First successful hand-rearing and release to the wild of two orphan brown bear cubs in Greece

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**INTRODUCTION**

Orphan brown bears (*Ursus arctos L.*) are becoming more common in Greece due to increasing interactions with humans. Several options for dealing with orphan bear cubs exist, but hand-rearing them in captivity and then releasing them back to the wild is one of the most attractive (Beecham et al., 2015). Rehabilitation and release efforts provide managers the opportunity to support the welfare of disabled animals, attract public attention to conservation efforts and collect important biological information (Moore et al., 2007). Brown bears are considered to be endangered in Greece; in recent years however the population has made a significant recovery and it is estimated that >450 individuals currently live in the country (Karamanlidis et al., 2015). At the same time negative human – bear interactions have also been increasing (Karamanlidis et al., 2011) and therefore informed, science-based management and conservation measures are urgently required to safeguard the recovery and survival of this endangered brown bear population. In this study we present and evaluate the first rehabilitation and release attempt of brown bears in Greece.

**CASE HISTORY**

In April 2011, two orphan male, unrelated, brown bear cubs, aged 2 and 2.5 months respectively, were found in separate locations in north-western Greece. Following unsuccessful attempts to re-unite them with their mothers, the two cubs (named Little John and Nikitas) were transferred to the Bear Rehabilitation Centre of the non-governmental organization ARCTUROS. All research activities were carried out under the research permit 119628/1442 of the Hellenic Ministry of
Upon arrival the cubs were placed in a quarantined area (2.5 x 1.5 x 1.5m), and a detailed physical examination was performed. Both cubs were in a moderate body condition (i.e., Little John: 4kg; Nikitas: 3kg) and mildly dehydrated. Auscultation findings, respiratory rate and pulse rate were normal. Mucopurulent discharge was noticed in the eyes of Little John and was treated for 7 days with an antibiotic eye ointment (Fucidin® 1% eye drops, LEO Pharmaceutical Products Hellas Ltd). Mucus diarrhoea was observed in both cubs and was treated with nutritional measurements and administration of Diarsanyl Plus® oral paste (Ceva Animal Health Inc., KS USA) and Pro-Enteric Triplex® and Enterochronic® paste (Bioberica Veterinaria, Spain) in order to balance the intestinal flora. Abdominal palpation did not reveal anything abnormal, and faecal parasitological examinations were negative; however, as bear cubs of this age are very susceptible to parasitic infestations (Beecham 2006), a prophylactic treatment for endoparasites with Banmith paste® (Pfizer Inc., U.S.A) was administered and repeated 3 weeks later. The parasitic treatment continued again once every two months, with Drontal plus® (Bayer HealthCare LLC, U.S.A) (Papageorgiou et al., 2002). No other clinical signs of disease or any injuries were observed.

Rehydration with liquid-electrolytes, Almora plus® (oral pd, ELPEN Co. Hellas) and bottle milk feeding was initiated. In addition, vitamins and supplements, such as Vetamix Osteoform® (Lloyd, U.S.A), VetStar Vital® (Provet Hellas) and Calo Pet® (Vetoquinol, France) were also administered to the cubs as part of their regular diet. No vaccines...
were administered because their use is controversial and depends on the endemic risk of disease in the release area (Beecham, 2006).

As both cubs showed natural avoidance behaviour towards people, they were considered good candidates for re-introduction into the wild and were entered into an intensive rehabilitation program for nine months. During the initial stabilization period (i.e., 10 days), both animals were housed together in a small enclosure and had minimal contact with humans; only two caretakers and the supervising veterinarian had access to them. After the first ten days both cubs were transferred to a small fenced area (80 m²) where they remained for two months (acclimatization period) (Figure 1a). This area had two artificial dens, a pool and several stimulating objects (i.e., tree trunks, rocks) that were used to reduce the possibility of developing stereotypic behaviours (Criswell and Galbreath 2005). Contact with humans remained minimal, with only two caretakers entering the enclosure to feed the animals. Then, at the age of approximately 4.5 – 5 months the cubs were transferred to an enclosed one-hectare forest of beech (Fagus sp.) and oak (Quercus sp.) where they remained for approximately seven months (pre-release period) (Figure 1b, 1c); restricted contact with humans was continued until their release on 23 January 2012.

During the initial stabilization period the cubs were bottle-fed with canine milk formula (Ormilak®ORSCO - Laboratoire Vétérinaire, France) and honey (1-2 teaspoons); meals were provided around the clock, every three hours. The quantity of food provided was slowly tapered off and 10 days later bottle feeding was discontinued. From then on and for 2 months (i.e., during the acclimatization period) the bear cubs were fed 10-20% of their body mass daily; they received 4 – 5 bowls of milk (Veta-Lac Milk Replacer®, Lloyd, U.S.A.), rice baby cream (Nounou baby rice cream, Royal Friesland Campina, The Netherlands), honey, yogurt, fruits, Quaker (Quaker, Pepsiko, U.S.A), muesli, corn flakes, eggs and canned dog food (Royal Canin puppy, Royal Canin, U.S.A) in four meals. When moved to the forest enclosure in July (i.e., at the age of 4.5-5 months) the two bear cubs were fed twice daily. By mid-December feeding was gradually reduced to once every other day and then discontinued one month later. During this phase of rehabilitation (i.e., during the pre-release period), the bear cubs were fed milk, cereal, nuts, dry dog food, and several fruits and vegetables collected from the nearby forest. Solid foods were presented to bears in various ways to encourage them to search for their food as they do in the wild (Lintzenich et al., 2006).

The release of the two rehabilitated bear cubs was carried out on the 23rd of January 2012, when Little John and Nikitas weighed 50 and 45 kg respectively. At this time both bears had entered their hibernation phase and were slightly lethargic (inside the den in the enclosure). The two cubs were anaesthetized with a dart gun using a combination of xylazine (2mg/kg b.w.) and ketamine (3mg/kg b.w.) intramuscularly. Fifteen minutes later the drugs went into effect and the bears went down, showing no response to auditory stimuli and no head–lifting reaction to tactile stimuli (Caulkett and Cattet 2002). Handling was initiated and the eyes of both bear cubs were covered with a piece of cloth to protect the corneas from direct sunlight and to reduce optic stimuli. Weight and body measurements were taken and a detailed clinical examination was performed. Rectal temperature was taken with a standard digital thermometer. Their body temperatures ranged between 36.5-37.2°C. Monitoring of anaesthesia was carried out by checking the palpebral reflex, the positioning of the eye globe, the presence of nystagmus, the respiratory and heart rate, as well as by examination of the colour of the mucus membrane, the capillary refill time, the peripheral pulse and the body temperature. To evaluate the health status, blood samples were taken from the jugular vein for haematologi-
cal and biochemical analyses (Table 1) and faeces were collected for parasitological examination. 3-D GPS-GSM (global positioning system, global system for mobile communication) collars (GPS Plus, Vectronic Aerospace GmbH, Germany) with cotton spacers were placed on both animals for post-release monitoring.

After the end of this procedure Little John appeared cyanotic and therefore supplementary oxygen was given through a nasal catheter for 10 min. In order to reverse xylazine and accelerate recovery, atipamezol (0.20.1 mg/kg b.w. intramuscularly) was administered, and Little John recovered from anaesthesia 10 min later without any further complications. Nikitas recovered from anaesthesia one hour later without additional intervention. One hour after full recovery both bear cubs were transported to a forested area in Mount Vitsi in northwestern Greece (altitude 1,400 m above sea level) where an artificial den had been constructed in order to increase the cub’s chances of survival in the wild. Approximately 8 hours after the first anaesthetic episode, the two cubs were anaesthetized again using xylazine (2 mg/kg b.w.) and ketamine (3 mg/kg b.w.) intramuscularly. It was necessary to administer supplemental doses of xylazine (1 mg/kg b.w.) and ketamine (6 mg/kg b.w.) intramuscularly to Little John; this was most likely necessary due to the injection of the drugs into the fat during the first injection or due to a residual action of atipamezole. Both bear cubs were placed in the den and the entrance was partially sealed off with snow. The

<table>
<thead>
<tr>
<th>Haematological exam</th>
<th>Biochemical exam</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>Little John</td>
</tr>
<tr>
<td>RBC 8.69 M/μl</td>
<td>8.44 M/μl</td>
</tr>
<tr>
<td>HGB 19.0 gr/dL</td>
<td>18.4 gr/dL</td>
</tr>
<tr>
<td>HCT 55.0%</td>
<td>52.3%</td>
</tr>
<tr>
<td>MCV 63.3 fl</td>
<td>61.9 fl</td>
</tr>
<tr>
<td>MCH 21.8 pg</td>
<td>21.8 pg</td>
</tr>
<tr>
<td>MCHC 34.5 gr/dL</td>
<td>35.1 gr/dL</td>
</tr>
<tr>
<td>RDW-CV 16.0%</td>
<td>16.0%</td>
</tr>
<tr>
<td>WBC 5.3 K/μL</td>
<td>5.6 K/μL</td>
</tr>
<tr>
<td>NEUT 3.6 K/μL</td>
<td>3.8 K/μL</td>
</tr>
<tr>
<td>LYM 1.1 K/μL</td>
<td>1.3 K/μL</td>
</tr>
<tr>
<td>MONO 0.3 K/μL</td>
<td>0.2 K/μL</td>
</tr>
<tr>
<td>EOS 0.29 K/μL</td>
<td>0.27 K/μL</td>
</tr>
<tr>
<td>BASO 0.01 K/μL</td>
<td>0.01 K/μL</td>
</tr>
<tr>
<td>LUC% 0.02 K/μL</td>
<td>0.01 K/μL</td>
</tr>
<tr>
<td>PLT 349 K/μl</td>
<td>307 K/μl</td>
</tr>
<tr>
<td>MPV 7.9 fl</td>
<td>7.5 fl</td>
</tr>
<tr>
<td>TS 6.8 gr/dL</td>
<td>7.4 gr/dL</td>
</tr>
</tbody>
</table>
gested that the formula should be high in calories, protein and fat and low in carbohydrates; in order to achieve the desired rapid growth curves bears in their initial stages of their rehabilitation should be fed around the clock every 2-3 hours (Huber et al., 1993). In our cases the formula used was tolerated well and the cubs gained body mass rapidly. Based on our experience we recommend that during these initial phases of feeding (but also throughout the entire rehabilitation process) rehabilitated animals should be closely monitored for bloating, inap - petence, discomfort and constipation. Gradually, the number of feedings per day and the quantity of food can be tapered off to 10-20% of their body mass. It has been suggested that at weaning (i.e., approximately at the age of 5-6 months) the diet of bears in rehabilitation should consist primarily of fruits, dry dog food (for nutritional balance), hard mast, some vegetables, and occasionally fish or the carcasses of wild animals (Lintzenich et al., 2006). In our cases the feeding regime followed (i.e., Quaker, muesli, corn flakes, eggs, as well as several fruits and vegetables collected from the nearby forest) proved to be very successful, as until the time of their release both cubs gained signifi - cant weight and were, based on the results of their haematological, biochemical and parasitological exams, in a good health condition.

Haematological and biochemical exams are an important tool to assess health and understand the impact of disease on wildlife populations (Geffre et al., 2009); the baseline values of these exams may be affected by various host and ecological factors and therefore caution is advised when interpreting diagnostic data. As no baseline bear hematology and biochemistry studies have been conducted in Greece, our results were compared to other relevant data (Græsli et al., 2014) and were normal.

Bears are known to show exposure to a variety of disease pathogens. Active cases of disease are rare in the wild (Binninger et al., 1980), but there is always the possibility that released cubs may

interior of the den was monitored with an infrared camera. Monitoring of anaesthesia was achieved solely through the infrared camera. Full recovery from anaesthesia was completed uneventfully 90-100 min after the administration of the drugs (Figure 1d). Following a three-month denning period, both bear cubs emerged from their den in the wild in March 2012, and started moving in the wider area of northwestern Greece.

DISCUSSION
Raising orphan wildlife in captive-rearing facilities for release back to the wild is expanding globally (Kelly et al., 2010) and has been performed with success with orphan birds, marine mammals and primates (Agoramoorthy and Hsu, 1999; Golightly et al., 2002; Moore et al., 2007). For more than 30 years bear conservationists and managers around the world have been experimenting with the rehabilitation and release of bears; in north America the rehabilitation and release of American black bears (Ursus americanus) and brown bears has proved to be a very effective and valuable conservation tool (Beecham et al., 2015). In contrast, in Europe, where several small and endangered bear populations still exist (Zedroser et al., 2001), there has been little experience with bear rehabilitation (Bereczky, 2010). In this study we provide information and evaluate the usefulness of the rehabilitation and release of orphan brown bears as a management tool for the species in Greece.

New-born bear cubs are totally dependent on their mothers to receive adequate nutrition for rapid growth in the early stages of their life. Depending on species, bear cubs may nurse for up to 28 months and the bear milk they feed on has a very high caloric value (Jenness et al., 1972). Young bears in rehabilitation need to be fed 15-25% of their body mass daily (Lintzenich et al., 2006) of a substitute that has the same effect of rapid growth as the mother’s milk. It has been sug-
grow up together and socialize with a minimum of human contact proved to be successful, as neither cub showed any signs of habituation towards humans. This was true, not only for the time during the rehabilitation, but also after the release and during the one year of post-release monitoring (Beecham et al., 2015).

Anaesthetizing and handling bears has been an integral part of bear research for decades now and several studies have been carried out on the effects and dosages of drugs used during anaesthesia (Cattet et al., 2003a; b). In the present cases we used a combination of xylazine and ketamine. Although these drugs have been used extensively in anaesthetic protocols for wild and exotic animals, there is now a tendency to use other drug combinations, i.e. more potent α₂ adrenergic agents instead of xylazine, combined with tiletamine (plus zolazepam) instead of ketamine in bear anaesthesia (Caulkett and Arnemo, 2007). Nonetheless we chose this particular drug combination in order to achieve more rapid recovery (Caulkett N, 2007): during the first anaesthetic episode we strived for rapid recovery because a second anaesthesia was going to follow and we would not have any access to the cubs after they had been placed in the den. Our decision to use this drug combination was supported furthermore by the short-term duration of the procedure, and the small size of the bears (Caulkett and Arnemo, 2007). About 45 min after Little John was anaesthetized for the first time, his buccal mucosa appeared cyanotic. Although respiratory rate and heart rate showed no change, we decided to accelerate recovery, especially since the procedure had been completed.

CONCLUDING REMARKS

In the present study we present information on the procedures followed during the first successful attempt of hand-rearing and release in the wild of two orphan brown bear cubs in Greece. The pos-
sibilities of finding orphan bear cubs in Greece are likely to increase in the future, since the bear population in the country and human-bear conflicts have been increasing (Karamanlidis et al., 2011; 2015). The procedures that were followed and the experience that was gained during this first successful rehabilitation and release attempt will prove valuable in the future management and conservation of this endangered species in Greece and other countries.

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CONFLICT OF INTEREST STATEMENT
The authors of this article declare that they do not have any conflicting interests.

REFERENCES


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