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DEVELOPMENT OF EXPLORATIVE BEHAVIOR AND NEOPHOBIA IN CAPTIVE SUN BEARS (*HELARCTOS MALAYANUS*)

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Keywords Ursidae Exploration novel object test bear cubs captivity rehabilitation Neophobia is the aversion to novelty and is a widespread phenomenon in the animal kingdom. In bears (Ursidae) neophobic responses seem to develop around the age of 5 months, and disturbance of this development may be the cause of rehabilitation failures. However, little is known about the behavioral development of bear cubs, which may be helpful for successful rehabilitation as well as for zoo animal management. Here, the development of explorative behavior and neophobia is investigated in two captive sun bear cubs (*Helarctos malayanus*). The behavior of the animals is observed between the ages of 4 to 6 months and neophobic responses are tested during general observations and in novel object tests. It would be expected that explorative behavior decreases while neophobia increases and that this development is in parallel with a growth in (social) distance to their mother. The results show that there is no decrease in explorative behavior of the cubs and no evidence for the development of neophobia is found. However, the distance to the mother does increase between the ages of 4 to 6 months, indicating that this would be an evident timeframe for neophobia to develop. These findings suggest that either no development of neophobia occurs in the sun bear, or the captive environment has disturbed their natural development. Further research comparing captive and wild cubs should clarify this.

ABSTRACT

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INTRODUCTION

Neophobia, or the aversion to novelty, is a widespread phenomenon in the animal kingdom (Greenberg, 2003). The development of neophobia is often demonstrated by a change from extensive explorative behavior during the juvenile phase to a fear of novelties when growing towards adulthood. It is specifically common in birds and mammals (Corey, 1978). For example, in neotropical raptors, chacma baboons, geladas, spotted hyenas and bats, juveniles have shown more exploration and less neophobia compared to adults (Bergman & Kitchen, 2009; Benson-Amram & Holekamp, 2012; Biondi, Bó, & Vassallo, 2010; Carter, Forss, Page, & Ratcliffe, 2018). The development of neophobia in animals can be explained by the uncertainty of costs and benefits that are associated with exploration of novelties (Greenberg & Mettke-Hofmann, 2001; Greenberg, 2003). During the juvenile phase, an individual has little information about its environment and gains many benefits from exploring. At the same time, the juvenile has little costs since it is still under maternal protection. As the individual grows older and familiarizes with its environment, the benefits of exploring decrease and the costs of potential danger increase due to the decline of maternal protection.

In bears (Ursidae), there have been signs of the development of neophobia in brown bear cubs (Pazhetnov & Pazhetnov, 2005), but no quantitative study on this has been reported so far. Mazur & Seher (2008) studied the learning of foraging behavior in wild black bears (Ursus americanus) and suggest neophobic tendencies in a hazardous human environment. Dahl et al. (2020) try to change stereotypical behavior in captivity by adding enrichments to polar bears (Ursus maritimus) and mentioned significant individual differences in neophobia and exploratory behavior. However, knowledge on this development may be of great value in rehabilitation programs. In total, there are eight bear species and most of them are according to the IUCN listed as vulnerable (Scotson et al., 2017). Conservation and rehabilitation programs are aiding in the recovery and stabilization of populations, but survival of released bears is not always accomplished (Fredriksson, 2001). This is often thought to be caused by the lack of fear for local humans, conflicts with resident bears, or underdeveloped foraging skills. It has been shown that successful rehabilitation depends on knowledge of cub behavior with as most important behavioral components their feeding, defensive, and social behaviors (Skripova, 2013). Sun bears are largely solitary animals, except during the mother-cub rearing phase (Scotson et al., 2017). The female gives birth to usually one or two highly altricial cubs and the denning period has previously been observed to be around 3 months (Nowak & Walker, 1999; Hall & Swaisgood, 2009). Cubs usually stay with their mother for up to three years in which they learn skills and gain information about foraging and predators via social learning (Gilbert, 1999). The first signs of explorative behavior of a sun bear cub have been observed around the age of 2 months (Hall & Swaisgood, 2009). This study by Hall and Swaisgood (2009) is the first quantitative study on the early development of a sun bear cub in a zoo environment and they have described the first three months of development. After a short phase of extensive exploration, sun bear cubs are thought to show a sudden decrease in exploratory behavior and develop neophobia as they grow older, as has been observed in one of their relatives, the brown bear (Ursus arctos), by Pazhetnov & Pazhetnov (2005). Fear reactions of these brown bear cubs were described by cessation of movement, followed by squatting, and climbing into a tree. In Asian black bears, similar behaviors have been attributed to the development of fear and include hiding in trees, escaping,

and attacking (Skripova, 2013).

There are different factors that may interplay in the development of neophobia in bears. Firstly, the emergence and development of fear reactions could be a contributing factor. In brown bears and Asiatic black bears, the emergence of fear and avoidance behavior has been observed at the age of 4.5 to 5 months (Pazhetnov & Pazhetnov, 2005; Skripova, 2013). Secondly, the development of adult foraging skills may reduce the need to further explore and may favor the development of neophobia. It has previously been shown that Asiatic black bears develop foraging patterns in parallel with fear behaviors between the ages of 2 to 5 months (Skripova, 2013) and that in brown bears basic foraging behaviors have developed at the age of 5 months (Pazhetnov & Pazhetnov, 2005). This also indicates the development of nutritional independence and therefore a dissociation from their mother. This dissociation involving an increase in (social) distance between mother and cub could be a third role playing factor in neophobia. It decreases maternal protection and increases the costs of exploration. However, even though they start to develop foraging skills and start dissociating, the cubs do not become completely independent at this age. The bear cubs still need to further develop their (foraging) skills which includes social learning from their mother (Gilbert, 1999). This could be for example in terms of learning and remembering locations of food sources or practicing skills, which is the case in polar bears that also have a prolonged period of dependence (Gilbert, 1999). Based on these three factors, it can be suggested that around the age of 4.5 to 5 months neophobia could develop in bears. This would be different from for example bird species in which the switch to neophobia occurs relatively later, namely around the time of independence or adulthood (Greenberg & Mettke-Hofmann, 2001).

So far, no quantitative research has been conducted on the development of neophobia in bears. Neophobia is often experimentally tested by so-called novel object tests (NOTs), in which animals are presented with novelties. Neophobic responses are measured by avoidance behavior, overall activity, and social grouping, amongst others (Crane & Ferrari, 2017). There have been studies using novel objects on bears, but only in the context of personality (Myers & Young, 2018; Martin-Wintle *et al.*, 2017) or in the context of zoo enrichment (e.g. Swaisgood *et al.*, 2001; Wagman *et al.*, 2018; Renner & Lussier, 2002). These studies all took place in captivity and NOTs have not been done on wild bears so far. Studying and testing bear cubs in the wild is difficult and therefore a zoo environment can offer more stability for observations and conduction of tests.

The goal of this study is to investigate the development of explorative behavior and neophobia in two captive sun bear cubs. The following parts of their behavioral development will be followed: the differences and development of explorative behavior, the development of neophobia, and the development of social distance between the cubs and their mother. The cubs will be studied between the ages of 4 to 6 months. They are tested for neophobia by different novel objects tests and are observed for their general behavior after and between the tests using focal-animal sampling. It is expected that the cubs show more explorative behavior than their mother and that the amount of exploration of the cubs declines over time. Furthermore, it is expected that the latency to approach novel stimuli and the amount of fear increases as the cubs grow older and that this is different for each stimulus category. The (social) distance between the cubs and their mother is expected to increase, indicating that the age of 4 to 6 months is a reasonable time frame for neophobia to develop. With this research a better understanding of sun bear cub development may be gained which could function as a tool for wildlife conservation and rehabilitation as well as for successful management of cubs in captivity.

METHODS

Animals and Housing

The subjects of this study were captive sun bears that are part of Koninklijke Burgers' Zoo, Arnhem (The Netherlands). This study was focused on one female with two cubs that are from now on indicated with A, B, and C (A and B for the cubs; C for the female). During the study, there were some interactions and influences from a male bear (their father) that was introduced to the cubs in the 5th week of the study by giving him entrance to their enclosure in the mornings. After some aggressive reactions from the female, the zoo decided not to continue with the introductions in the beginning of week 6.

The female with the two cubs was born on 4 August 2000 and grew up in Cologne Zoo, Köln (Germany) after which she was moved to Arnhem in 2008. Her two male cubs were born on 7 May 2019 and had stayed in the maternal den (a separate enclosure containing nesting materials) until the age of 2.5 months, after which they got access to the display enclosure. The cubs were 4 months old at the start of the study in the second week of September 2019. The female was kept in several night enclosures together with her two cubs and access to the display and outside enclosure was regulated by zookeepers and kept to a minimum. The animals were fed at irregular times during the day and their diet consisted of a variety of vegetables, fruits, dairy, fish, and meat.

General Observations

The general behavior of the mother and cubs was observed for a period of 7 weeks (from 16 September until 1 November 2019). The observations took place from Monday to Friday and generally lasted 4 to 5 hours a day (between 9.00 and 15.00 hour). Depending on the weather, the animals were observed either in their display or outside enclosure. The area the bears could move freely was about 600 m². The animals were observed by focalanimal sampling and the behaviors were scored by using one-zero coding with an interval of 1 minute (Martin & Bateson, 1993). A timer was set to produce an audible sound at the end of each 1-minute interval. The ethogram of the behaviors that were measured consists of 42 behaviors that are divided into locomotor, explorative, solitary, social, and anticipatory behaviors (Table 1). Besides these behaviors, the distances from the cubs to their mother, from the cubs to the ground, and between the cubs were estimated at the end of each 1-minute interval. Baseline observations were made prior to the test phase (from 10 September to 13 September 2019) and lasted 16 hours. In total, 167.5 hours of general behavioral observations were collected. The animals were observed live by one observer who recorded the behaviors manually. The observer was positioned in the visitor's area and capable to move around.

Novel Object Tests

The cubs were tested specifically for neophobia during novel object tests. These tests were performed in the same 7 weeks, mostly on Monday, Wednesday, and Friday between 9.00 and 10.00 hour. The novelties were divided into objects, olfactory stimuli, and auditory stimuli (Table 2). The objects and stimuli were chosen in such a way, that they do not occur in the natural habitat or in the current and past environment of the animals. This was to ensure that the reactions were not related to innate responses or to recognition of the stimulus. Besides, the object or stimulus was not edible or related to food, so that the response of the animals was not directly influenced by the motivation to consume or search for food.

Behavior	Abbr.	Definition		
Locomotor behavior				
Solitary resting	AR	The individual lays down or sits on the ground or on an enclosure enrichment with musculature relaxed and is not moving or showing another behavior. The eyes can be open or closed.		
Social resting	SR	The individual lays down or sits on the ground or on an enclosure enrichment with musculature relaxed and is not moving or showing another behavior. The individual is close (within the mother's arm length) to a conspecific. The eyes can be open or closed.		
Bipedal standing	BS	The individual stands vertical on its two hindlimbs without holding on to leaning against an object.		
Walking/running on ground	WR	The individual moves over the ground using four limbs.		
Climbing	CL	The individual moves over enclosure enrichments or trees/wood above ground using its limbs.		
Going inside/outside	IO	The individual moves from outside to inside (the display enclosure) or vice versa.		
Exploratory behavior				
Investigating food	IF	The individual licks, bites, sniffs, touches, or manipulates food with its claws.		
Investigating object with claws	IC	The individual touches, turns over, lifts, manipulates and/or holds an object with its claws.		
Investigating target object	IT	The individual licks, bites, sniffs, touches, or manipulates the target object/stimulus that is placed during the novel object test.		
Sniffing object	SO	The individual smells an object with its nose from a short distance or touches the object with its nose to smell.		
Biting object	BO	The individual uses its teeth to bite an object.		
Licking object	LO	The individual licks an object with its tongue.		
Sniffing air	SA	The individual sniffs the air with its nose while having its head horizontal or lifted.		
Sniffing ground	SG	The individual sniffs the ground underneath while bending its head downwards.		
Moving ground	MG	The individual moves ground or pieces on the ground with its front claws or nose.		
Solitary behavior				
Eating	EA	The individual chews and subsequently swallows food.		
Drinking	DR	The individual ingests water.		
Tongue flicking	TF	The individual's tongue comes out of its mouth and moves around its lips or in the air.		
Body shake	SH	The individual shakes its body sideways quickly for a short duration.		
Scratching	SC	The individual uses its claw to scratch another body part.		
Yawning	YA	The individual has its mouth wide open to yawn.		
Selflicking	SL	The individual licks its own fur or body with its tongue.		
Self biting	SB	The individual bites its own fur or body with its teeth.		
Urinating	UR	The individual urinates.		
Defecating	DE	The individual defecates.		
Stationary alert	ST	The individual suddenly stops to look at something before it continues its initial behavior (e.g. walking).		

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Table 1. Ethogram	of the scored	behaviors	during the	general	observations
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Solitary play movement	SP	The individual plays by making rolls, jumps or other vigorous movements with or without holding an object. The individual is not in contact with a conspecific.
Reaction to visitors	RP	The individual responds positively to the presence of or stimuli from zoo
(positive/alert)		visitors by approaching or looking into their direction.
Reaction to visitors	RN	The individual responds negatively to the presence of or stimuli from zoo
(negative/scared)		visitors by avoiding contact or walking away.
Smearing face	SF	The individual grabs a piece of food, places it on the lower arm or paw and
		subsequently tries to smear the piece of food on its own forehead or cheek.
		This sequence of behaviors is usually repeated several times.
Social behavior		
Suckling	SU	The individual (cub) suckles from its mother's nipple.
Body contact	BC	The individual has any form of body contact with a conspecific.
Following conspecific	FC	The individual is following another individual by walking behind it in proximity.
Being followed by	BF	The individual is being followed by another individual that is walking behind
conspecific		it in proximity.
Social play	SP	The individual bites, slaps, pushes, and/or pulls another individual gently
		(sometimes mouth is opened and/or ears are lifted towards the back). This is
		often seen together with one or both individuals rolling over. Mother's play
		behavior often only involves an open mouth and/or biting.
Vocalizing	VO	The individual produces a sound with its mouth or throat.
Licking conspecific	LC	The individual licks the fur of another conspecific with its tongue.
Anticipatory behavior		
Sniffing enclosure entrance	SE	The individual smells one of the entrances in the enclosure from a short
		distance with its nose or touches the entrance with its nose. Often happens
		after the individual has noticed a caretaker.
Pacing	PA	The individual walks back and forth in the same path at least twice, often after
		noticing a caretaker.
Awaiting	AW	The individual does not move or show any other behavior while having its
		attention clearly focused at an individual or situation around.
		cubs at the end of every interval)
Distance from ground	DG	The height (in meters) at which the individual is located from the ground is
		estimated and rounded up to whole numbers.
Distance from mother	DM	The distance (in meters) from each of the cubs to their mother is estimated
	5.6	and is rounded up to whole numbers.
Distance between cubs	DC	The distance (in meters) between the two cubs is estimated and rounded up
		to whole numbers.

It was aimed to choose the objects in such a way, that they were about similar in potential scariness (e.g. in terms of size, complexity, and color intensity), but varying in color, shape, and material. Little is known about sun bears' vision, but there is a possibility that they can discriminate between colors, as one of their closest relatives, the American black bear, has shown indications for hue discrimination (Bacon & Burghardt, 1976). The olfactory stimuli were aimed to be experienced as neutral; being neither directly repulsive nor attractive. For the auditory stimuli, a subdivision was made between animal vocalizations and non-vocalizations. Each auditory stimulus had a duration of 7.5 s and was repeated with intervals of 22.5 s of silence during the entire trial of one hour (or longer in case one of the individuals had not entered immediately and the trial was continued until all had been in the enclosure for at least 30 min).

Week	Date	Trial	Object/stimulus	Category
38	Tuesday 17/9	1	Traffic cone	Object
	Wednesday 18/9	2	Lavender	Olfactory stimulus
	Friday 20/9	3	Collar bell	Auditory stimulus (non-vocalization)
39	Monday 23/9	4	Skateboard	Object
	Wednesday 25/9	5	Vinegar	Olfactory stimulus
	Friday 27/9	6	Chicken vocalization	Auditory stimulus (vocalization)
40	Monday 30/9	7	Watering can	Object
	Wednesday 2/10	8	Anise	Olfactory stimulus
	Friday 4/10	9	Trumpet	Auditory stimulus (non-vocalization)
41	Monday 7/10	10	Rubber duck	Object
	Wednesday 9/10	11	Peppermint	Olfactory stimulus
	Friday 11/10	12	Sheep vocalization	Auditory stimulus (vocalization)
42	Monday 14/10	13	Tin plate	Object
	Wednesday 16/10	14	Rosemary	Olfactory stimulus
	Friday 18/10	15	Horse vocalization	Auditory stimulus (vocalization)
43	Monday 21/10	16	Plastic dice	Object
	Wednesday 23/10	17	Perfume	Olfactory stimulus
	Friday 25/10	18	Cartoon boink	Auditory stimulus (non-vocalization)
44	Monday 28/10	19	Fire hose cube	Object
	Wednesday 30/10	20	Vanilla	Olfactory stimulus
45	Monday 4/11	21	Seal vocalization	Auditory stimulus (vocalization)

Table 2. Objects and stimuli that were used during the novel object tests.

The object or stimulus was placed in the (12.15 by 6.00 meter) display enclosure after which the mother and cubs got access to the enclosure. The animals could not be separated and were therefore tested all at the same time. The objects were placed in the center of the enclosure at the exact same position and were immediately visible upon entering the enclosure. The olfactory stimuli were smeared in liquid form on one of the enrichment branches as close as possible to the center of the enclosure. The auditory stimuli were played by a speaker that was placed in the corridor right behind the zookeepers' entrance and started playing when all individuals were present in the display. Food was present during most of the novel object tests in the form of vegetables and/or fruits, as to be in accordance with the feeding regulations of the zoo.

The behavior of the cubs and their mother during the novel object tests was monitored for one hour after the animals entered the display enclosure. The behaviors were measured by one observer and were videotaped with a GZ-MG630 camcorder (JVZ) from the visitor's area. In case not all animals entered the display enclosure at the start of the trial, video recording was continued until all animals had been present in the enclosure for at least half an hour. Measurements that were made during these tests include latency to approach, distance from the stimulus/ground/mother, number of visits, total visit time, amount of stress behaviors, and amount of fear behaviors (as defined in Table 3.

In addition to the fear behaviors mentioned in the table, the other fear reactions that exist in bears were not possible (e.g. hiding) or did not occur (e.g. attacking). In case an animal did not approach the stimulus during a given trial, data on latency were not taken into account. For auditory stimuli, visits were counted when the animal approached the part of the fence behind which the speaker was placed.

Behavior	Definition		
Latency to approach	The total time (in seconds) it takes for the individual to approach and		
	investigate the object or stimulus after the individual has entered the area.		
Distance from	The distance (in meters) between the individual and the stimulus, ground, and		
stimulus/ground/mother	mother estimated at the end of each 1-minute interval and averaged for one		
	trial.		
Number of visits	The amount of times that the individual visits the object or stimulus (separated		
	by >5 seconds) during one trial.		
Total visit time	The total amount of time (in seconds) that the individual has visited the object		
	or stimulus during one trial.		
Amount of stress behaviors	The total amount of potential stress behaviors that an individual shows during		
	one trial. These behaviors include body shaking, yawning, scratching, self biting,		
	self licking, and tongue flicking.		
Amount of fear behaviors	The total amount of potential fear behaviors that an individual shows during		
	one trial. These behaviors include returning to mother, climbing (counted each		
	time the individual starts climbing), and preparing to climb (counted each time		
	the individual grabs a tree without leaving ground level).		

Table 3 Measurements during the novel object tests.

Data Analyses

The data from the general observations and from the novel object tests were processed in R3.6.1 (R Core Team, 2019). Normality of data was tested with Shapiro-Wilk tests and equality of variance with Levene's tests using the "car" package (Fox & Weisberg, 2019). Based on the outcome of these tests, differences between individuals and between responses to stimulus categories were analyzed by either a two-sampled t-test/ANOVA or by a Kruskal-Wallis (KW)/Mann-Whitney U (MWU) test in case of non-normality of data. In some cases, a Median test (MT) was applied using the "agricolae" package (de Mendiburu, 2020).

The changes in responses and behaviors over time were determined by linear models (LMs) in which homogeneity and heteroscedasticity were checked graphically. Autocorrelation was checked graphically and tested for significance by using Breusch Godfrey tests in the "bgtest" package (Zeileis & Hothorn, 2002). In the linear model for exploration, all explorative behaviors that were not related to foraging (i.e. investigating with claws, investigating target object, sniffing object, biting object, and licking object) were combined, and for the model for foraging all foraging behaviors (i.e. investigating food, sniffing air, sniffing ground, and moving ground) were combined. In one case (the latency across trials) a generalized linear model (GLM) was used with including latency as dependent variable and trial as independent variable with a Gamma (log link) distribution. All tests were based on a significance level of α =0.05. However, Holm correction was used in case of multiple testing and time correction (multiplying the variables according to the amount of time the animal was present) was applied in case the animals were not present in the enclosure during the entire NOT.

RESULTS AND DISCUSSION

Explorative and Foraging Behavior

A significant difference in explorative behavior is found between the individuals (MT: $\chi^2 = 55.55$, df = 2, P < 0.001), and post-hoc analysis using pairwise median tests with Holm-adjusted p-values shows a significant difference between each cub and its mother (post-hoc A-B: $\chi^2 = 0.22$, df = 1, P = 0.64; A-C: $\chi^2 = 40.33$, df = 1, P < 0.001; B-C: $\chi^2 =$ 40.33, df = 1, P < 0.001). For foraging behavior, post-hoc analyses with Holm-adjusted p-values following a Kruskal-Wallis test (KW: $\chi^2 = 43.39$, df = 2, P < 0.001) show that the mother spends more time on this behavior than cub A (MWU: W = 155, P < 0.001), and cub B (MWU: W = 180.5, P < 0.001), while no difference was found between the cubs (MWU: W = 653, P = 0.74).

Linear regression was used to determine the changes in explorative and foraging behavior over time for all individuals separately (Figure 1). No significant increase or decrease has been found in the number of explorative behaviors between the ages of 4 to 6 months shown by individual A ($F_{1,35}$ = 3.14, P = 0.09), individual B ($F_{1,35}$ = 0.44, P = 0.51), and individual C ($F_{1,36}$ = 2.09, P = 0.16). Regarding the foraging behavior of the animals, linear regression

shows no significant change for individual A ($F_{1,35}$ = 1.78, P = 0.19), but a significant increase for individual B ($F_{1,35}$ = 6.70, P = 0.01) and individual C ($F_{1,36}$ = 9.25, P = 0.004).

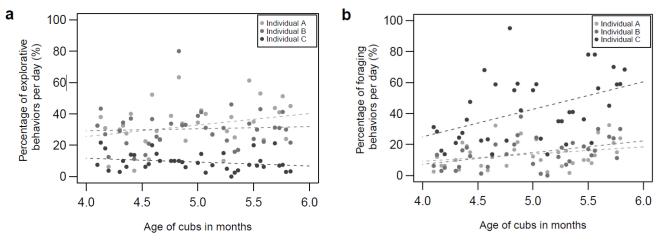


Figure 1. Changes in explorative and foraging behaviors of the three sun bears during the time the cubs were aged between 4 to 6 months. The values are plotted as percentages (the precentages of 1-minute intervals in which explorative/foraging behavior occurred each day). a = Linear regressions show no significant change in the number of explorative behaviors. b = Linear regressions show an increase in foraging behavior for all individuals, however, this is only significant for individual B (P = 0.01) and individual C (P = 0.004)

Neophobia: Change Over Time

GLMs were used to visualize the changes in latency to approach novelties across the different trials of the novel object test (Figure 2). A significant decrease is found in latency to approach novel stimuli over time for individual A ($F_{1,12} = 16.032$, P = 0.002), while only a decreasing trend is found for individual B ($F_{1,13} = 3.17$, P = 0.10). This is in contrast with the hypothesis stating that an increase in latency was expected when neophobia would develop in the cubs. For the mother, individual C, no change in latency is found across the trials ($F_{1,16} = 0.36$, P = 0.56).

In addition to the latency, the distance from the stimulus, ground, and mother were measured as well to determine the degree of fear for the novel stimulus (Figure 3). Both cubs do not differ from each other in the distance from the stimulus (t-test: $t_{39} = -0.07$, P = 0.95), distance from the ground (t-test: $t_{39} = 0.35$, P = 0.73), and distance from their mother (t-test: $t_{39} = 0.64$, P = 0.53). Therefore, data from both cubs were averaged in the linear regression analysis.

No significant change was found in the distance from the stimulus ($F_{1,19} = 0.59$, P = 0.45) or in the distance from the mother ($F_{1,19} = 0.31$, P = 0.58) across the trials. However, there is an increasing trend found in the distance from the ground ($F_{1,19}$ = 3.52, P = 0.08) of both cubs during the trials. Besides the latencies and distances of the cubs, the amount of fear and stress behaviours during the trials could be another indicator for fear and neophobia. There is no difference in the amount of fear and stress behaviours between both cubs (MWU: W = 154.5, P =0.10; MWU: W = 272, P = 0.20, for fear and stress respectively). Linear regression was performed for A and B separately regarding their fear and stress behaviours during the novel object tests over time (Figure 4). The results show that there is no significant increase in the amount of fear behaviours for individual A ($F_{1,18} = 0.85$, P = 0.37) and individual B ($F_{1,18}$ = 0.14, P = 0.71) and no increase in stress behaviours for individual A ($F_{1,18} = 0.99$, P = 0.33) and B ($F_{1,18} = 0.41$, P = 0.53).

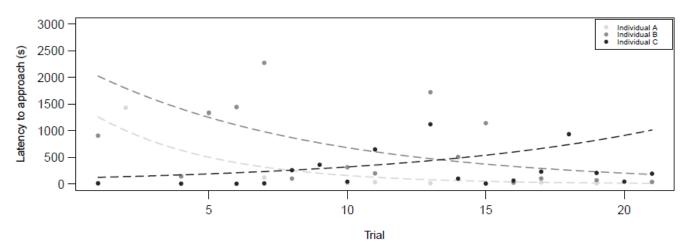
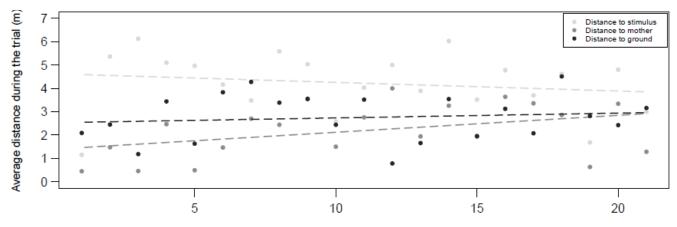


Figure 2. The latency (s) of the subjects to approach novel stimuli across 21 novel object test trials. GLMs show only a significant decrease in latency for individual A (P = 0.002).



Trial

Figure 3. The average distance (m) to the stimulus, to the mother, and from the ground for both cubs during the novel object tests. The values for each distance are plotted as the average of all distances measured at the end of each 1-minute interval of both cubs for each trial. Linear regression shows an increasing in distance to the ground (P = 0.08).

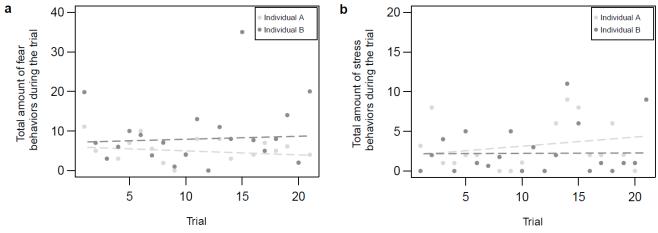


Figure 4. The amount of fear and stress behaviors of both cubs during the novel object tests. The values are plotted as the average of the total amount of fear/stress behaviors of cub A and B separately during one trial. a = Linear regression shows no change in the amount of fear behaviors across the trials, b = neither a change is shown for the amount of stress behaviors.

Neophobia: Differences in responses to stimuli

Different behavior measurements of the novel object tests were used to examine whether a certain stimulus category (objects, olfactory stimuli, or auditory stimuli) elicits more fear or neophobia in the cubs than the others (Figure 5). It was found that the latencies to approach do not differ between objects, olfactory stimuli, and auditory stimuli ($\chi^2(2) = 1.35$, P = 0.51). The amount of stress and fear behaviors of the cubs does not differ between the stimuli ($\chi^2(2) = 0.07$, P = 0.96).

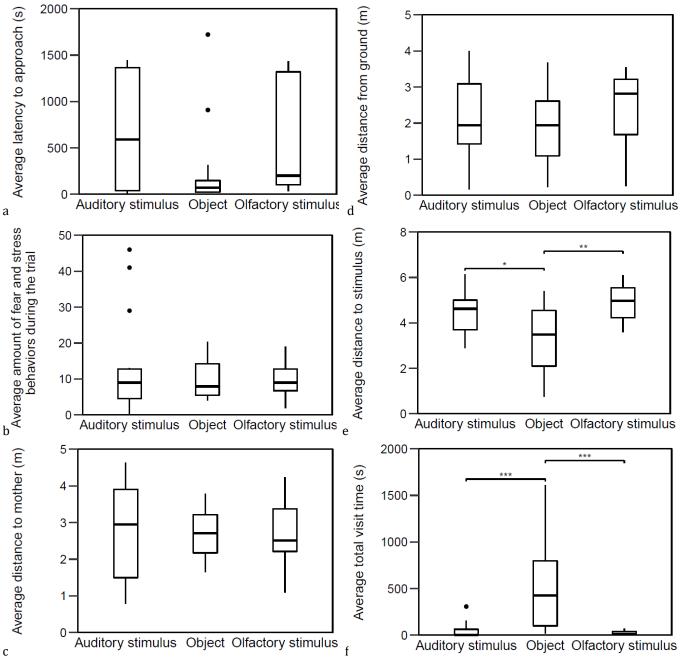


Figure 5. Differences in responses to the different kinds of stimuli. The values are plotted as the averages per trial. a = There is no significant difference in latencies between the stimuli, b = neither in the amount of stress and fear behaviors, c = in the distance from the mother, d = or in the distance from the ground. e = The distance from the stimulus, however, is significantly smaller for objects than for auditory (P < 0.05) and olfactory stimuli (P < 0.01). f The total visit time is significantly larger for objects than for auditory (P < 0.001) and olfactory stimuli (P < 0.001).

Furthermore, the distance from the mother and the distance from the ground do not differ per stimulus ($F_{2,25} = 0.07$, P = 0.94; $\chi^2(2) = 1.35$, P = 0.51, respectively). However, a difference was found for the distances from the stimulus and post-hoc analysis revealed that the distance from the stimulus was significantly smaller for objects than for olfactory stimuli and auditory stimuli ($t_{20} = -3.77$, P = 0.001; $t_{23} = -2.65$, P = 0.01, for olfactory and auditory stimuli respectively). In addition, analysis on the total visit time showed that the time animals spent visiting objects was significantly larger than for odors (W = 184, P < 0.001) and sounds (W = 18, P < 0.001). This suggests less fear and/or a

larger interest for the category of objects.

The category of auditory stimuli was further subdivided into animal vocalizations and non-vocalizations. To determine the difference in response towards animal vocalizations and non-vocalizations, analysis was performed on data of all individuals together (Figure 6). The amount of fear and stress behaviors is higher during trials that present sounds in the form of animal vocalizations compared to non-vocalizations, but this is not significant (W = 35.5, P = 0.20). However, the total visit time is significantly higher for animal vocalizations than for non-vocalizations (W = 19, P = 0.01).

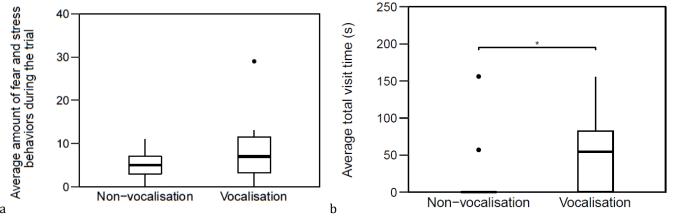


Figure 6. Differences in responses to vocalizations and non-vocalizations. The values are plotted as averages per trial. a = More stress and fear behaviors are shown during trials that present animal vocalizations compared to non-vocalizations, but this is not significant. b = The animals spend more time visiting animal vocalizations than non-vocalizations (P < 0.05).

Other Indications of the Development of Neophobia

Besides the novel object tests, there are several parts in the general observations that could be used to look at the change in fear and thereby the development of neophobia over time. Firstly, the amount of stress behaviors in general (thus outside of the novel object tests) was compared between the individuals and was compared over time (Figure 7a). There was a significant difference found between the average amount of stress behaviors shown per day by A, B, and C (KW: $\chi^2(2) = 38.43$, P < 0.001). Post-hoc analysis using pair-wise test with Holm-Bonferroni adjusted p-values showed that A shows more stress behaviors than B (MWU: W = 913.5, P = 0.01) and B shows more stress behaviors than C (MWU: W = 1065.5, P < 0.001), regardless of the time point. When looking at the change in stress behavior over time using linear regression, none of the animals showed a significant increase or decrease (A: *F*_{1,35} = 0.68, *P* = 0.42; B: *F*_{1,35} = 1.13, P = 0.30; and C: $F_{1,36} = 0.50$, P = 0.48). Another indicator of neophobia could be the distance from the ground, which may be related to the amount of flight behavior. Since the enrichment trees in the outside enclosure are significantly higher than in the inside enclosure, the data for outside and inside observations were separated (Figure 7bc). There was no difference in distance from ground while be inside or outside between individual A and B (t-test: t_{52} = 0.05, P = 0.96; MWU: W = 154, P = 0.32, for inside and outside respectively). Linear regression has shown that there is no change in the distance from the ground when being inside for individual A ($F_{1,24} = 2.86$, P = 0.10) and for individual B ($F_{1,26} = 0.56$, P = 0.46). The distance from the ground when being outside has not changed either, for both individual A ($F_{1,17}$ = 2.85, P = 0.11) and B ($F_{1,18}$ = 1.63, P = 0.22). Thus, both the amount of stress behaviors and the distance from the ground during the general observations do not indicate that there is some development of fear and neophobia within this time frame of the cubs being 4 to 6 months of age.

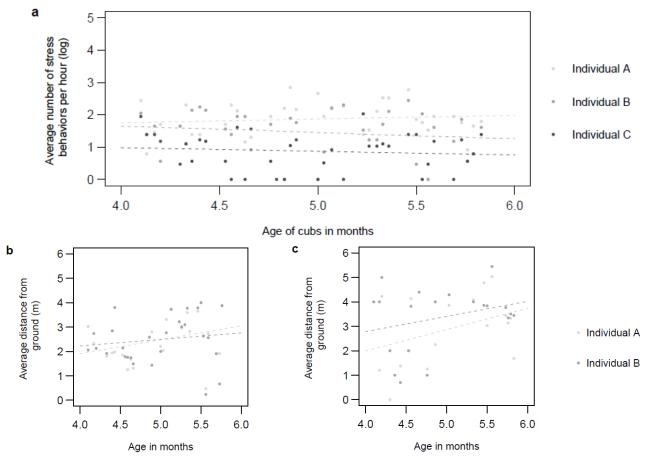


Figure 7. Indicators of the development of stress and fear. a = The change in the average amount of stress behaviors that individual A, B, and C show in 1 hour is shown. The values are plotted as the averages of the total amount of stress behaviors per 1-minute interval multiplied by sixty. Linear regression shows no change in the average amount of stress behaviors over time for individual A, B, and C. b = The change in average distance from the ground (m) per day measured for individual A and B in their inside enclosure is shown. The values are plotted as averages per day of the distances measured at the end of each 1-minute interval. Linear regression shows that there is no change in the distance from the ground when being inside for individual A and B. c = There was no change in distance from the ground when being outside either.

Contributing Factor: distance from mother

Among the hypothesized factors that may contribute to the development of neophobia in sun bear cubs are the development of fear reactions, the development of adult foraging skills, and the increase in distance from their mother. The distance between the mother and the cubs was estimated at the end of each 1-minute interval during the general observations and the average of both cubs together was calculated per day (Figure 8a). Only the observations from the outside enclosure are included, since little variation in distance is possible in the 12-m wide inside enclosure. The distance between the cubs and their mother appears to increase significantly between the ages of 4 to 6 months ($F_{1,19} = 5.76$, P = 0.03).

In addition, the development of social distance between

the cubs and their mother was determined by looking at the change in the amount of suckling behavior, body contact with mother, following behavior, and social play with mother (Figure 8b-e). LMs were used to assess whether the changes in these behaviors were significant. The results show that there is a significant decrease in the amount of suckling behavior of the cubs ($F_{1,35} = 9.82$, P =0.003). This behavior seems to be even fading around the age of 5.5 months, indicating the development of nutritional independence of the cubs. It has also been shown that there is a significant decrease in the amount of body contact between the cubs and their mother ($F_{1,35} =$ 7.62, P = 0.009) and a significant decrease in the amount of social play between the cubs and their mother ($F_{1,35} =$ 4.61, P = 0.04). Interestingly, the amount of times that the cubs follow their mother has increased over time ($F_{1,35}$ = 9.48, P = 0.004). This increase seems to start around the age of 5.3 months, which is the time point at which the male (individual D) was being introduced to the cubs

several times. Combined, these results show that there is an increase in the (social) distance between the cubs and their mother.

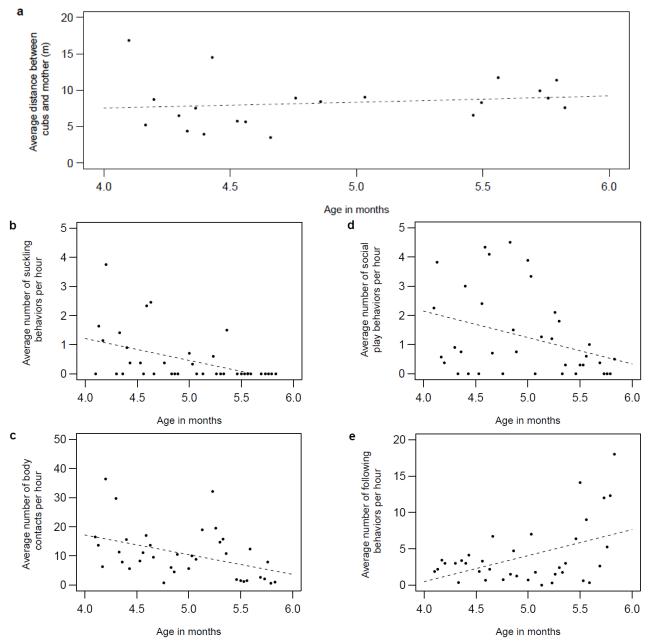


Figure 8. Indicators of the development of (social) distance between the two sun bear cubs and their mother. The values in *a* are plotted as the averages per day of all distances of A and B measured at the end of each 1-minute interval. The values in figures *b*-*e* are plotted as the average amount of 1-minute intervals per hour in which the behavior occurred. LMs are used to test for significant changes in behavior. a = A significant increase in the distance (m) between the cubs and their mother between the ages of 4 to 6 months is found (P < 0.05). b = There is a significant decrease in the amount of suckling behavior of the cubs (P < 0.01), c = in the amount of body contact between the cubs and their mother (P < 0.01), d = and in the amount of social play between the cubs and their mother (P < 0.01). e = The amount of following behavior, however, increases (P < 0.01), starting around the age of 5.3 months, which is the time at which the introduction of the male (individual D) took place.

DISCUSSION

This study aimed to investigate the development of explorative behavior and neophobia in two captive sun bear cubs. It was expected that the cubs would show more explorative behavior compared to their mother and that the amount of exploration of the cubs would decline over time. In addition, it was expected that the cubs would develop fear and neophobia, while at the same time the (social) distance to the mother would increase. The results show that the cubs explore more compared to their mother. However, the amount of exploration of both cubs does not decline over time. When it comes to neophobia, no indications are found for the development of this phenomenon; the latency to approach novelties even declines over time. The (social) distance between the cubs and the mother increases between the ages of 4 to 6 months, suggesting that this age could be the proper time frame for neophobia to develop.

The explorative behavior of the cubs differed from their mother for almost all different components. This is in line with prior studies stating that juvenile animals explore more than adults (Bergman & Kitchen, 2009; Benson-Amram & Holekamp, 2012; Biondi *et al.*, 2010; Carter *et al.*, 2018). Regarding the foraging-related explorative behaviors, the mother spends by far more time on this than her cubs.

No decline in exploration was shown by either individual A or B. This is in contrast with the hypothesis stating that exploration would decline as a consequence of the development of neophobia. The development of neophobia in these two sun bears is therefore questioned, especially when combining this with the results of the novel object tests. No indications of an increase in fear were found during the novel object tests or during the general observations. This contrasts the findings in brown bear cubs that had qualitatively demonstrated fear responses when presented with new odors, sounds, and objects (Pazhetnov & Pazhetnov, 2005). Fear reactions in brown bear cubs had fully developed at the age of 5 months, however, the development of this reaction depended on the degree of isolation from humans in their early life. The degree of isolation could be an explanation for the lack of fear in the two sun bear cubs studied here, who had been in close contact with humans during their development. Further research comparing wild and captive bear cubs should determine the extent to which the development of neophobia is affected by human presence. In case an absence of neophobia in wild cubs will be found as well, the development of neophobia in this species could be excluded.

Remarkably, the latency to approach novelties even decreased. This decrease may be caused by the fact that none of the stimuli was associated with danger, which is of course not the case in their natural habitat. These captive cubs may have learned that each novelty, especially the objects, are harmless toys which they can use to play with. Therefore, habituation may have occurred across the trials of the novel object tests and could have played a role in the fact that neophobia was not detected. The larger interest in objects is shown by the longer visit time and smaller distance from the stimulus during these tests. Objects appear to be more interesting/less frightening than olfactory and auditory stimuli. This has also been shown in other studies that used smells and sounds to test neophobia in animals (Crane & Ferrari, 2017). From an ecological perspective, wild animals, especially those in complex environments, are exposed to a large amount of sounds and smells in their daily life and tracking all of these would probably not be beneficial in terms of energy allocation. Novel objects may be rarer, and therefore more worth investigating. There is also a difference found in the total visit time and in the amount of fear/stress behaviors between the two different kinds of auditory stimuli. Both the visit time and the amount of fear/stress behaviors were larger during exposure to animal vocalizations compared to non-vocalizations, although the latter not significantly. Apparently, sounds that are meaningless are not worth tracking, while sounds that come from potential prey or predators are perceived as stressful and are investigated thoroughly.

Besides the lack of evidence for the development of neophobic responses, there is another argument for the fact that neophobia may not have developed in these two sun bears. This argument is the increase in (social) distance from the mother. Both the distance in meters and multiple parameters for social distance increased between the ages of 4 to 6 months. Similar results have also been found in polar bear cubs after leaving the maternal den (Greenwald & Dabek, 2003). This increase indicates that if neophobia would develop in sun bears, this age would be the proper moment. Thus, it is possible that no neophobia develops in sun bears. This would be in line with the neophobia threshold hypothesis, stating that neophobia would be ecologically more beneficial/likely for specialist species (Greenberg, 2003), and sun bears are opportunistic omnivores. Living in a complex tropical forest as an omnivorous generalist species, sun bears may be obstructed in their foraging if they would develop neophobia. Future research could compare neophobic responses of sun bear to a more specialist relative, for example the polar bear, to further test this theory.

However, there were some limitations in this study that may have influenced neophobic reactions of the cubs. During the novel object tests, the animals entered the area with the novel stimulus all together and thereby may have influenced each other's responses. Previous studies have shown the influence of a social context during novel object test and both increasing and decreasing effects on latencies have been found (e.g. Stöwe, Bugnvar, Heinrich, & Kotrschal, 2006; García et al. 2019; Oostindjer, Muñoz, Van den Brand, Kemp, & Bolhuis, 2011). In addition, there were some differing circumstances that may have had an effect on the behavior of the cubs, including the introduction of the male and the varying number of zoo visitors during the trials. More importantly, the neophobic responses, especially during the novel object tests, were difficult to assess because of the interplay between neophobia and the tendency to explore. These two motivations can occur simultaneously creating ambiguous behavior and masking the development of neophobia (Greenberg & Mettke-Hofmann, 2001).

In conclusion, this study shows that the explorative behavior of the two captive sun bear cubs does not decline between the ages of 4 to 6 months. Furthermore, there are no indications of the development of neophobia at this age, even though an increase in (social) distance to the mother was observed. Concerning wildlife rehabilitation, this study implies that close contact with humans, as is the case in captive environments, may disturb the development of proper fear reactions and neophobia needed for survival in the wild. However, the possibility remains that no development of neophobia occurs in sun bears at all, but further research comparing captive and wild sun bears should clarify this.

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REFERENCES

Bacon, E. S., & Burghardt, G. M. 1976. Learning and color discrimination in the American black bear. In M. R. Pelton, J.W. Lentfer, G.E. Folk (Eds.), *Bears: Their Biology and Management* (pp. 27-36). Morges, Switserland: International Union For Conservation of Nature and Natural Resources.

- Benson-Amram, S., & Holekamp, K. E. 2012. Innovative problem solving by wild spotted hyenas. *Proceedings of the Royal Society B: Biological Sciences*, *279*(1744), 4087-4095. <u>https://doi.org/10.1098/rspb.2012.1450</u>
- Bergman, T. J., & Kitchen, D. M. 2009. Comparing responses to novel objects in wild baboons (Papio ursinus) and geladas (Theropithecus gelada). *Animal cognition*, *12*(1), 63-73. https://doi.org/10.1007/s10071-008-0171-2

Biondi, L. M., Bó, M. S., & Vassallo, A. I. 2010. Interindividual and age differences in exploration, neophobia and problem-solving ability in a Neotropical raptor (*Milvago chimango*). *Animal cognition*, 13(5), 701-710.

https://doi.org/10.1007/s10071-010-0319-8

- Carter, G. G., Forss, S., Page, R. A., & Ratcliffe, J. M. 2018. Younger vampire bats (Desmodus rotundus) are more likely than adults to explore novel objects. *PloS* one, 13(5), e0196889. https://doi.org/10.1371/journal.pone.0196889
- Corey, D. T. 1978. The determinants of exploration and neophobia. *Neuroscience & Biobehavioral Reviews*, 2(4), 235-253.

https://doi.org/10.1016/0149-7634(78)90033-7

Crane, A. L., & Ferrari, M. C. 2017. Patterns of predator neophobia: a meta-analytic review. Proceedings of the Royal Society B: Biological Sciences, 284(1861), 20170583.

http://dx.doi.org/10.1098/rspb.2017.0583

- Dahl, F. M., Hansen, H. H., Vorup, L. D., Jensen, L. Ø., Spyridopoulos, P. S., Jensen, T. H., Pertoldi, C., Alstrup, A. K. O., & Pagh, S. 2020. Effect of enrichments on behavioural reaction norms of two captive polar bears (Ursus maritimus) in Aalborg Zoo, Denmark. *Genetics and Biodiversity Journal*, 61-72.
- de Mendiburu, F. 2020. agricolae: Statistical Procedures for Agricultural Research. R package version 1.3-3. <u>https://CRAN.R-project.org/package=agricolae</u>
- Fox, J., Weisberg, S. 2019. An {R} Companion to Applied Regression, Third Edition. Thousand Oaks CA: Sage.

https://socialsciences.mcmaster.ca/jfox/Books/C

ompanion/

- Fredriksson, G.M. 2005. Conservation threats facing sun bears, *Helarctos malayanus*, and experiences with sun bear reintroductions in East Kalimantan, Indonesia. In L. Kolter, J. van Dijk (Eds), *Rehabilitation and release of bears* (pp. 35-42). Köln, Germany: Zoologischer Garten Köln.
- García, G. O., Nicolli, A. R., Castano, M. V., Zumpano, F., Favero, M., & Biondi, L. M. 2019. Evaluation of neophobia in a threatened seabird: Olrog's Gull (Larus atlanticus) as a case study. *Emu-Austral* Ornithology, 119(2), 166-175.

https://doi.org/10.1080/01584197.2019.1568197

- Gilbert, B.K. 1999. Opportunities for social learning in bears. In H.O. Box, K.R. Gibson (Eds), *Mammalian social learning: comparative and ecological perspectives* (pp. 225-235). Cambridge, United Kingdom: Cambridge University Press.
- Greenberg, R.S. 2003. The role of neophobia and neophilia in the development of innovative behaviour of birds. In S.N. Reader, K.N. Laland (Eds), *Animal innovation* (pp. 175-196). New York, United States of America, Oxford University Press.
- Greenberg, R.S., Mettke-Hofmann, C. 2001. Ecological aspects of neophobia and neophilia in birds. In V. Nolan Jr, C.F. Thompson (Eds), *Current ornithology*, vol 16 (pp. 119-178). Boston, United States of America: Springer.
- Greenwald, K. R., & Dabek, L. 2003. Behavioral development of a polar bear cub (Ursus maritimus) in captivity. *Zoo Biology: Published in affiliation* with the American Zoo and Aquarium Association, 22(5), 507-514.

https://doi.org/10.1002/zoo.10095

- Hall, S.S., Swaisgood, R.R. 2009. Maternal care and cub development in the sun bear. *Ursus*, *20*(2):143-152. <u>https://doi.org/10.2192/09SC001.1</u>
- Martin, P., Bateson, P.P.G. 1993. *Measuring behaviour: an introductory guide*. United States of America: Cambridge University Press.
- Martin-Wintle, M. S., Shepherdson, D., Zhang, G., Huang, Y., Luo, B., & Swaisgood, R. R. 2017. Do opposites attract? Effects of personality matching in breeding pairs of captive giant pandas on reproductive success. *Biological Conservation*, 207, 27-37. https://doi.org/10.1016/j.biocon.2017.01.010
- Mazur, R., & Seher, V. 2008. Socially learned foraging behaviour in wild black bears, Ursus

americanus. Animal behaviour, 75, 1503-1508.

- Myers, P. J., & Young, J. K. (2018). Consistent individual behavior: evidence of personality in black bears. *Journal of Ethology*, *36*(2), 117-124. https://doi.org/10.1007/s10164-018-0541-4
- Nowak, R.M., Walker, E.P. 1999. Carnivora; family *Ursidae*. In *Walker's Mammals of the World*, vol 1 (pp. 678-694). Baltimore, United States of America: JHU press.
- Oostindjer, M., Muñoz, J. M., Van den Brand, H., Kemp, B., & Bolhuis, J. E. 2011. Maternal presence and environmental enrichment affect food neophobia of piglets. *Biology Letters*, 7(1), 19-22. http://dx.doi.org/10.1098/rsbl.2010.0430
- Pazhetnov, V.S., Pazhetnov, S.V. 2005. Re-introduction of orphan brown bear cubs. In L. Kolter, J. van Dijk (Eds), *Rehabilitation and release of bears* (pp. 53-61). Köln, Germany: Zoologischer Garten Köln.
- R Core Team. 2019. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. <u>https://www.Rproject.org/</u>.
- Renner, M. J., & Lussier, J. P. 2002. Environmental enrichment for the captive spectacled bear (Tremarctos ornatus). *Pharmacology Biochemistry and Behavior*, 73(1), 279-283.

https://doi.org/10.1016/S0091-3057(02)00786-4

Scotson, L., Fredriksson, G., Augeri, D., Cheah, C., Ngoprasert, D., Wai-Ming. W. 2017. *Helarctos malayanus* (errata version published in 2018). The IUCN Red List of Threatened Species 2017:e.T9760A45033547.

http://dx.doi.org/10.2305/IUCN.UK.2017-3.RLTS.T9760A45033547.en

Skripova, K. V. 2013. The behavior of Asiatic black bear cubs (Ursus (Selenarctos) thibetanus G. Guvier, 1823) in the process of adaptation to the natural environment. *Contemporary Problems of Ecology*, 6(1), 113-120.

https://doi.org/10.1134/S1995425513010162

- Stöwe, M., Bugnyar, T., Heinrich, B., & Kotrschal, K. 2006. Effects of group size on approach to novel objects in ravens (Corvus corax). *Ethology*, *112*(11), 1079-1088. <u>https://doi.org/10.1111/j.1439-0310.2006.01273.x</u>
- Swaisgood, R. R., White, A. M., Zhou, X., Zhang, H., Zhang,G., Wei, R., Hare, V. J., Tepper, E. M., & Lindburg, D.G. 2001. A quantitative assessment of the efficacy

of an environmental enrichment programme for giant pandas. *Animal Behaviour*, 61(2), 447-457. https://doi.org/10.1006/anbe.2000.1610

Wagman, J. D., Lukas, K. E., Dennis, P. M., Willis, M. A., Carroscia, J., Gindlesperger, C., & Schook, M. W. 2018. A work-for-food enrichment program increases exploration and decreases stereotypies in four species of bears. *Zoo biology*, *37*(1), 3-15. <u>https://doi.org/10.1002/zoo.21391</u>

Zeileis, A., Hothorn, T. 2002. Diagnostic Checking in Regression Relationships. *R News* 2(3), 7-10. URL: <u>https://CRAN.R-project.org/doc/Rnews/</u>

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