors were allied to improve fertility and differential rates of growth.

Materials and Methods

Study design, site and duration

The observational cross-sectional study was conducted. The study was carried out in Nepal Biomedical Research Centre (NBRC) Lele, Lalitpur, Nepal for five years starting from 2006 - 2010.

Figure 1: *Nepal Biomedical Research Centre (NBRC) Lele, Lalitpur.*

Animals

All the rhesus monkeys used in this study were brought from various eco zones which had obtained the animals by capturing them in their natural environment. A total of 44, 68, 123, 175, 225 rhesus macaques were studied in the year 2006, 2007, 2008, 2009, 2010 respectively all of them were of 2.5 years of age or above having normal reproductive history. The animals were kept into the individual cage in the ratio of 1:10 (male: female).

Figure 2: *Rhesus macaque (Macaca mulatta) playing around in the cage.*

Experimental designs

The variation on breeding pattern of rhesus monkey were observed on the basis of two main factors

(i) Proper nutritional diet

(ii) Light

Proper Nutritional diet

Macaques were fortified with following standard balanced diet in the form of pellet and supplemented with 100 gms of pumpkins and seasonal fruits. Euroguard treated water was supplied throughout the day.

Ingredients	Percentage
Wheat	27.5
Soyameal	18
Ground corn	26.5
Skimmed milk	1
Wheat bran	8
De-oiled rice bran	8
Sugar	2.9
Soyabean oil	3.6
Salt	0.3
Digestible crude protein	1
Vitamins	1
Minerals	1
$CaCo_3$	1.2

Table 1: *Standard diet plan followed for the preparation of pellet food supplement.*

(A) (B)

Figure 3: *(A) Rhesus macaques having seasonal fruits and (B) Staff members involved in cleaning the food supplements with euro guard water.*

Light

All together 14 hours (natural and artificial) of light per day was provided. During winters 10 hours of natural light and 4 hours of artificial light was provided whereas during summer 12 hours of natural and 2 hours of artificial light was maintained.

Observation for variation on breeding pattern

Changes like sexual swelling of the face and their sex skin which turns palest at the time of menstruation and slightly swollen as the animal nears ovulation was observed on the basis of which parturition occurs.

Results

After the provision of proper nutritional diet and 14hrs of light daily, parturition was found to occur throughout the year.

(A) (B)

Figure 4: *(A) Pregnant rhesus monkey having swelling face and change in their sex skin, (B) New born rhesus macaques (parturition).*

Number of off-springs born in year 2006

A total of 44 rhesus macaques were studied in 2006, where total number of birth was 2 in the month of February which is the normal month for parturition.

Figure 5: *A total of 2 rhesus macaques born in the month of February, 2006.*

Number of off-springs born in 2007

The total number of birth was found to increase slightly (i.e. 2 to 11) from 2006 to 2007. From February to July the number of birth was 1, 3, 2, 1, 3 and 1 respectively.

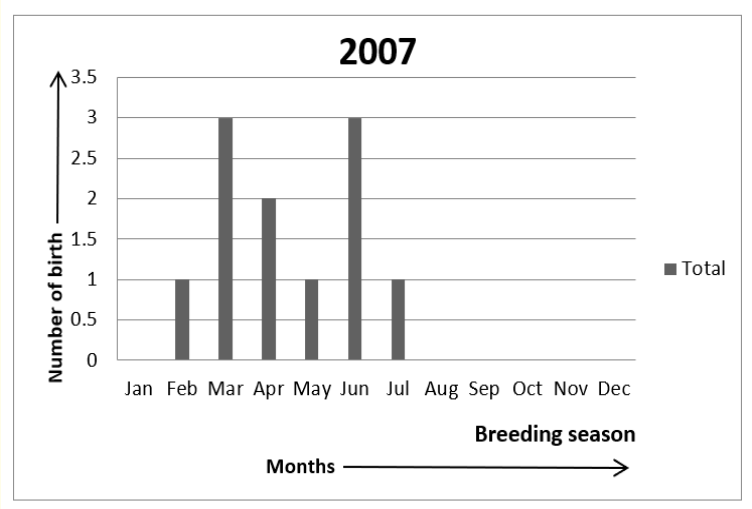

Figure 6: *A total of 11 rhesus macaques born in the month February to July, 2007.*

Number of off-springs born in 2008

During 2008, a total of 123 rhesus monkeys were studied, and the total number of birth was increased rapidly from 11 to 35. After the provision of standard balanced diet and light these monkeys were able to parturition even during their breeding month.

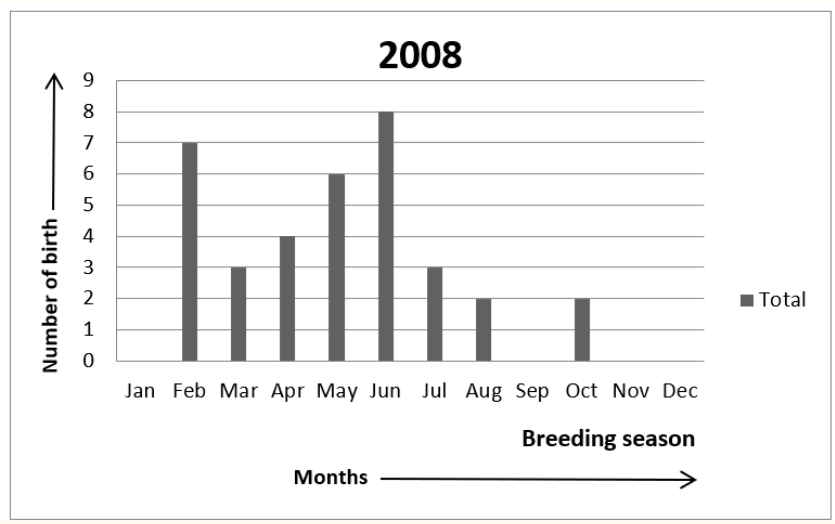

Figure 7: *Increased number of births observed during February to October 2008.*

Number of off-springs born in 2009

Almost in every month, parturition was observed and the total number of birth was 38. The highest number of birth was 8 i.e. during June. Even in September and October (breeding season) a total of 5 macaques were born.

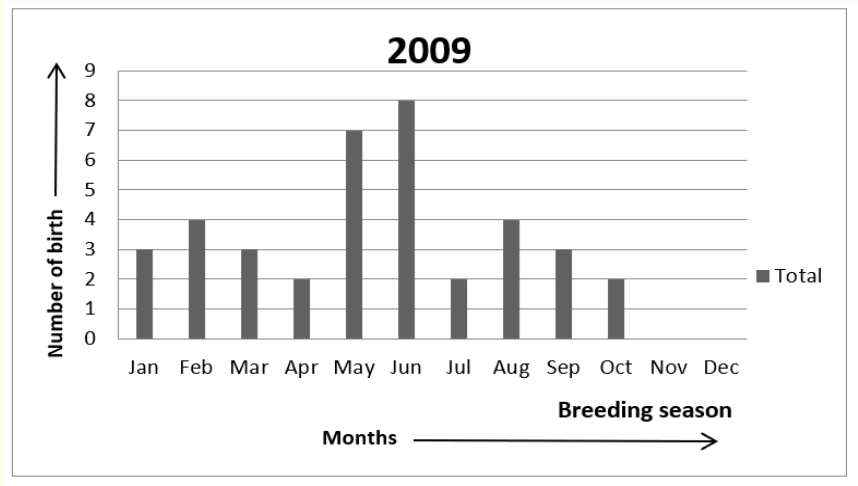

Figure 8: *Parturition observed almost in every month and the total of 38 macaques born during 2009.*

Number of off-springs born in 2010

A total of 225 macaques were studied in 2010, in every month even during their breeding season parturition was observed and the number of off-spring born was escalated from 38 to 45. The highest number of birth was observed in July (i.e. 9) and the lowest number of birth was observed in January, September and November (i.e. 2).

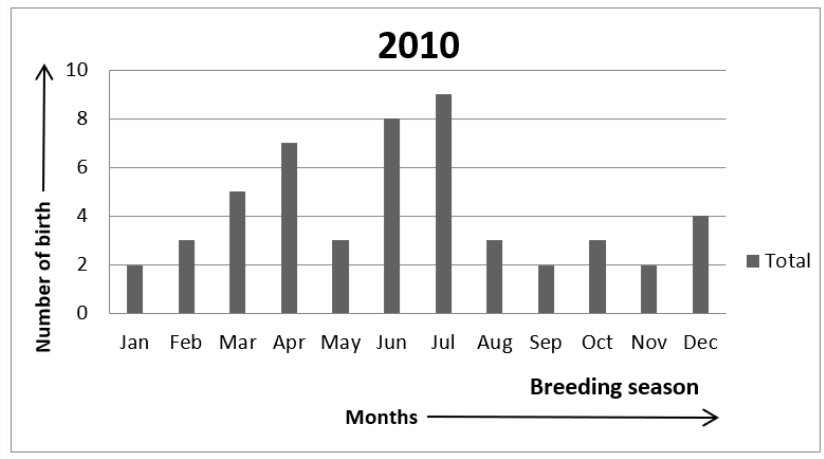

Figure 9: *Parturition observed in every month of the year.*

Comparison of parturition in rhesus macaques (2006-2010)

During 2006 - 2010 the number of birth was gradually increased from 2 to 45. In 2006 the number of birth was only 2 i.e. in the month of February. As the year increases, the parturition also increased to 45 and occurred in every month (even in their breeding months) in the year 2010 due the provision of proper diet and light. The highest number of births was observed in the month of June and July whereas the lowest was observed January, September and October.

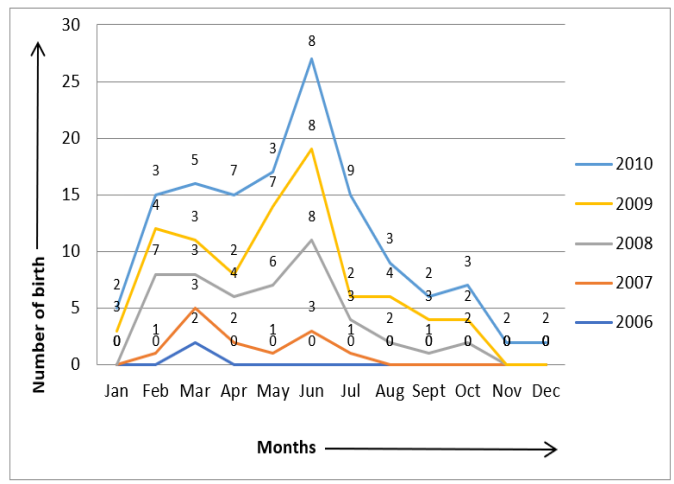

Figure 10: *Comparison of parturition in rhesus macaques from 2006 to 2010.*

Discussion

Study observing the breeding seasonality of rhesus macaques is limited in a developing country like Nepal and this work is the first to observe the variation in their breeding pattern with the provision of ambient photoperiod and standard nutritional diet. Observational study was carried out where 44, 68, 123, 175, 225 rhesus macaques were studied in the year 2006, 2007, 2008, 2009 and 2010 respectively with the ratio of 1:10 (male: female) and this study has demonstrated that if these macaques are exposed to ambient photoperiod and given proper nutritional diet, then parturition can occur throughout the year.

Rhesus macaques display a distinct seasonal pattern of breeding activity i.e. they breed mainly from September to January and parturition occurs from February to July. In the first year of our study i.e. in 2006, no distinct variation in the breeding pattern was observed. During that year only 2 macaques were born in the month of February which is the normal month for parturition. This may be due to slow adaptation to a new stable environment. In 2007, a slight change was observed, in that year parturition occurred from February to July and the total number of off-spring was increased to 11. Similarly, in 2008 the total number of birth was increased rapidly from 11 to 35. Macaques were born even during their breeding month (Figure 7). Meanwhile in the 2009 and 2010, parturition occurred almost every month of the year even during their breeding months (October, November and December) and the total number of off-spring born was increased gradually from 35 to 45. During that year the highest number of birth was observed in May, June and July.

This variation in their breeding pattern may be due to the melatonin production, this melatonin acts within the pars tuberalis of pituitary to activate TSH production which decreases Type III iodothyronine deiodinase (DIO3) and increases Type II iodothyronine deiodinase (DIO2), leading to a local increase of T3 production within the medio-basal hypothalamus (MBH). Other reason might be due to their

imitational or observational learning behavior; they have does tendency to observe the act or behavior of human or their same species and further change their own which can act as a sexual motivation. According to Deborah., *et al.* (1980) [5] reported that housing rhesus macaques under conditions that closely simulate the temperature, humidity and lighting during the breeding season results in establishment of ovulatory menstrual cycles within 2 months from their natural environment.

Our study contradict with the study carried out by Masumi., *et al*. (1990) [11] where the sexually mature female Japanese monkeys were exposed to alternating four-month periods of short days (8L16D) and long days (16L8D) with proper standard food supplements and demonstrated that their breeding seasonality was not influenced by the suitable photoperiod. This variation in the result might be due to the difference in the species of macaques. Similarly, Wehrenberg and Dyrenfurth (1983) [15] in their study maintained macaques *(Macaca mulatta and M. assamensis)* on a 12L :12D light cycle for 4 years and showed that light alone doesn't regulate seasonal breeding in macaques other factors should also be considered.

Meanwhile in the study conducted by Wilson., *et al*. (2015) [16] on how the unseasonal environment influenced the timing of menarche and first ovulation in rhesus monkeys and how these events were related to differential rates of growth revealed that when the female macaques are raised in the artificially maintained unseasonal environment exhibited a significantly earlier age at both menarche and first ovulation. Similarly, Vandenbergh and Vessey (1968) [14] reported that seasonal breeding in free-ranging rhesus monkeys was correlated with rainfall and vegetation. In a study conducted by Bank and AynedJlan (1963) [1] where a proper diet consisting of potatoes, breads, carrots, root and green vegetables and fresh fruits was given and as result of which theses macaques were found to have good health and the fertility of the female breeding macaques where also enhanced.

The results of the present study indicate that the type of environment in which animals are raised has a profound effect on their breeding pattern hence summarizes the development of a novel method to control seasonal breeding among rhesus macaques. The present data support the hypothesis that exposure to a non-seasonal environment, in terms of photoperiod and nutritional diet, can advance the parturition throughout the year. However, these light treatments do not allow efficient synchronization of ovulation hence cannot be used for AI protocols. Nowadays, the implementation of light treatments is combined with the male effect or hormonal treatments for AI purposes. On the other hand, the present results justify the need for studies in the field, to observe the efficiency of light treatment and nutritional diet along with other factors like ambient temperature, rainfall, humidity and vegetation.

Conclusion

The present study revealed that rhesus macaques being a strict seasonal breeder can give birth throughout the year if they are provided with ambient light and standard nutrition diet. In this study, an endeavor was made to observe the effect of light and proper diet on breeding pattern of rhesus macaques and from this conducted research; it has demonstrate that from the study period of 2006 to 2010, the parturition among the studied rhesus monkeys escalated and found to occur in every month of the year. From a practical point of view, the use of photoperiod to control seasonal breeding among macaques is simple, cost-effective, and limits the use of exogenous hormones. It can be combined with other various factors to maximize its impact. Nevertheless, it was just an observational study where the effect of light and diet was guarded on the basis of parturition. However, future work must proceed to evaluate the effect of light and diet along with other factors like ambient temperature, rainfall, humidity and vegetation on parturition, tempo of maturation and ovulatory menstrual cycle among rhesus macaques.

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