

The feeding behaviour and phytochemical food content of grey-shanked douc langurs (*Pygathrix cinerea*) at Kon Ka Kinh National Park, Vietnam

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Summary

The grey-shanked douc langur¹ is a critically endangered and endemic leaf-eating primate to Vietnam. The population of the species is decreasing and highly fragmented due to hunting pressure and loss of habitat. The species is restricted to several provinces in the Central Highlands. Kon Ka Kinh National Park is home of less than 250 individuals. Currently, there is insufficient understanding about the feeding behaviour and phytochemical content in food selection among the species. This study was conducted in Kon Ka Kinh National Park from February 2009 to June 2010. We collected 212 hours of feeding behaviour data. Grey shanked douc langurs ate 135 plant species of 44 plant families during the study period. The plant species *Pometia pinnata* is the most preferred item. We collected 33 plant samples from eaten species for phytochemical analysis which revealed that protein comprised of 11.4% of dry matter, lipids 2.6%, minerals 5.0%, sugar 4.9%, starch 12.8%, and Neutral Detergent Fiber (NDF) 40.8%. Protein in leaves is higher than in whole fruits: 12.8% and 8.3% respectively. However, protein in young leaves and mature leaves was not significantly different; 14.1% and 11.5% respectively.

Thành phần hóa dinh dưỡng và tập tính ăn của loài voọc chà vá chân xám (*Pygathrix cinerea*) tại Vườn Quốc Gia Kon Ka Kinh, Việt Nam

Tóm tắt

Voọc chà vá chân xám là loài khỉ ăn lá đặc hữu và cực kỳ nguy cấp của Việt Nam. Quần thể của loài trong tự nhiên đang bị suy giảm do tình trạng săn bắt và mất môi trường sống. Hiện nay, loài chỉ phân bố tại một số tỉnh miền Trung và Tây Nguyên, Việt Nam. Vườn Quốc gia Kon Ka Kinh là nơi sinh sống của gần 250 cá thể voọc chà vá chân xám. Những dẫn liệu khoa học về tập tính sinh thái dinh dưỡng của loài hiện vẫn còn rất hạn chế. Nghiên cứu này được tiến hành từ tháng 2 năm 2009 đến tháng 6 năm 2012. Khoảng 212 giờ quan sát trực tiếp tập tính ăn của loài ngoài tự nhiên đã được thu thập. Kết quả cho thấy, loài voọc chà vá chân xám ăn 135 loài thực vật thuộc 44 họ. Trong đó loài

¹ There remains some scholarly debate about the common names for the *Pygathrix* species, although this does not affect systematic placement. For the purpose of consistency within the Vietnamese Journal of Primatology, douc langur is used for all *Pygathrix* species. Please refer to the article in this issue for a discussion of vernacular names [Nadler (2012): Why Sea Lions don't catch Zebras—Thoughts about common names of Indochinese primates. Vietnamese J. Primatol. 1(2), 3-5].

Pometia pinnata là loài thực vật được ưa thích nhất của loài tại khu vực nghiên cứu. 33 mẫu thức ăn của loài voọc chà vá chân xám đã được thu thập để phân tích thành phần hóa dinh dưỡng. Kết quả cho thấy, các mẫu thức ăn chứa 11,4% là protein, 2,6% chất béo, 5,0% chất khoáng, 4,9% đường, 12,8% tinh bột, và 40,8% chất xơ. Hàm lượng protein trong lá nhiều hơn trong quả với 12,8% và 8,3% hàm lượng chất khô. Hàm lượng protein trong lá non (14,1%) nhiều hơn trong lá già (11,5%), tuy nhiên sự sai khác không có ý nghĩa thống kê.

Introduction

Food components of colobines consist of leaves, fruits (including seeds), flowers, lichen, bark and partly animals (insects). According to Yeager & Kool (1997), colobines select foods of high nutritional value, and young leaves are preferred over mature leaves. Milton (1998) argued that young leaves contain more protein than mature leaves, and that young leaves contain less fiber. Therefore, young leaves tend to be consumed more than mature leaves.

Colobines are folivorous primates, but in some species their diets also contain large proportions of unripe fruits and seeds. For example, Davies (1991) found that *Presbytis rubicunda* eat over 80% seeds in some months. In Africa, other colobines also feed heavily on fruits such as *Colobus satanas* in Cameroon and Gabon (McKey, 1978; McKey et al., 1991; Harrison, 1986) or *Colobus polykomos* in Sierra Leone (Dasilva, 1992).

Diet and feeding behaviour of the genus *Rhinopithecus*, *Pygathrix* and *Nasalis* have been studied by Yeager (1989), Boonratana & Le Xuan Canh (1993), Kirkpatrick (1998), Lippold (1998), Otto (2005), Hoang Minh Duc (2007), Ha Thang Long (2009), and Grüter (2009).

Black-shanked douc langurs in Nui Chua National Park and Binh Chau Phuoc Buu National Park eat leaves, flowers, fruits and seeds from at least 152 plant species of 37 plant families (Hoang Minh Duc, 2007). Red-shanked douc langurs eat leaves and fruits of 50 plant species (Pham Nhat, 1993). Otto (2005) provided a list of 87 species from 36 families consumed by the red-shanked douc langurs, but the list of feeding species was collected in captivity, containing many species of limestone areas around Cuc Phuong National Park. Feeding behaviour of the grey-shanked douc langurs was first studied by Ha Thang Long (2009) in Kon Ka Kinh National Park and the author found that the species fed on 166 plant species belonging to 40 plant families. In terms of food items, the grey-shanked douc langurs ate 49.5% young leaves, 21.9% ripe fruits, 19.1% unripe fruits and only 9.3% mature leaves. This study aims to provide further understanding of grey-shanked douc langurs feeding behaviour as well as the phytochemical contents of their food items.

Material and methods

Study site

Kon Ka Kinh National Park is located in north-east Gia Lai Province (14° 09' – 14° 30'N and 108° 16' – 108° 28'E) with an area of 41,710 ha (Fig.1). The park belongs to Kon Phe, Dak Roong and Kroong Communes, K'Bang District, and Ha Dong and Ayun Communes, Mang Yang District, Gia Lai Province. Altitude ranges from 570 m along Ba River to 1,748 m at Mount Kon Ka Kinh (Le Trong Trai et al., 2000). The rainy season lasts from May to November. Average annual rainfall is 1,700 mm; the peak is in July or August with 400-450 mm/month. The dry season starts in December and ends in April. The peak of the dry season is in January or February with zero rainfall. Eighty percent of Kon Ka Kinh National Park is montane and lowland forest (33,565 ha).

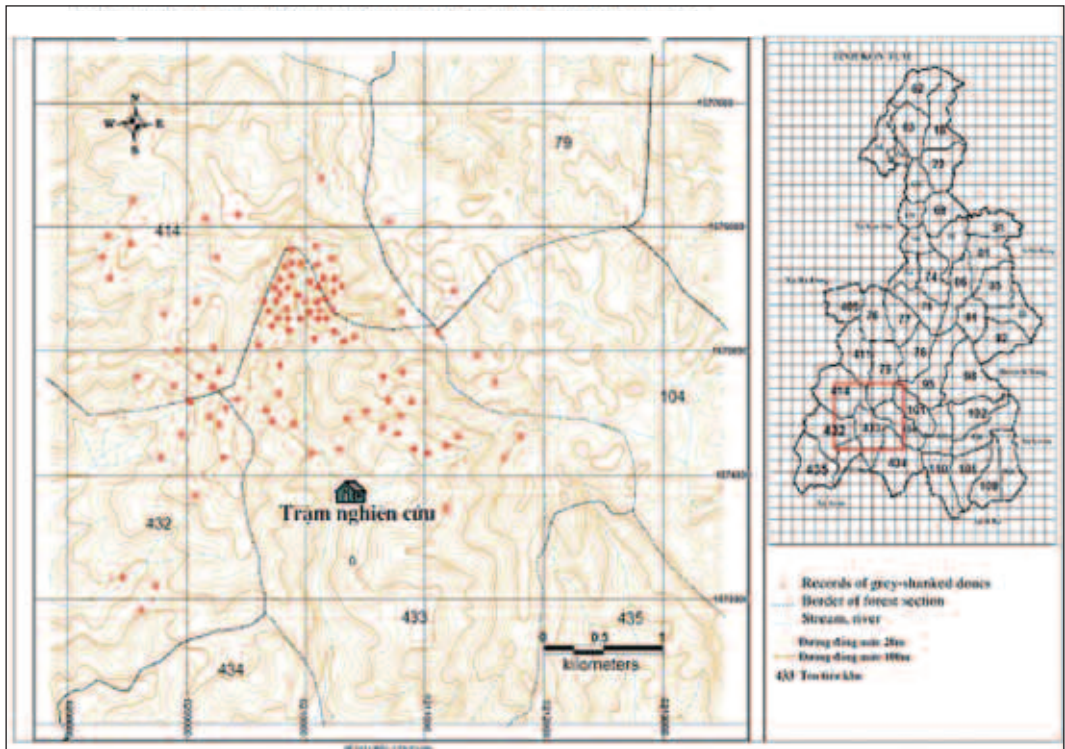


Fig.1. Study area in Kon Ka Kinh National Park.

There are seven species of primates recorded in Kon Ka Kinh National Park, including the northern yellow-cheeked gibbon (*Nomascus annamensis*), grey-shanked douc langur (*Pygathrix cinerea*), stump-tailed macaque (*Macaca arctoides*), pig-tailed macaque (*M. leonina*), rhesus macaque (*M. mulatta*), pygmy loris (*Nycticebus pygmaeus*), and northern slow loris (*N. bengalensis*). The population of grey-shanked douc langurs in the national park is less than 250 individuals (Ha Thang Long, 2009), which represents the largest known population.

Behavioural observation

Behavioural observations were made on different family units of a super-troop of 88 individuals. The observations were made from dawn to dusk 7 days every month from June 2009 to July 2010. Approximately 212 hours of observation were made. The observations were focused on adult males and females. Instantaneous focal animal sampling method (Altmann, 1974) was used to record feeding behaviour. If the animal was feeding, the species and plant parts (e.g. young leaf, mature leaf, fruits, seeds, and flowers) were recorded (Fig. 2).

Herbarium specimen collecting and classification

Herbarium specimens were collected and identified. Feeding trees were tagged with a number and located by GPS. The herbarium specimens of the feeding tree were collected within the month that observations were conducted.



Fig.2. Grey-shanked douc langurs (*Pygathrix cinerea*) eating at the study site. Photo: Nguyen Thi Tinh.

Nutritional Analyses

Every month 2 plant samples frequently eaten and 1 plant sample occasionally eaten were collected. In total 34 plant samples were collected (14 young leaves, 8 mature leaves, 9 seeds, 2 fruits, and 1 flower), but one fruit sample was not analyzed. The samples represent 32 plant species of which 12 species were on the top 20 most selected plants. The samples were dried by sunshine. They were stored in airtight plastic bags until analysis. Samples were sent to the biochemistry lab in Hue University. In the laboratory, samples were re-dried in an oven (60°C) to a constant weight to determine total dry matter (DM). Samples were ashed at 550°C from 3 to 6 hours to determine the percentage of inorganic constituents (total ash content). Total nitrogen content was determined using a macro-Kjeldahl procedure with the machine Kjeltex-2200. Crude protein (CP) was determined using the standard formula: $N \times 6.25$ (Williams, 1984).

Neutral Detergent Fiber (NDF) (cellulose+hemicellulose+lignin), Acid Detergent Fiber (ADF) (cellulose + lignin), and Acid Detergent Lignin (ADL) were sequentially analyzed along with the sulfuric acid lignin fraction, using the methods described in Van Soest, (1991).

Sugars were determined from a phenol-sulfuric acid colorimetric assay (Dubois et al., 1956) as modified by Strickland and Parsons (1972) with sucrose used as a standard. Data was reported on 100% of dry matter (DM). The nutrient value of the plant samples was analyzed by several methods (Table 1).

Table. 1. Methods for nutrient analysis.

Parameter	Test method
Crude protein	Kjeldahl test (TCVN 4328-01)
Crude fat	Soxhlet (TCVN 4331-01)
Sugar	Dubois et al. (1956)
Starch	Hovencamp & Hermelink (1988)
NDF, ADF, ADL	Van Soest (TCVN 4329-93)
Minerals	Atomic absorption spectrometry AAS(AOAC-2000)

Statistical Analyses

Nutrient and fiber content of young and mature leaves were compared. The difference was determined using independent sample T-Test. Levene's Test was used to check the equality of variances before T-Tests were conducted. The same procedure was applied to nutrient and fiber contents in leaves and fruits. All tests set the significant value $\alpha=0.05$, 2-tailed.

Results

Plant species eaten by the grey-shanked douc langurs

In this study, 135 plant species of 44 plant families were recorded as foods of the grey-shanked douc langurs. 81 species were observed directly and 54 species identified from food remains. The top 10 most eaten plant families contain 54.8% of all eaten species. The 10 families were Myrtaceae, Sapindaceae, Moraceae, Lauraceae, Euphorbiaceae, Meliaceae, Elaeocarpaceae, Fagaceae, Papilionoideae and Flacourtiaceae. The family Moraceae contained the most selected foods with 14 species, including 12 species for young leaves and two species for fruit. The family Euphorbiaceae contained the second most selected foods with 13 species, including 12 species for young leaves and flowers of one plant species.

In terms of time spent feeding, 10 different families occupied 64.4% of total feeding time. The 10 families were Sapindaceae, Myrtaceae, Lauraceae, Guttiferae, Moraceae, Flacourtiaceae, Burseraceae, Theaceae, Loganiaceae and Betulaceae (Table 2). The family Sapindaceae was eaten most extensively with 28.5% of total feeding time. The family Myrtaceae was second with 7.13% of total feeding time. Five plant families (Myrtaceae, Sapindaceae, Moraceae, Lauraceae, and Flacourtiaceae) were very important food sources for grey-shanked douc langurs. Three families (Moraceae, Theaceae, Loganiaceae) were important sources of leaves while four families (Burseraceae, Sapindaceae, Myrtaceae, and Guttiferae) were important sources of seeds and fruits.

Table 2. The top 10 most eaten plant species.

TT	Species	Family	Young leaves	Mature leaves	Fruit	Seed	Flower	Time spent (%)
1	<i>Pometia pinnata</i>	Sapindaceae	1.18	0.77	.	21.53	0.71	24.19
2	<i>Syzygium tramnion</i>	Myrtaceae	.	1.35	0.20	1.42	.	2.97
3	<i>Garcinia merguensis</i>	Guttiferae	.	.	.	2.86	.	2.86
4	<i>Canarium bengalense</i>	Burseraceae	0.14	.	0.91	1.76	.	2.81
5	<i>Strychnos ovata</i>	Loganiaceae	2.35	2.35
6	<i>Syzygium oblatum</i>	Myrtaceae	.	.	0.69	1.47	.	2.15
7	<i>Nephelium lappaceum</i>	Sapindaceae	.	.	.	2.09	.	2.09
8	<i>Betula alnoides</i>	Betulaceae	1.94	1.94
9	<i>Homalium ceylanicaum</i>	Flacourtiaceae	0.97	0.81	.	.	.	1.78
10	<i>Camellia fleuryi</i>	Theaceae	1.73	0.04	.	.	.	1.77

The species *Pometia pinnata* and *Nephelium lappaceum* were the most eaten foods. The douc langurs spent nearly a quarter of their feeding time (24.1%) on *Pometia pinnata* (Fig. 3). For this plant species, all type of food items were eaten including young leaves, mature leaves, flowers and seeds. The seeds of *Pometia pinnata* were available in peak from June to July and young leaves were available at peak in February. *Syzygium tramnion* and *Syzygium oblatum* were also important for the douc langurs since they feed on young leaves and seeds of the both species through-out the year. The species *Homalium ceylanicaum* was an important source of young leaves and mature leaves from January to April.



Fig.3. Plant species *Pometia pinnata* eaten by the grey-shanked douc langurs (*Pygathrix cinerea*). Photo: Nguyen Thi Tinh.

Among the most important food sources to the douc langurs were 3 families (Sapindaceae, Myrtaceae and Flacourtiaceae) and 5 species (*Nephelium lappaceum*, *Pometia pinnata*, *Syzygium tramnion*, *Syzygium oblatum* and *Homalium ceylanicaum*).

Phytochemical content of food items

Leaves vs. fruits

The fiber content of leaves was greater than of fruits including NDF ($t=4.188$; $P<0.001$), ADF ($t=4.379$; $P<0.001$), and ADL ($t=2.976$; $P=0.006$) (Table 3). Protein in leaves is greater than in fruits ($t=2.257$; $P=0.031$). However, sugars and starches in fruits were greater than in leaves ($t=-2.989$; $P=0.012$) and ($t=-3.184$; $P=0.007$). Lipid content in leaves and fruits was similar.

Table 3. Nutrient and fiber content between leaves and fruits.

Parameter	Leaves (n=22)		Fruits (n=11)		T-Test		
	Mean (g/kg DM)	sd	Mean (g/kg DM)	sd	t	df	P (2-tailed)
Protein	127.68	46.37	83.30	65.40	2.257	31	0.031
Sugar	37.47	16.19	75.52	40.63	-2.989	31	0.012
Starch	83.96	73.23	222.66	134.90	-3.184	31	0.007
Lipids	20.88	12.73	40.13	74.98	-0.845	31	0.417
Ash	58.87	33.00	32.44	16.40	2.493	31	0.018
NDF	466.38	97.33	280.07	158.43	4.188	31	0.001
ADF	324.65	107.14	155.97	98.08	4.379	31	0.001
ADL	163.15	75.66	84.92	60.76	2.976	31	0.006

Young vs. mature leaves

Nutrients (protein, sugar, lipid, and starch) in young leaves were almost identical to mature leaves. Protein in young leaves was generally higher than in mature leaves, but the difference was not statistically significant ($t=1.46$; $P=0.16$). Fiber content (NDF, ADF and ADL) in young leaves and mature leaves was similar (Table 4).

Table 4. Nutrient and fiber contents in young leaves and mature leaves.

Parameter	Young leaves (n=14)		Mature leaves (n=8)		T-Test		
	Mean (g/kg DM)	sd	Mean (g/kg DM)	sd	t	df	P (2-tailed)
Protein	141.27	41.47	115.23	37.93	1.46	20	0.160
Protein/ADF	0.44	0.21	0.42	0.15	0.32	20	0.750
Sugar	35.47	16.11	39.01	9.59	-0.56	20	0.579
Starch	75.37	30.96	65.00	15.83	0.88	20	0.390
Lipids	16.96	8.19	27.16	16.88	-1.92	20	0.069
Ash	66.90	37.03	46.08	17.82	1.48	20	0.153
NDF	487.26	104.98	462.78	92.13	0.55	20	0.589
ADF	350.88	111.36	291.21	82.13	1.32	20	0.202
ADL	185.32	77.63	127.71	58.70	1.82	20	0.084

Nutrient and fiber content in selected plant species

The greatest amount of protein was found in seeds of *Canarium bengalense* with 210.7 g/kg DM. Protein in young leaves of *Strychnos ovata* and *Antidesma velutinsums* was also high with 201.4 g/kg DM. The greatest amount of lipids was found in seeds of *Nephelium lappaceum* with 208.6 g/kg DM. The greatest amount of sugar was found in fruits of *Syzygium oblatum* with 178.1 g/kg DM. The greatest amount of starch was found in seeds of *Lithocarpus ceriferus* with 428.9 g/kg DM. The species *Antidesma velutinasum* contains the greatest amount of NDF (622.2 g/kg DM), ADF (560.1 g/kg DM) and ADL (351.8 g/kg DM) of the young leaves that were eaten (Table 5).

Discussion

Diversity of plant species in diets of grey-shanked douc langurs

Ha Thang Long (2009) reported that the grey-shanked douc langurs fed on 166 species of 40 families in Kon Ka Kinh National Park. In this study, 135 species of 44 families were identified. There are only 25 food species overlapping between this study and the study of Ha Thang Long (2009). Difference in location of feeding observations and trees combined with high diversity of plant species in Kon Ka Kinh National Park may constitute to the low overlap of consumed plant species. In total, the grey-shanked douc langurs ate 251 identified plant species in Kon Ka Kinh National

Table 5. Mean for nutrient and fiber content in all food items.

Nutrient and fiber contents	Mean (g/kg DM) (n= 33)	sd	Min	Max	Top 3 species with the highest nutrient and fibers
Protein	114.99	57.03	25.2	210.7	<i>Canarium bengalense</i> , <i>Strychnos ovata</i> , <i>Antidesma velutinsums</i>
Lipid	26.83	43.55	0.7	208.6	<i>Nephelium lappaceum</i> , <i>Canarium bengalense</i> , <i>Tetradium glabrifolium</i>
Sugar	49.59	31.61	12.5	178.1	<i>Syzygium oblatum</i> , <i>Syzygium zimmermannii</i> , <i>Syzygium tramnion</i>
Starch	128.02	115.60	19.8	428.9	<i>Lithocarpus ceriferus</i> , <i>Syzygium tramnion</i> , <i>Garcinia merguensis</i>
NDF	408.75	148.42	98.3	622.2	<i>Antidesma velutinsum</i> , <i>Betula alnoides</i> , <i>Wightia speciosissima</i>
ADF	272.84	131.17	29.8	560.1	<i>Antidesma velutinsum</i> , <i>Garcinia oligantha</i> , <i>Maesa parvifolia</i>
ADL	141.86	83.07	4.1	351.8	<i>Antidesma velutinsum</i> , <i>Maesa parvifolia</i> , <i>Garcinia oligantha</i>

Park, 109 additional plant species were identified in this study. Among *Pygathrix*, the grey-shanked douc langur shows the highest recorded diversity in plant species consumed. Black-shanked douc langurs consumed 152 plant species of 37 families (Hoang Ming Duc, 2009), and red-shanked douc langurs 87 species from 36 families (Otto, 2005). Among odd-nosed monkeys, the grey-shanked douc langurs also show very high diversity of plant species in their diets. For example, the proboscis monkey (*Nasalis larvatus*) ate 188 plant species (Matsuda et al., 2009) while *Rhinopithecus roxellana* ate 84 plant species (Guo et al., 2007). Food resources in Kon Ka Kinh National Park are likely more diverse and dense than other sites due to greater plant density and diversity.

Nutrition in the diet of grey-shanked douc langurs

In this study, we found that the grey-shanked douc langurs met the adequate 7-11% protein of dry matter required for growth and maintenance as suggested by Oftedal (1991). Among *Pygathrix*, the diets of the red-shanked douc langurs contained the highest amount of protein (173 g/kg dry matter) in young leaves, followed by the amount of protein in the diets of the grey-shanked and black-shanked douc langurs. The study of red-shanked douc langurs was conducted in captivity at Cuc Phuong National Park which is located in limestone forest of northern Vietnam outside of the known range of *Pygathrix* (Otto, 2005), while the studies on grey-shanked and black-shanked douc langurs were conducted in southern Vietnam in the wild. Difference in vegetation and habitat may affect the amount of protein in their diets.

The nutrition contained in eaten plant parts was analyzed in the Delacour's langur (*Trachypithecus delacouri*) also showing that protein in young leaves is higher than in mature leaves (Workman & Le Van Dung, 2009) (Table 6).

Table 6. Comparing nutritional value in the diets of all douc langur species and the Delacour's langur (*Trachypithecus delacouri*).

Species	Study site	Nutrition Value (g/kg dry matter)		Plant parts	Reference
		Protein	NDF		
<i>Pygathrix cinerea</i>	Kon Ka Kinh NP, Gia Lai	141.2	487	Young leaves	This study
		115.2	462	Mature leaves	
		83.3	280	Fruits	
<i>Pygathrix nemaeus</i>	Cuc Phuong NP, Ninh Binh	173	365	Young leaves	Otto (2005)
		128	384	Mature leaves	
<i>Pygathrix nigripes</i>	Bu Gia Map NP, Binh Phuoc Nui Chua NP, Ninh Thuan	123	466	Mixed leaves	Hoang Minh Duc (2007)
		75.7	442	Fruits	
<i>Trachypithecus delacouri</i>	Van Long Nature Reserve, Ninh Binh	122	380	Young leaves	Workman & Le Van Dung (2009)
		104	334	Mature leaves	

Important food sources for the grey-shanked douc langurs in Kon Ka Kinh National Park

Dietary studies that identify the important food in a primate diet are important for conservation. In this study, we determined 3 species which are the most important foods including *Pometia pinnata*, *Syzygium trannion*, and *Canarium bengalense* (Fig. 4). Among those, *Pometia pinnata* was the most vital since up to 24.9% of time spent feeding was on this species, which corroborates an earlier study that grey-shanked douc langurs spent the greatest amount of time (14,3%) feeding on *Pometia pinnata* (Ha Thanh Long, 2009). Grey-shanked douc langurs ate several plant parts of these species including

young leaves, mature leaves, fruits, seeds and flowers. During the peak of fruiting (June, July, and August) the grey-shanked douc langurs spent significant time harvesting fruits and seeds. They also intensively ate young leaves of this plant during 4 months of the dry season (January to April). Following

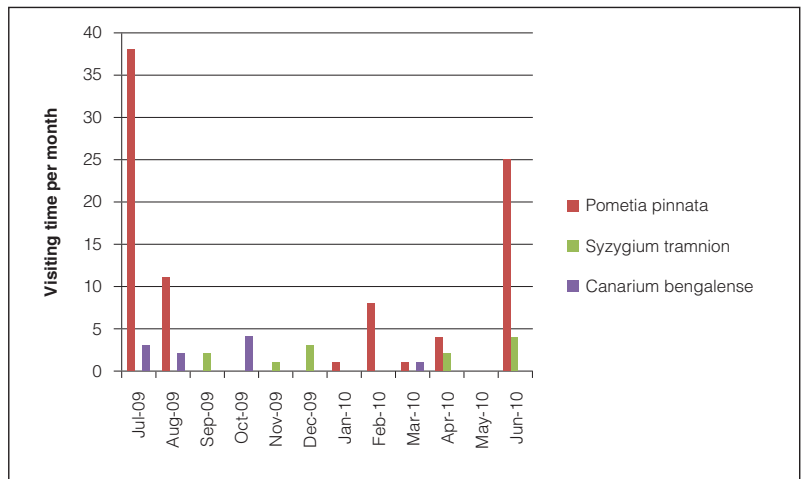


Fig.4. The three plant species most visited by the grey-shanked douc langurs (*Pygathrix cinerea*).

Marshall & Wrangham's (2007) definition of 'preferred food' there is sufficient evidence to conclude that *Pometia pinnata* is the 'preferred food' of grey-shanked douc langurs, because the density of the plant species in the study site is only 1.4 trees/ha, so the availability is low. Further, the usage of the plant species by the grey-shanked douc langurs is very high. *Pometia pinnata* is an important agro forest tree in the Pacific Island (Thomson & Thaman, 2006), where it has commercial value and ecological value in Samoa. In Kon Ka Kinh National Park, however, local people did not harvest this plant species for any purpose, and does not seem to be in high local demand currently.

Conclusions

The diet of grey-shanked douc langurs is highly diverse in terms of plant species. There were 135 species of 44 plant families eaten by the douc langurs in this study. Combined with previous studies, there are 251 plant species recorded as foods for the grey-shanked douc langurs in the Kon Ka Kinh National Park. The nutritional component in selected food consists of protein at 11.4% of dry matter, lipids at 2.6%, minerals at 5.0%, sugars at 4.9%, starches at 12.8%, and NDF at 40.8%. The consumption of protein is similar to that of red-shanked and black-shanked douc langurs. At the study site, the plant species *Pometia pinnata* is the preferred food.

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