Title: Darting primates in the field: A review of reporting trends and a survey of practices and their effect on the primates involved

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Abstract

Capture is one of the top ethical concerns of field primatologists, and darting is a common method of capturing primates. Little is published, however, about the safety of darting practices and conditions for the animals concerns. We conducted a literature review to examine trends in the reporting of darting methods and results, and two anonymous surveys of primatologists to gather information on darting methods and their effect on the primates involved. Among 111 papers reporting studies that darted primates, only 18 included full details of procedures, the total numbers of primates darted and the number, if any, of injuries and complications. In the surveys, 73 respondents reported on 2,092 dartings, including 44 injuries. The results show that smaller primates are more likely to be injured. 91% of seriously and fatally injured primates were arboreal, although arboreal species accounted for only 54% of the dartings. All primates who were fatally injured due to a dart hitting the abdomen or head were darted with a rifle, which were used for 45% of dartings. The presence of a veterinarian appears to reduce primate mortality in the event of injury or complications. Severe social effects of darting are not common, but include forced copulations, partner changes, and fatal attacks on infants. Lack of information about primate darting hinders refinement in methods that could improve safety. We hope this study will lead to greater sharing of information and the formation of a committee of experts in capture and immobilization to evaluate and regularly update protocols.

Keywords: Methods, Capture, Anesthesia, Ethics

Introduction

Capture is one of the top ethical concerns of field primatologists (Fedigan 2010). Direct contact with primates may be needed to remove snares or for medical intervention (Sleeman et al. 2000), and can further conservation and research goals such as the study of behavior, energetics and disease (Glander 2013; Unwin et al. 2011). Small primates, such as *Microcebus*, can be trapped or netted and manually restrained. Larger primates must be chemically immobilized (Unwin et al. 2011). Veterinarians caution that such field anesthesia is particularly challenging because the pre-anesthetic assessment of an animal's medical condition is limited to characteristics that are easily observed (Lynch and Bodley 2014; Mosley and Gunkel 2007). Primates may be chemically immobilized by injection after trapping or they may be chemically restrained by remote drug delivery (Glander 2013; Jolly et al. 2011). The latter, which we refer to as darting (following Glander 2013; Sapolsky and Share 1998), is a common method of capturing primates (Glander 2013). The practice may be increasing due to advances in radio tracking and telemetry (Honess and Macdonald 2011; Juarez et al. 2011) which require the capture of primates for the attachment of devices.

Primates present a wide range of challenges and risks to a darting program, due to their variation in size, habitat, and behavior. Darting is fast and effective, but it may cause trauma at the injection site and injury from a fall (Ølberg 2007) and there is inherent risk of serious injury or death (Glander 2013; Karesh et al. 1998; Unwin et al. 2011). Mammals under 20 kg, which covers the majority of primates, are at greater risk of fractures, injury to vital organs and other trauma from dart impact than larger mammals (Gunkel and Lafortune 2007; Hahn et al. 2007; McKenzie and Burroughs 1993). The degree of risk also varies with the age, health, and reproductive status of the animal (Unwin et al. 2009; Unwin et al. 2011). For example, mild cardiac disease or dehydration can exacerbate stressors common to chemical immobilization (Mosley and Gunkel 2007) such as hyperthermia, hypothermia, hypoxemia and respiratory and cardiovascular depression (Arnemo et al. 2014; Ko and West 2007; Williams and Junge 2014).

Two papers from the 1990s outline practices for darting arboreal (Glander et al. 1991) and terrestrial primates (Sapolsky and Share 1998). More recent papers provide general information on darting techniques (Glander 2013), detailed instructions on the use of anesthesia (Unwin et al. 2009) and guidelines for handling primates in the field (Unwin et al. 2011). However, there is a paucity of data on the relative safety of various darting practices with free-ranging primates (Juarez et al. 2011), although this is essential for refining practices.

As a first step in making darting safer for primates, we examine trends in reporting darting practices via a literature review and present the results of two surveys of darting practices in primatology. The surveys included reasons for darting, characteristics of the primates darted, procedures, injuries incurred and other consequences of darting to the animals concerned. Even in completely anonymous surveys, respondents tend to provide answers that will be viewed positively by others and, in doing so, compromise the accuracy of

results (Zuber and Kaptein 2014). This tendency may have been heightened in our survey as participants might have been concerned that they could be indirectly identified through their descriptions of the darting episodes. Some individuals with negative experiences may have preferred not to participate in the survey. Thus, we do not consider our results as accurate rates of overall injury rates and death. With this caveat, we analyze the risk of injury or death to primates from factors such as age and sex, arboreality, the delivery system of the dart, the experience of the darting team, the drug(s) used, the conditions in which the primate is kept, and the presence of a veterinarian.

METHODS

Literature Review

We searched PrimateLit (<u>http://primatelit.library.wisc.edu/</u>) for articles from 1940 to 2010 with the following search terms: "darting", "telemetry", "radio-collar", "capturing", and "capture". For 2011-2013 we used New York University's search engine "What am I searching?" with the keywords "dart and primate", "telemetry and primate", "radio-collar and primate". We also tried "capture and primate" but a scan of the articles indicated that most concerned the capture of genetic material or psychological attributes such as attention and it was not productive to investigate the list further. The searches included the complete text of articles, but were restricted to scholarly journals in English. We did not include abstracts and only followed up on original investigations of free-ranging primates, excluding *Microcebus* which are too small to dart. Four papers did not fit into our categories (for example they provided details on one aspect of darting, but did not provide information on other aspects of darting) and five papers presented information on a single primate, but it was not possible to determine whether other primates were darted; these nine papers are not included in our analysis.

For capture by darting, we noted the number of primates, the thoroughness with which the procedures were described, and whether or not the authors indicated harm, if any, to the primates. We coded the papers by the degree of information provided (Table 1). As we are interested in general trends, rather than precise numbers, we did not attempt to exclude papers which might be describing the same darting events.

Surveys

Two anonymous surveys were conducted using Survey Monkey. Email invitations with a link to the survey were sent to members of the PRIMATE SCIENCE List Serve (Survey 1) and the International Primatological Society (Survey 2). Based on a preliminary analysis of the response to Survey 1, we modified the survey by changing the format of several questions, eliminating some questions and adding three new ones. In both surveys, we asked respondents to provide information on their last darting period (consecutive or nearly consecutive days of darting in

one area). We asked for information about the species, sex and age class of the primates, the conditions of the darting, the composition and experience of the darting team, reasons for darting and, when applicable, injuries or problems that occurred. The surveys used "logic paths": a respondent who answered "yes" to the question "Were any primates injured or did any primates have an adverse drug reaction?" progressed to a series of questions about the injury, whereas someone who answered "no" skipped those questions. Survey 1 is available from the first author; Survey 2 is included online (Appendix S1).

The surveys included primates darted in various environments. Here we analyze data for free-range primates only. Survey 1 did not include a specific question about the environment in which the dartings took place, so JMS and EPC identified the environment based on answers to other questions separately and agreed in all 25 cases. In Survey 2, respondents could select between the following environments: free range, semi-free range, sanctuary, zoo, indoor research, and other. We used data for "free range" from this survey, but also included data from one well-known wild population who were categorized as "semi-free range" in Survey 2 because we judged that this was a mistake by a respondent who was not a native English speaker.

We combined the results of the two surveys for analysis at the level of the individual primate darted. In some cases information was incomplete or ambiguous. Although the survey was anonymous, respondents could provide contact information for follow-up questions. We included information obtained from follow-up emails in our dataset. When questions remained, we included as much data as possible without making assumptions that might skew the results. Although we asked respondents to report on their last darting period, we included data from 10 respondents reporting on 1008 dartings that appear to extend over more than one period. In two cases it seems two respondents reported the same events: we included the results of all four responses, but weighted the data 0.5 to account for this replication. We also used data weighting when respondent said 2-3 individuals, we entered two individuals with a weight of 1.0, and a third with a weight of 0.5. We weighted all other data 1.0.

We asked respondents to report their responsibilities during the darting event and how many primates they had been involved in darting in the past ten years. We asked them to give the reason for the darting, the species darted, how many males and females and how many infants and juveniles were darted. We used taxonomic information for descriptive analysis and to derive additional variables, using genera where appropriate species-level data were not available. We divided primates into terrestrial (most of waking time on ground), arboreal (most of waking time in trees) or semi-arboreal (waking time divided between ground and trees) categories to investigate whether risk factors varied for these categories. We assigned species to a category based on the preferred height data on "All the World's Primates" website (Rowe and Myers) and obtained body mass data from the same website. For a few species we needed to consult Primate Info Net Factsheets website or Fleagle (1999) for information. Where sex was unknown, we used the mean mass for males and females. We divided primates into small (< 1 kg), medium (1-10 kg) and large size categories (> 10 kg). We selected three categories as a compromise between detecting a non-linear relationship between risk and size (which requires >2 categories) and ease of interpretation.

Question 6 of the survey asked for the number of males and the number of females that were darted. We summed these numbers to obtain the total number of primates darted. Question 7 asked "Were any juvenile or infant primates darted?" with boxes for respondents to fill in the number of infants and the number of juveniles. When respondents provided an answer for the number of infants and juveniles that were darted, we considered all the other primates as adults. When respondents gave the numbers of juveniles, but not the numbers of infants, we assumed that the non-juveniles were adults. If respondents provided no answer to Question 7, we coded the age of all the primates as "unknown". For one respondent who wrote "three juveniles (subadults)", we coded three juveniles, based on the first part of the answer. In a second case where a respondent listed "subadults", and in a third where a respondent listed "adults, possibly subadults" we coded these primates as adults.

The combined sex and age class for individual primates was not always clear. However, we had information concerning the species, age class and sex of each seriously-injured primate. As we were not able to test more than one independent variable at a time in our analyses (see below), ambiguity about some age-sex classes does not affect our analyses.

We asked respondents about the experience of the darter. Based on their answers we formulated the following categories: very little experience (< 10 dartings), little experience (11-30 dartings), moderate experience (31-90 dartings), very experienced (> 90 dartings) and unknown. We also asked respondents about the anesthetic and dose they used. If a report listed more than one anesthetic or combination of anesthetics, but did not indicate the number of primates anesthetized with each combination, we counted both possibilities for each primate, but weighted the data 0.5. When respondents provided a range for the dose, we used the mean. We excluded one case where a respondent provided an answer that was obviously a mistake as it was 200 times higher than the next highest dose that was reported. We asked how primates were kept prior to release and provided a text box for respondents to describe how long primates were monitored before being released.

We asked respondents whether primates were injured and to describe the seriousness of the injury. We gave them the following choices to describe the **Degree of injury**: Released on schedule, recovered completely; Release delayed, recovered completely; Complete recovery not expected, survival chances good; Long-term survival not good; Died within a year; Died within a week. However, respondents selected one of these choices for only 14 of 43.5 injuries. Moreover, while cases in which primates died were unambiguous, other categories did not distinguish between the degree of the initial injury or problem and the outcome. For example, primates who suffered respiratory distress under anesthesia and primates with minor bruising were both categorized as "Released on schedule, recovered completely". However, in the first situation a primate might die without the intervention of a veterinarian, whereas expert care is not essential in the second situation. Respondents provided specific information, as comments, on most of the injuries whether they selected one of the choices we presented or not. Based on this information we grouped the responses into five categories: None, Minor (scrapes, small cuts, bruises), Serious (potential to affect survival or reproduction, required expert care), Fatal, Unclear. We also asked respondents about the cause of injuries.

We also provided text boxes for general comments and recommendations.

Data analysis

We used SPSS 20 for analysis. We summarized most data by category as % responses (the total N varies as not all respondents answered each question). We tested for a relationship between anesthetic dose and mass using the Pearson correlation coefficient. We tested the relationship between injury and each of age class, sex, size category, strata, delivery system, and experience of darter using Fisher's exact tests. We used a logistic regression to test for a relationship between injury and mass. Due to the small number of serious and fatal injuries, we could not assess the risk for more than one independent variable in the same analysis. We used a chi square test to test the association between presence of a veterinarian and the risk of injury and a Fisher's exact test to test the association between a fatal or not-fatal outcome for seriously-injured primates with and without the presence of a veterinarian.

Ethical note

This research was granted exemption from New York University Committee on Activities Involving Human Subjects.

RESULTS

Literature Review

The literature searches found 111 papers which reported data from primates who were darted. The first record of primate darting was in 1976 and there were few dartings during the 1980s (Table 1, Fig. 1). The 1990s saw a more than five-fold increase in the number of papers which include darting in their methods and in the 2000s there were more than twice as many dartings as in the previous decade. Since 2010, the rate of increase appears to have slowed.

Some primatologists provided detailed information on the darting procedures they use and the results (Fig. 1). Such reports, however, were relatively scarce (18 of 111 papers, 16 % overall). Moreover, fewer recent papers provided a comprehensive report on procedures and complications: prior to 2000, 13 of 32 papers (42 %) included these details; from 2000-13 only 5 of 79 papers (6 %) included this information (Category 1, Table 1 and Fig. 1). The 18 papers providing comprehensive information represent approximately 654 dartings. In this group there were 33 serious complications and serious or fatal injuries, a rate of 5 %. The 13 deaths reported were due to drug overdose (3), dart trauma (4), injury from the fall (3), sunstroke (1), unknown cause (1) and an infant baboon killed by a male between the time the mother was darted and the darting team found her (1). Non-fatal complications and injuries included an aborted pregnancy, the need for artificial respiration, broken bones and paralysis (one of the paralyzed primates died some time later) (Table S1). Three papers mention problems with darted animals overheating and in some cases this caused convulsions. Darting teams attempted to catch falling primates, with a mean success rate of 43 % (N=122) (Table S1).

An additional 11 papers representing approximately 561 darting captures provide less information on procedures but indicate whether or not primates were injured or died (Category 2, Table 1 and Fig. 1). Ten of these papers indicate no adverse effects from darting; one indicates that of 68 darted *Alouatta pigra*, two died and an infant was abandoned. Six papers in Category 3, representing 388 dartings, indicate no fatalities, but do not provide information about injuries. 54 papers in category 4, representing over 2,284 dartings, provide no information on fatalities or injuries. 22 papers in Category 5 do not provide information on fatalities, or injuries, and most did not include numbers of primates darted.

Surveys

Characteristics of respondents

We report on data provided by 73 people (unweighted) on 2091.5 (weighted) darted primates. Respondents had a range of responsibilities during the darting events, including respondents who assisted, non-veterinarians who darted, project supervisors or principal investigators, observers and veterinarians (Fig. 2). Respondents reported being involved with darting 0 to "1000s" of primates over the past 10 years, with a mean of 88.7 and a median of 30 (N = 63). 51 % of respondents described their answers to the surveys as "accurate", 41 % described them as "accurate and recollection" and 8 % described them as "recollection" (N = 59).

Reasons for darting and characteristics of the primates

Respondents gave a variety of reasons for darting primates in the field (Fig. 3). The largest categories were collection of genetic samples, attaching or removing telemetry equipment or marking individuals, and biomedical assessment. Other reasons were medical treatment (which included 9 cases of removal of snares and one treatment of wounds from a trap), "colony management" (although these were free-ranging primates, see note in Methods regarding language concerns), and translocation of at-risk populations. Genetic material was collected from 717 primates. Genetic sampling was given as the sole reason for darting 450 of these (63%) while respondents listed more than one reason for the captures in the other 267 cases (37%).

43 species of primates from 22 genera are represented in the data. The age and sex breakdowns for each species or genera, as well as the size and strata categories we assigned to the taxa are available online (Table S2). Respondents reported on darting 1083 adults (89 %), 125 juveniles (10 %), and 8 infants (<1 %). Slightly more females (N = 662; 56 %) were darted than males (N = 510; 44 %). Respondents reported darting primates from < 1 kg to 170 kg: most were medium (N = 1,341 of 2092, 64 %) and large species (N = 709, 34 %), with few small primates (N = 42, 2 %) darted. More than half of the primates darted were arboreal (N = 1,135 of 2092, 54 %), 41 % were terrestrial (N = 849), and only 5 % were semi-arboreal (N = 108). Of the arboreal primates, 96.1 % (N = 104) were medium-sized.

Procedures

Of the 1776 primates for whom we had data concerning the experience of the darters, 83.0 % were darted by very experienced darters, 10.2 % by darters with moderate experience, 3.1 % by darters with little experience, and 3.7 % by darters with very little experience. A veterinarian was present in 47.3 % of 2084.5 cases for which we had this information.

Overall, the most common delivery system for the dart was blowpipe, followed by rifle; few primates were darted using pistols. Rifles were used more commonly than blowpipes to dart arboreal primates (Fig. 4).

Respondents reported using ketamine for immobilization in the majority of cases (Fig. 5). Ketamine was used on its own for 36.4% of captures of arboreal primates. It was frequently used in combination with other drugs (Fig. 5). Doses of ketamine (including cases in which ketamine was used with a second drug) varied 14-fold (range 3.5 - 50 mg / kg, mean = $12.9 \pm SD$ = 12.2 mg / kg, N = 219). For 90.4 % of the dartings the dose was 10 mg / kg or less; for 9.6 % of the dartings it was 50 mg / kg). There was a negative correlation between dose and mass (r = -0.230, N = 219, P = 0.001), but this relationship was non-significant when we excluded outliers of 50 mg / kg (r = -0.008, N = 198, P = 0.906).

Equal parts of tiletamine and zolazepam were used in 23.6 % of dartings (Fig. 5). Doses ranged from 1-162.5 mg / kg (mean = $20.28 \pm SD 25.54$ mg / kg, N = 294) and were ≤ 30 mg / kg for 97.3 % of dartings. Acepromazine was used in combination with tiletamine-zolazepam in two (0.7 %) dartings (no information on doses). Phencyclidine, which is no longer available, was used in 8 cases. In 151 cases phencyclidine or propionylpromazine or a combination of the two drugs was used.

Between the time that primates were darted and released they were kept in a range of conditions. Of the 1,371 primates on whom we have data, 28 % were kept in a sack, 27 % in a kennel, and 19 % in a cage. Some primates were left on the ground (11 %), usually in the shade close to where they were darted. These primates were large or medium-sized. Some (2 %) small or medium-sized primates were wrapped in a blanket, towel or tarpaulin. 13 % of primates

were kept in "other" conditions which included immobilizing their limbs to prevent injury and placing them on a table in the shade.

Primates were monitored for 1.0 - 48.0 h after darting, with a mean of 5.6 h (+/- SD 7.1) and a median of 3.0 h (N = 878). In addition to these quantitative descriptions, a further 26 respondents, reporting on 651 primates, gave behavioral descriptions of monitoring, including 12 (46 %) respondents who described monitoring primates until "completely recovered" or "fully awake". Of these, two respondents reported monitoring overnight. A further 11 (42 %) described monitoring animals until they were at least partially recovered (e.g., "until the lemur was able to climb"; "until came round and walked away itself"). Three respondents answered "several hours." One respondent reported observing the primate for ten minutes after it woke up. Only one respondent reported using antidotes to reverse the effects of anesthesia.

Injuries

43.5 primates were injured or had problems with anesthesia as a result of darting (Table 2). Specific minor injuries were cuts to the gingiva, Achille's tendon or back. The number of minor injuries is an under-representation of the actual number of minor injuries as respondents did not always specify the number and/or age and sex of individuals who sustained minor injuries. 35.5 serious injuries include a case of a likely broken limb where the primate died from an unspecified cause. Of 25.5 fatal injuries, most were the result of falling, followed by the dart hitting an inappropriate part of the body, and one primate that died from a respiratory problem (Table 2).

Neither age-class (Fisher's Exact Test: P = 0.163) nor sex (Fisher's Exact Test: P = 0.072) was a significant risk factor for serious or fatal injury. However, we found a significant relationship between mass and risk of injury (Wald statistic 4.91, df 1, P = 0.027). For every increase of 100 g in mass the odds of serious injury decreased by 1.2 % (mass range = 0.59 – 120.95 kg; mean = 29.13 \pm SD = 45.14 kg; N = 2085). 100 % of injured primates were mediumsized (Fisher's Exact Test: P < 0.001). Arboreal primates, who were almost all medium-sized, were over-represented among injured primates (Fisher's Exact Test: P < 0.001): they accounted for 91.2 % of serious injuries (Fig. 6), although only 54 % of darted primates were arboreal. In contrast, while 41 % of darted primates were terrestrial, this group accounted for only 5.9 % of injuries (Fig. 6). Semi-arboreal primates accounted for a small proportion of dartings and injuries.

There was a significant association between darting with rifles and injury (Fisher's Exact Test: P < 0.001): rifles were used in 82.1 % of cases in which serious injuries occurred and we know the delivery system (Fig. 6) but were used in only 39.9 % of the all cases in which we know the delivery system (N = 1,188). This finding may be partly explained by the fact that rifles were used more to dart arboreal primates (see above), but even among arboreal primates, the proportion of injured primates was significantly greater for those darted with a rifle than with a

blowpipe (Fisher's Exact Test P = 0.022). In all eight cases in which a primate died from the dart hitting the abdomen or cranial cavity (Table 2), a rifle was used. In all 16.5 cases in which a primate died from the fall (Table 2), the primates were arboreal and were darted with a rifle. When we excluded the primate who had a problem with anesthesia from the analysis, we found that all the arboreal primates who had fatal injuries were darted with a rifle (Fisher's Exact Test P = 0.001). Among three primates who likely suffered from broken bones (Table 2), one was on the ground when darted with a blowpipe, climbed a tree and fell, and one was darted with a blowpipe and fell from mid-canopy. These two primates were immobilized with ketamine, which causes muscle rigidity when used on its own (Glander 2013). Dart trauma caused the third fracture. None of the primates who had fatal injuries from falls were immobilized with only ketamine. There was no significant relationship between the experience of the darter and the risk of injury (Fisher's Exact Test: P = 0.510, N = 1766).

A veterinarian was present for 37.3 % of injuries. The presence of a veterinarian was not significantly related to the number of injuries (Chi Square = 1.369, df = 1, P = 0.242). However, when we included only cases in which an injury occurred (N = 33.5), the risk of death was 25.5 times greater if no veterinarian was present than if a veterinarian was present (Fisher's Exact Test: P < 0.002) (Fig. 7). Respondents reported that veterinarians addressed a respiratory problem by administering doxapram, treated wounds, and delayed the release of a primate for several days while she recovered from paralysis of the forelimbs.

Other consequences of darting

Most respondents (82 % of 68) reported no changes in social behavior due to darting, 10 % noticed social changes, and 8 % found the question non-applicable. Some of the changes were relatively mild. For example, respondents wrote that: darted Ateles were usually solitary for 24 hours, but then rejoined the group; Alouatta returned to their groups within a week; and one Pongo individual became agitated and stayed away from the darting team. However, the comments indicate that the social impact of darting can be substantial. For example, an alpha male Cebus did not rejoin his group successfully and pair-partner changes sometimes occurred in Hylobates lar. Explaining why they only used darts for skin plugs in Hamadrayas baboons, one respondent wrote "If a leader male is anaesthetized, he will likely lose his females to another male. This may also occur if the female is anaesthetized, though that is less common." In another species, the alpha male died as a result of darting, changing the hierarchy in the group and the group travelled less than usual for two days. A few respondents described aggressive reactions to darting. In one case, a dominant male Gorilla attacked another Gorilla as it was regaining consciousness and "making weird drawn-out cries" but no injuries were observed. A respondent noted that Macaca nemestrina females had attacked their offspring in a previous darting (i.e., data not analyzed here). Although it was not the mating season, a darted female Eulemur was forced to copulate with all the adult males of her group. One respondent reported that a *Papio anubis* male from another troop killed an infant while the mother was unconscious during a previous darting (i.e., data not analyzed here).

Respondents' comments include recommendations on ways to minimize risk and avoid negative consequences, descriptions of specific difficulties, and general impressions, both positive and negative, of darting, summarized in Appendix S2.

DISCUSSION

Survey respondents reported darting primates for a variety of reasons, including research into primate ecology, behavior, genetics and morphology, individual welfare and conservation goals. The results of our literature review and surveys cannot give an accurate rate of overall injury rates and death, but suggest a 5 % rate of serious and fatal injuries and complications. Our discussions with numerous people involved in darting primates suggest that these surveys under-represent true rates of serious and mortal injury. However, even the 5 % serious injury rate is alarming. For comparison, capture experts find a mortality rate > 2 % unacceptable and that it merits re-evaluation of capture protocols for the chemical capture of artiodactylids (Spraker 1993), moose (*Alces alces*), brown bears (*Ursus arctos*), wolverines (*Gulo gulo*), Eurasian lynx (*Lynx lynx*), gray wolves (*Canis lupis*) (Arnemo et al. 2006) and African buffalo (*Syncerus caffer*) (Oosthuizen et al. 2009). Moreover, capture experts stress the importance of including deaths directly attributed to the capture event and those caused by secondary events such as myopathy, instrumentation with radio transmitters, and predation (Arnemo et al. 2006; Kock et al. 1987; Oosthuizen et al. 2009) which we do not consider in our study.

Sharing of information that could lead to refinement of protocols and reduced risk is rare among primatologists. Our literature review shows that some primatologists provide detailed, specific information on the darting procedures they use and the results. Such reports, however, are scarce, and the rate of acknowledged harm in papers that give few details of darting is very low, suggesting that researchers may provide information when there are no injuries, but not when problems occur. If this is true, it is understandable, but contributes to an unwarranted impression of safety and is detrimental to refining methods to reduce risk of injury. It also seems likely that the most disastrous darting attempts never result in publication. Disturbingly, despite a clear call for information in 1998 ("The capture and handling of free-ranging primates is always accompanied by risk of injury or mortality . . . sharing the undesirable impacts with the scientific community enables informed decisions to be made during future project development", Karesh et al. 1998), research published since 2000 is less likely to share information on darting than studies published earlier.

Our findings suggest that injury from darting is due to a combination of factors: many species are relatively small; falling is a serious hazard for arboreal species and a danger for semi-arboreal and terrestrial species; some species may become aggressive towards

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conspecifics from the disruption and stress of darting. Under-reporting of adverse outcomes may skew our results. Nevertheless, some of the results are compelling and have clear implications for practice. For others areas it is difficult to determine the safest approach given multiple risk factors.

Procedures

Experience of the darter Although many respondents commented on the importance of training and an experienced darting team (Appendix S2), we found no relationship between the experience of the darter and the likelihood of serious injury. This result is not surprising, as over 80 % of the primates in our survey were darted by very experienced darters and the sample size of seriously injured primates is small. The results indicate that serious injuries occur even with experienced darters and highlight the need for caution. We did not have information on the training of the darter, which may have been an important factor. The Canadian Council on Animal Care requires personnel involved in chemical restraint to complete a recognized training course and have continual practice or participation in immobilization events (Austin-Smith et al. 2003). This seems to be a reasonable requirement which could be applied more widely.

Drugs used Ketamine was used most often to immobilize primates (alone or in combination with other drugs), followed by tiletamine-zolazepam. Ketamine causes rigidity of muscles and increases the possibility of fractures (Glander 2013), and the two primates who sustained fractures from falling in our survey were darted with only ketamine. However, all of the other primates who had fall-related injuries (1 case of temporary paralysis and 16.5 fatal injuries) were arboreal and none were darted with only ketamine. Some researchers (e.g., Glenn et al. 1998) combine ketamine with drugs such as xylazine that relax muscles to counteract the effects of ketamine on muscles. Xylazine and other alpha-2 agonists such as medetomidine and dexmedetomidine can also be reversed safely with atipamizole; a further advantage of these drugs. The use of these anesthetic combinations also reduces the dose of ketamine needed and can thus reduce the side effects.

Anesthesia can also lead to changes in emotional state. For example, captive *Macaca* who had been anesthetized with ketamine for a routine health check spent significantly more time engaged in behaviors indicating anxiety and stress for two subsequent days than monkeys who had not undergone the anesthesia and health check (Bethell et al. 2012). Ketamine causes behavioral disassociation, but it does not induce amnesia, and the brain continues to register and act on sensory input during the induction and recovery phases. This input may increase stress for future anesthesia (Leopold et al. 2002). This may explain why four respondents to our survey reported difficulty in darting the same primates repeatedly (Appendix S2). Alternatively, these primates may have learnt to recognize the darter or the darting apparatus.

Doses of both ketamine and tiletamine-zolazepam varied greatly. The appropriate anesthetic and dose varies with species and circumstance (Unwin et al. 2011). Recommended

doses of ketamine range from 5-20 / kg for primates in sanctuaries (Unwin et al. 2009) to 25-33 mg / kg for free-ranging primates (Fernandez-Duque and Rotundo 2003; Glander 2013). Most respondents to the survey reported doses under 10 mg / kg, although about 10 % used 50 mg / kg, which is higher than the recommended doses. Recommended doses for a tiletamine-zolazepam, which is more potent, range from 1-15 mg / kg for primates in sanctuaries (Unwin et al. 2009), to \geq 20 mg / kg for free ranging primates (Glander 2013; Larsen et al. 2011). Respondents to the surveys reported doses that ranged far higher, although 97% of the doses were \leq 30 mg / kg. This suggests that primatologists do not always follow recommended dosages. However, it is also possible that respondents may have given incorrect information.

Although ketamine and tiletamine-zolazepam have wide margins of safety (Karesh et al. 1998), there are health concerns associated with their use, so the minimum effective doses should be used. Ketamine and tiletamine-zolazepam are processed by the liver and kidneys and higher doses may contribute to the death of weaker individuals (Karesh et al. 1998). Both ketamine and tiletamine-zolazepam can cause hyperthermia, which can cause brain damage or death (Glander 2013; Unwin et al. 2009). High ambient temperatures and stressful inductions increase the likelihood of hyperthermia (Ko and Krimins 2014). Five survey respondents reported hyperthermia and/or hypothermia. It is essential that the planning for anaesthesia minimises the risk of hypothermia and/or hyperthermia. A rectally-placed thermometer remains the best method to measure core body temperature and such monitoring will allow the detection of developing problems before they become life threatening. It is difficult to deal with hyperthermia and hypothermia under field conditions. Hypothermic animals should be warmed slowly by wrapping them in blankets and allowing them to recover in a draft-free area. Although ice water immersion is recommended in humans with hyperthermia, this would impractical under field conditions and an evaporative technique (e.g., intermittently spraying the animal's body with warm water while blowing a fan across the body, allowing the heat to evaporate) may be more appropriate. Severely hyperthermic animals will need an intravenous drip to mitigate other pathophysiological issues associated with hyperthermia, such as hypoglycaemia.

Under-dosing is also a concern as it may necessitate re-darting a primate (Karesh et al. 1998). *Cebus* took longer to become immobilized and fall with lower doses of tiletaminezolazepam (mean of 150 s at 26.3 mg / kg vs. mean 317 s at 17.5 mg / kg), increasing the possibility that the darting team lost sight of them (Crofoot et al. 2009). However, *Ateles* fell from trees quickly with low doses of tiletamine-zolazepam (mean 142.5 s at mean of 14 mg / kg), possibly due to the use of a low-impact darting system which minimized muscle trauma and hematoma formation and allowed for more efficient drug absorption (Karesh et al. 1998). Three respondents to the survey recommended use of small darts and needles (Appendix S2), which may promote drug absorption by minimizing trauma, and allow the use of lower doses without under-dosing. One possible reason for the wide range of dosages reported is that it is difficult to control the delivery of anesthesia when darting. All the anesthetics discussed work best when delivered deep into a muscle mass. However, optimal results cannot be guaranteed with darting as some anesthetics may be delivered subcutaneously, depending how the dart hits the animal. As a result, the animal may take longer than expected to go to sleep and the quality of the anesthesia may be poor. Darts may also deliver anesthetic too deep, for example into a bone, resulting in fast, but short-lived anesthesia, as well as possible fracture to the bone. In such situations, the full dose may not be administered, which may explain the wide range of dosages reported. We recommend, when possible, waiting until an animal is not moving before attempting darting. Trying to hit a moving target is another reason for reduced anesthetic effectiveness as it is less likely the anesthetic will be delivered deep into the muscle.

Where animals are kept Between the time that primates were darted and released they were most often kept in cloth sacks, kennels or cages. Large primates were often left on the ground or placed on a tarpaulin. Although sacks may be acceptable for short periods, respondents reported that some primates were kept in sacks overnight, without access to food or water for 24 hours. This allows full recovery from anesthesia, but primates may urinate, defecate and vomit in the sack, leading to discomfort (EC personal observation). Sacks also limit the possibility of observing or assisting a primate in distress. A primate with a fracture in a sack cannot freely adjust their body and pain from the fracture may be greatly increased.

Glander (2013) discourages the use of cages as visual stimulation may cause stress, resulting in self-inflicted injury and/or diarrhea. Respondents, however, reported that cages covered with blankets to reduce visual stimulation worked well (see also Fernandez-Duque and Rotundo 2003). We are not aware of primates in *covered* cages who have damaged themselves.

Guidelines recommend that an animal held for more than twelve hours be kept in an appropriate environment that minimizes stress and provided with suitable food and water (Austin-Smith et al. 2003; Sikes et al. 2011). We, therefore, suggest that covered cages be available during captures in which the primate is kept overnight and as a back-up for cases in which there is an injury or additional observation is needed.

Observation Survey respondents reported keeping primates for between one and 48 hours. The length of time needed to keep primates post anesthesia before releasing them requires knowledge of the species, and perhaps of individuals, and weighing of relevant risks. For example, keeping primates under observation for longer time periods should reduce the probability that a mildly-sedated primate will be attacked by a predator or conspecific, an occurrence reported by a few respondents. However, researchers report that longer recovery periods for *Aotus azaria* increased the possibility that the primates would be rejected by their social group (Fernandez-Duque and Rotundo 2003; Juarez et al. 2011), similar to fighting after separation longer than 12 hours in pack animals (Larsen and Kreeger 2014).

Size Respondents to the surveys reported darting primates ranging in mass from < 1 kg to 170 kg. The results indicated that smaller primates were more likely to be injured. The results of the literature review also showed that all primates who sustained dart-induced injuries were < 10 kg. Non-primate mammals < 20 kg are considered to be at greater risk of dart-induced injury than larger mammals (Gunkel and Lafortune 2007; Hahn et al. 2007; McKenzie and Burroughs 1993). The relatively small target areas of many primate species can be difficult to hit (Ølberg 2007). In our surveys, all of the primates who died from the dart hitting the head were < 10 kg. In an additional case, the dart fractured the hind limb of a primate < 10 kg. All these primates were darted with rifles. The Canadian Council on Animal Care cautions that high-velocity dart rifles are capable of killing most mammalian species, and are less accurate than firearms (Austin-Smith et al. 2003). They note that low-velocity systems cause less trauma than high-velocity systems, but are more limited in use. Blowpipes provide the greatest margin of safety and minimize the risk of dart-induced trauma for small primates (Williams and Junge 2014). Blowpipes are also suggested for wild aardwolves (Proteles cristata) (Hahn et al. 2007), while other smaller mammals such as clouded leopards (Neofelis nebulosi) and Asiatic golden cats Catopuma temminckii) are trapped (Grassman et al. 2004). Based on the results of our survey, and reviews of primate and non-primate literature, we recommend that researchers planning to capture primates consider the use of blowpipes and traps rather than rifles very carefully for primates under 20 kg.

Stratum Responses to the surveys show that all primates who died from injuries sustained from falling were arboreal and were darted with rifles. Blowpipes were the most common delivery system for the dart, but the use of blowpipes is limited to a maximum height of 7 m, whereas rifles can be used for greater distances (Fernandez-Duque and Rotundo 2003) and the relationship between rifles and fatal injuries from falls is probably due to the greater likelihood that primates found higher in the canopy were darted with a rifle, rather than a direct relationship between fatalities and the use of a rifle. Despite researcher's best efforts, the literature review indicates that only 43 % of falling primates were caught. There are multiple possible reasons for this: primates may move very quickly after being darted (Crofoot et al. 2009); chasing the primate may drive it higher into the canopy (Jones and Bush 1988); dense foliage may obscure the location of the primate (Olupot 1999); and undergrowth may impede the movements of the catchers (Lemos de Sá and Glander 1993; Olupot 1999) or make it impossible to spread a net (Glenn and Bensen 1998). For non-primate mammals, such as otariid seals and koalas (Phascolarctos cinereus), experts recommend avoiding situations in which there is danger of the animal falling (Haulena 2014; Holz 2007). Respondents to our survey reported no incidents of fatal injuries from falls for terrestrial and semi-arboreal species, although there were two cases of broken bones. These findings suggest that darting teams were careful and were able to minimize the risk of falls for terrestrial and semi-arboreal primates. For arboreal primates, however, the findings suggest that researchers should fully consider alternatives to darting, such as traps (Jolly et al. 2011). Traps have been used successfully for arboreal primates and may be safer and more efficient, capturing larger numbers of animals more quickly (Aguiar et al. 2007; Garber et al. 1993; Monteiro et al. 2007; Oliveira and Dietz 2011; Rocha et al. 2007; Savage et al. 1993). Traps, however, also involve risk (Brett et al. 1982) and in some situations they are time-consuming and unfeasible (Fernandez-Duque and Rotundo 2003). For many wild animals, darting is less stressful than trapping or netting (Arnemo et al. 2014).

Further environmental issues and procedures, which we did not address in our surveys, are also likely to influence the safety of animals concerned. These include the distance between the darter and the primate, the height of the primate, presence of water hazards or predators, the ambient temperature, the time taken to dart an individual, and the type of dart used. In particular, repeated attempts to dart an individual may increase activity and stress and result in greater likelihood of hyperthermia or other adverse events.

Veterinarians Although results of the surveys suggest that veterinarians can improve the chances of recovery when complications occur -- chances of death were 25.5 times greater without a veterinarian -- veterinarians were present for only half of the dartings. Respondents reported that veterinarians administered drugs, treated wounds, and delayed the release of a primate to recover from paralysis. The potential benefit of veterinary care might increase if primates can be kept for days or weeks to allow time for healing and for the administration of drugs, if needed. In addition, veterinarians' knowledge of reversal agents in drug combinations can reduce doses of ketamine which cannot be reversed and thereby reduce monitoring requirements and the risk of long-term sedation. Some countries (e.g., Tanzania, Kenya, United Kingdom) require anaesthetics to be used only under veterinary supervision (Craft 2008; Kenya Law Reports 2011; Royal College of Veterinary Surgeons 2015a,b). Although it is now illegal for non-veterinarians to anaesthetize wildlife in many countries, primatologists should consider such a requirement for darting primates even when it is not a legal requirement. The Code of Best Practices for Field Primatology states that there should be a plan of action in the event that an animal is injured, requires veterinary attention, or must be euthanized (Riley et al. 2014).

Conclusions

The results of the literature review and surveys indicate that darting primates involves risk of serious or fatal injury. Existing guidelines stress the importance of using the least invasive methods possible (Animal Behavior Society 2013). For example, samples for genetic and endocrinology studies can be collected non-invasively (Goossens et al. 2011; Hodges and Heistermann 2011). When direct contact is necessary, guidelines state that the minimum number of animals should be captured and pain and distress should be avoided (American Society of Primatologists; Riley et al. 2014). Researchers should collaborate to reduce the

number of primates darted. In some cases, they should refuse to conduct a study, if they are dissatisfied with their assessment of risks to the primates involved.

A striking result of the literature review is the need for greater sharing of information from each darting, in particular from instances in which injuries or fatalities occur. We join earlier authors (Karesh 1998, Glander 2013) in calling for detailed reporting of anesthetic and darting protocols to refine darting methods. Authors should include full details of their methods and justify them, while reviewers and editors should check for this. This information will aid researchers wishing to capture primates in considering their methods carefully, and exploring all possible options.

Specific information on best practices for each species and circumstance is needed to minimize the risk of serious injury or death to darted primates (Osofsky and Hirsch 2000). Those working with primates should take their inspiration from Scandinavia, where professionals with decades of experience, including thousands of immobilizations, have developed protocols that have substantially reduced direct, indirect and delayed mortality of chemically immobilized large mammals (Arnemo et al. 2006; Arnemo et al. 2012). A protocol for each species is subject to ongoing revision based on a mortality assessment undertaken after every capture-related death to evaluate how changes in anesthesia and methodological approaches could have prevented the death. The protocols are updated on a regular basis and are available online. These protocols have reduced mortality to below 2 % for some species and the ultimate goal is the elimination of mortality (Aremo et al. 2006).

We recommend a similar effort for primates. Those capturing primates should collect data on the immediate and long-term direct and indirect consequences of darting and other methods of capture. The data should be submitted to a committee of experts in capture and immobilization, who should evaluate various methods of capture – darting, trapping, netting – for each species and circumstance. Every serious complication or injury should be evaluated to determine whether a change in protocol could have prevented the problem. Based on this ongoing data stream, protocols should be updated on a regular basis and available online. We can and should do more than we are doing to reduce the risks associated with capture. As scientists who study our closest animal relatives and link to the animal world, we should be leading the way to more humane treatment for all animals.

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SUPPORTING INFORMATION

Appendices S1-S2, Tables S1-S2, and References for Electronic Supplementary Material are available online.

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Table 1. Criteria used to categorize papers by the level of information provided about darting (See Fig. 1 for numbers of papers at each level of information by decade).

Level of	Details	
information		
1	includes detailed and specific information on procedures and problems, if	
	they occurred, and responses to the problems	
2	includes the rate of fatalities and other health problems that resulted from	
	darting and more general information on procedures	
3	includes information on procedures and directly or in-directly reports that no	
	primates died as the result of darting, but does not indicate whether or not	
	there were injuries or other problems	
4	provides information on procedures, but does not indicate whether or	
	there were fatalities or other health problems	
5	Indicates only that primates were darted or refers the reader to other	
	manuscripts for the information on darting procedures. In some cases those	
	papers provided comprehensive reports; in other cases, the cited papers	
	contained few details of darting procedures or simply cite further papers	

Description	Number
minor	3
extent of injuries unclear	7
serious	8
broken bones	3
paralysis (lasted several days)	1
dart to testicle	1
hypothermia	1
respiratory problem	1
aborted fetus	1
fatal	25.5
fall	16.5
dart hit abdomen or cranial cavity	8
respiratory problem	1
Serious and fatal injuries	33.5
Total	43.5

Table 2. Frequency, seriousness, and cause of injuries to primates as a result of darting, basedon anonymous surveys of primatologists (N = 2,092)



Fig. 1 Number of studies published that included darting primates by decade, and the details provided of the darting (from most comprehensive (1) to no detail (5), see Table 1 for details of categories)



□assisted

non-veterinarian who dartedsupervised

∎observed

veterinarian

■other / unknown

Fig. 2 Roles of respondents (N = 73).



Fig. 3 Reasons for darting primates in the field, based on anonymous surveys of primatologists. N = 1,576 primates. Respondents gave more than one reason for darting 565 primates. "Other" reasons included physiological study and dental casts.



Fig. 4 Delivery systems used to dart primates, based on anonymous surveys of primatologists (N = 1,188). 'Rifle' includes CO_2 rifles, Telinject Vario 1V air powered breech-loading pistol custom-modified as a rifle, and a modified 22 rifle, while 'pistol' includes CO_2 and air powered pistols



Fig. 5 Drugs used when darting primates, based on anonymous surveys of primatologists. Black fill indicates only one drug was used. t-z = tiletamine-zolazepam (N = 1,596).

*Other: medetomidine (0.7 %), detomidine (0.2 %) levomepromazine (0.2 %), and midazolam (0.2 %)



Fig. 6 Strata and delivery systems used when primates were injured during darting, based on anonymous surveys of primatologists (N =35.5).



Fig. 7 Fatal and not-fatal outcomes for primates with serious injuries and complications with and without the presence of a veterinarian, based on anonymous surveys of primatologists.