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Effect of passive acoustic sampling methodology on detecting bats after declines from white nose syndrome

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Concomitant with the emergence and spread of white-nose syndrome (WNS) and precipitous decline of many bat species in North America, natural resource managers need modified and/or new techniques for bat inventory and monitoring that provide robust occupancy estimates. We used Anabat acoustic detectors to determine the most efficient passive acoustic sampling design for optimizing detection probabilities of multiple bat species in a WNS-impacted environment in New York, USA. Our sampling protocol included: six acoustic stations deployed for the entire duration of monitoring as well as a 4 x 4 grid and five transects of 5-10 acoustic units that were deployed for 6-8 night sample durations surveyed during the summers of 2011-2012. We used Program PRESENCE to determine detection probability and site occupancy estimates. Overall, the grid produced the highest detection probabilities for most species because it contained the most detectors and intercepted the greatest spatial area. However, big brown bats (*Eptesicus fuscus*) and species not impacted by WNS were detected easily regardless of sampling array. Endangered Indiana (*Myotis sodalis*) and little brown (*Myotis lucifugus*) and tri-colored bats (*Perimyotis subflavus*) showed declines in detection probabilities over our study, potentially indicative of continued WNS-associated declines. Identification of species presence through efficient methodologies is vital for future conservation efforts as bat populations decline further due to WNS and other factors.

Key words: White-nose syndrome, detection probability, Indiana bat, little brown bat.

INTRODUCTION

White-nose syndrome (WNS) is a disease of cave hibernating bats caused by the fungal agent, *Pseudogymnoascus destructans*, first documented in North America in 2006 (Blehert et al., 2009). Since its onset, WNS has caused the deaths of >6 million bats (USFWS, 2013) and has rapidly spread from central New York to at least 22 states and 5 Canadian provinces. *P. destructans* has been documented from four additional states, but infection in

bats with discernible lesions remains uncertain, that is, presence of *P. destructans* of DNA has been discovered without concomitant disease. In the context of severe bat population declines, sampling methodologies must be modified to account for these lower chances of detection. As WNS continues to spread and bat populations decline further, biologists invariably will need to rely more on acoustic detection, as a matter of necessity, as the primary

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method of monitoring bats on the landscape.

Acoustic monitoring is a non-invasive sampling technique that has become commonplace over the last decade for investigating ecology, species assemblages and relative abundance of bats within landscapes and/or relative to land management therein (Johnson et al., 2002; Milne et al., 2004; Ford et al., 2011; Johnson et al., 2011b). Use of acoustics has allowed the detection of greater overall species richness more quickly and over greater spatial extent than traditional capture methodologies such as mist-netting (Murray et al., 1999; O'Farrell and Gannon, 1999). Although most research suggests that use of both capture and acoustic techniques provides the more complete assessment of a bat community, the additive benefit of netting to increase detection probability is lessened in a post-WNS landscape because of low capture rates. Furthermore, in these severely impacted areas, the need for very high levels of sampling effort to detect declining species simply will be logistically prohibitive in that the efforts required may approach the entire duration of when temperate forest bats are present on the landscape (Coleman, 2013). As managers begin to incorporate acoustical methods, questions remain on the most effective deployment strategies in terms of duration and configuration of multiple detectors.

The Anabat acoustic detector (Titley Electronics Ballina, New South Wales, and Australia) is a type of frequency-division detector that has been used widely to evaluate species-specific habitat use and foraging activity (Betts, 1998; O'Farrell and Gannon, 1999; Britzke et al., 2002; Britzke, 2003; Ford et al., 2006). Although Anabats have been field evaluated (Johnson et al., 2002; Britzke, 2003; Milne et al., 2004; Brooks and Ford, 2005; Britzke et al. 2011), no research has assessed multiple passive sampling arrays for detecting numerous species in the context of WNS-impacted landscapes. Accordingly, the objective of our study was to assess detection probabilities and occupancy estimates of bats among differing spatial and temporal arrangements of Anabat detectors in a post-WNS landscape.

MATERIALS AND METHODS

This study was conducted at the Fort Drum Military Installation (Fort Drum) in Jefferson and Lewis counties in northern New York (44°00'N, 75°49'W). Fort Drum is a U.S. Army installation of approximately 43,000 ha that lies at the intersection of the St. Lawrence-Great Lakes Lowlands, the foothills of the Adirondack Mountains, and the Tug Hill Plateau ecoregions within the Black River and Indian River drainages. The nearby Niagara Escarpment (10–15 km west) contains karst formations with caves that support hibernating bats (Fenton, 1966). Largely undeveloped except in the Cantonment area, much of the Fort Drum landscape is a forested habitat dominated by northern hardwood types of varying successional stages. Approximately 20% of the installation area is comprised of wet meadows and beaver (*Castor canadensis*) impacted streams and ponds.

In the summers of 2011 and 2012, we deployed acoustic bat detectors across Fort Drum in various sampling arrays. We used a mix of Anabat II detectors connected to compact flash-storage Zero-

Crossings Analysis Interface Modules and wholly-contained SD1 and SD2 units (Titley Electronics, Ballina, New South Wales, Australia)¹. We calibrated all units using an ultrasonic insect deterring device prior to use in the field (Larson and Hayes, 2000). We placed Anabat units in weatherproof boxes with polyvinyl chloride (PVC) tubes attached that contained a small hole in the bottom for water drainage (O'Farrell, 1998). Boxes were placed on 1.5 m tripods aligned in a manner that allowed sound to enter the PVC tubes at a 45° reflective angle to be received by Anabat transducers perpendicularly (Britzke et al., 2010).

Randomly selected acoustic monitoring stations may not successfully detect endangered Indiana bats (*Myotis sodalis*) or little brown bats (*M. lucifugus*) due to recent WNS declines (Ford et al., 2011). Therefore, we focused monitoring efforts on these species by placing arrays of detectors near a little brown bat maternity colony in an artificial bat house (Dobony et al., 2011) and known historic Indiana bat maternity areas at Fort Drum (Johnson et al., 2011a). We deployed detectors to record passively at permanent stations that remained in the field for the entire summer season, linear stream transects of 5–10 detectors deployed for 6–8 nights at a time, and a 4 x 4 "grid" of detectors that also were deployed for 6–8 days per sampling session (Figure 1).

To ensure that more than one Anabat did not collect data on the same bat simultaneously, we separated individual detector sites by 200–250 m. The exception to this was a double transect of five sites where two detectors were pointed in opposite directions at each site and data from both units were combined. We chose deployment locations and the azimuth of microphone direction at each site to maximize call quality. For example, we specifically targeted sites with abundant open space such as forest canopy gaps, forested trails with open corridors, or over open water where many bat species would be expected to forage (Ford et al., 2005). We set Anabats to record data continuously from approximately 1900 to 0700 h over our sampling sites, once in 2011 for each sampling array and 4 times each for the arrays in 2012 for a total of 5 survey events. We changed batteries and memory cards as needed and downloaded data to a laptop computer using the CFCread program (Titley Scientific, Ballina, Australia).

We analyzed call files using EchoClass (v1.1, U.S. Army Engineer Research and Development Center, Vicksburg, MS, USA). Although the ability to identify bat calls to the species level has been criticized (Barclay, 1999), research has suggested that good quality calls of eastern North American bats can be identified both qualitatively (O'Farrell et al., 1999) and quantitatively (Britzke et al., 2002; Britzke et al., 2011). To minimize the impact of species identification when accuracy is less than 100%, we considered species of bats to be present at a site (detector station) if the maximum likelihood *P*-value estimate for an individual species' identified call was $\geq 90\%$.

We created nightly presence-absence detection histories from the acoustic data for the 9 possible species at Fort Drum (Gorresen et al., 2008). We considered each nightly survey independent due to the separation of sites and break in sampling during daylight hours. Because double transect detectors may have recorded the same bats simultaneously, both Anabats at each site were considered a single unit for computing detection histories, that is, if a species was detected by either Anabat at a site on a given night it was considered to be present at that site. For each bat species, we attempted to fit a candidate set of 15 models to determine whether sampling habitat ("wet" = riparian zone or lake, wet meadow, or beaver swamp or "dry" = upland forest or field), year, or time of season (Table 1) impacted estimates of overall occupancy or detection (MacKenzie et al., 2002) using program PRESENCE (version 2.4, Hines and Mackenzie, 2008). We ranked models using Akaike's

¹Any use of trade, firm, or product names is for descriptive purposes only and does not imply endorsement by the U.S. Government.

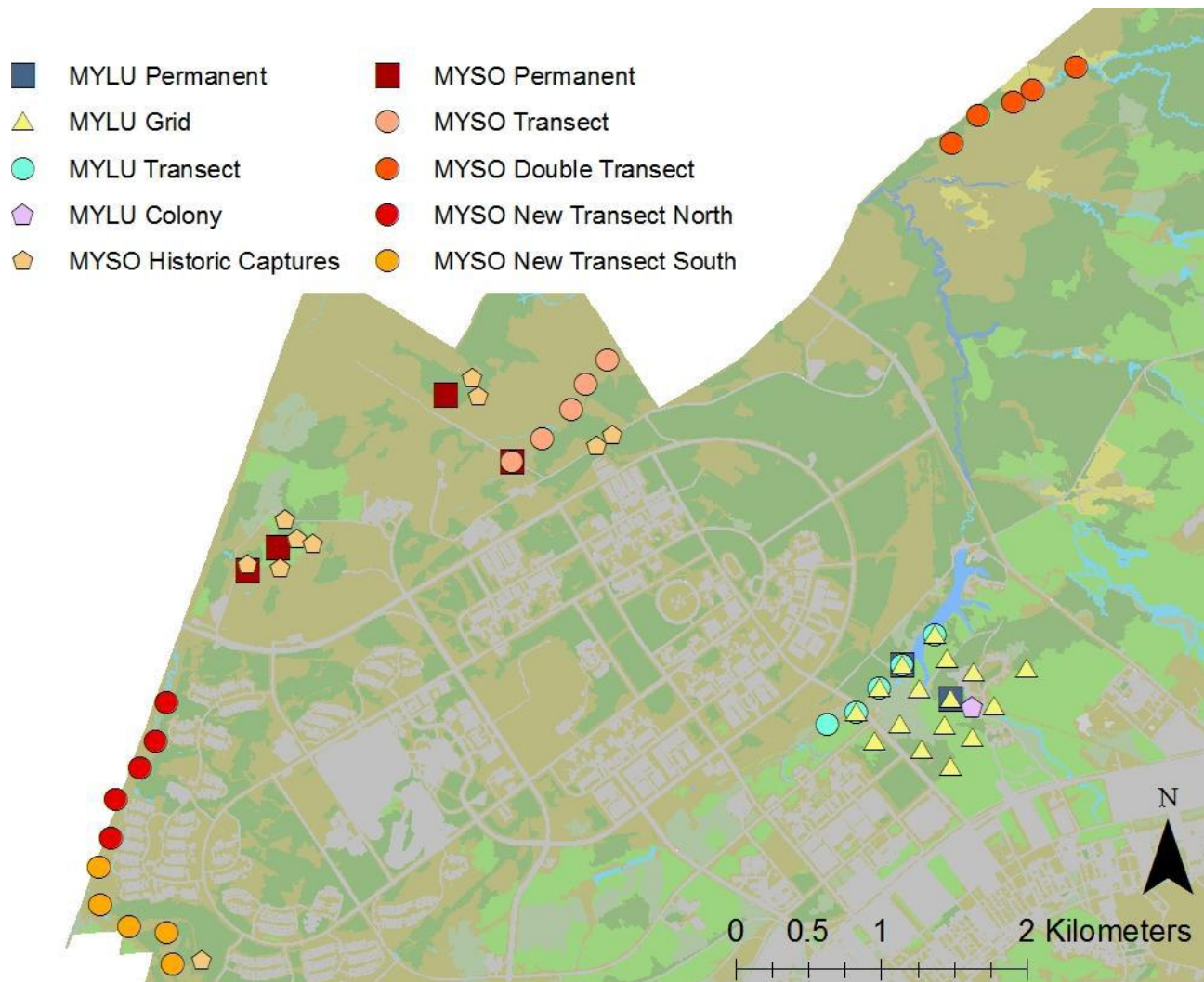


Figure 1. Passive acoustic sampling arrays in known maternity use areas of little brown bats (*Myotis lucifugus*; MYLU) and Indiana bats (*M. sodalis*; MYSO) at Fort Drum Military Installation Cantonment Area. Each point represents the site of a one passively deployed Anabat, except MYSO Double Transect which represents 2 Anabats per site. MYSO New Transect North and MYSO New Transect South deployed twice for 6-8 day sampling periods each in summer 2012. All other arrays sampled 4-5 times for 6-8 sampling periods in summers 2011 and 2012.

Information Criterion (AIC) corrected for small sample size and compared the weight of evidence among candidate models using Akaike weights (Burnham and Anderson, 2002). Following those analyses and retaining the significant covariates for the top approximating models, we collapsed detection histories so that each sampling array in its entirety was considered a single "site" for a given night. For example, if a species was detected at any of the 16 grid sites in a given night, it was considered present at that grid on that night. Because all sites in a particular sampling array were collapsed into a single representative site for the second set of models, we adjusted the covariate for habitat to a continuous variable representing the percent of wet sites in an array. We applied these covariates as a starting point for a single, additional model for each species to determine detection probability estimates among the varying sampling arrays. Included in the covariate set for sampling arrays were permanent, grid, transect and double transect sites. If the anticipated model failed to converge, we attempted to remove covariates from the previous candidate set in a step-wise fashion until the best possible model was reached to describe detection

probabilities based on sampling array. Finally, for WNS-impacted species, we calculated the effort required (in sampling nights) to determine true absence based on detection probabilities derived from occupancy models following Wintle et al. (2012).

RESULTS

We detected 9 species that occur at Fort Drum at least once during the 2011 and 2012 monitoring seasons. Two transects, "MYSO New Transect North" and "MYSO New Transect South" were only deployed once each in 2012. We removed one permanent site placed near the installation's artificial bat house from detection histories for each species because of high sound distortion caused by multiple bats being detected simultaneously. We were able to fit candidate models for all species to determine impacts of habitat, year and time of season on occupancy

Table 1. Candidate set of models for assessing impacts of habitat, year, or time of season on bat occupancy (ψ) and detection probability (p) estimates at Fort Drum Military Installation using passive acoustic sampling; “.” = constant, “habitat” = wet vs. dry sites, “year” = 2011 vs. 2012, “season” = 15 May-15 July/pre-volancy of young vs. 15 July-15 Aug/post-volancy of young.

| Model | ψ | p |
|-------|-------------------------|-------------------------|
| 1 | . | . |
| 2 | . | Habitat |
| 3 | . | Season |
| 4 | . | Year |
| 5 | . | Habitat + Season |
| 6 | . | Habitat + Year |
| 7 | . | Year + Season |
| 8 | . | Habitat + Season + Year |
| 9 | Habitat | Habitat |
| 10 | Season | Season |
| 11 | Year | Year |
| 12 | Habitat + Season | Habitat + Season |
| 13 | Habitat + Year | Habitat + Year |
| 14 | Season + Year | Season + Year |
| 15 | Habitat + Season + Year | Habitat + Season + Year |

and detection probability estimates (Table 2).

Placement of detectors at wet or dry habitats had an effect on occupancy estimates for silver-haired (*Lasionycteris noctivagans*), big brown (*Eptesicus fuscus*), and hoary bats (*Lasiurus cinereus*), with higher occupancy rates at wet versus dry sites (Table 2). Habitat also had an effect on eastern small-footed bat (*Myotis leibii*) occupancy, but the opposite trend was observed. The detection probabilities of all 9 species were greater at wet sites versus dry sites. Year had an effect on eastern red (*Lasiurus borealis*), hoary, little brown, Indiana, eastern small-footed and tri-colored bats (*Perimyotis subflavus*). Hoary and eastern small-footed bats showed slight increases in detection probabilities from 2011 to 2012 whereas the other species showed moderate to large declines from 2011 to 2012. Finally, time of season affected detection probabilities of eastern red, northern (*Myotis septentrionalis*), and eastern small-footed bats such that values increased in the late, post-volancy season when “new” bats are added to the landscape.

For array-specific detection histories, we created additional models to determine changes in detection probability based on a sampling array for 7 species (Table 3). We were unable to successfully model northern and eastern small-footed bats due to very low actual detections for these species. The percent-wet habitat covariate for the collapsed representative sites of permanent locations and the grid were 60 and 40%, respectively. Transects with single units and the double transect were all located at wet sites (100%).

Silver-haired bats were detected widely regardless of sampling array, year, or season. However, the detector

grids produced the highest detection probability estimates (Figure 2). Eastern red bats were also detected widely regardless of sampling array. The highest detection probability estimates were recorded at the grid, and an increase in detection probability was observed from early to late season. Hoary bats detection probability estimates were highest at the grid relative to other sampling arrays, though overall species’ detection probabilities were high regardless of array, year, or season. Big brown bats were detected at all sampling arrays, but showed the highest detection probability at the double transect regardless of year and season (Figure 2). Both little brown bats and Indiana bats were detected at higher levels at the grid relative to other array types. Finally, the tri-colored bat was detected at transects, the grid, and permanent stations in 2011 and 2012, however the species was not detected at the double transect in either year.

DISCUSSION

Even prior to the advent of WNS, occupancy modeling and detection probability estimates to assess bat species assemblages, relative activity, and habitat use with Anabat acoustic detectors had been gaining wider attention and use. Yates and Muzika (2006) reported an effect of year on Indiana bat detectability in the Ozark Mountain region of Missouri, pre-WNS. Similar to our within-year observations for eastern red bats, Hein et al. (2009) reported an increase in activity of eastern red bat over the sampling season, but also with little brown, and tri-colored bats in the South Carolina Coastal Plain, an area not been impacted by WNS at that time. Weller (2008) used

Table 2. Top (within 2.00 $\Delta AICc$) models for assessing impacts of habitat, year, or time of season on bat occupancy (Ψ) and detection probability (p) estimates at Fort Drum Military Installation using passive acoustic sampling at 40 sites; “.” = constant, “habitat” = wet vs. dry sites, “year” = 2011 vs. 2012, “season” = 15 May-15 July/pre-volancy of young vs. 15 July-15 Aug/post-volancy of young.

| Models | <i>K</i> | <i>AICc</i> | $\Delta AICc$ | <i>w_i</i> |
|---|----------|-------------|---------------|----------------------|
| Silver-haired | | | | |
| Ψ (habitat), p (habitat) | 4 | 1798.21 | 0.00 | 0.6480 |
| Eastern red | | | | |
| Ψ (.), p (habitat + year + season) | 5 | 2342.49 | 0.00 | 0.4967 |
| Ψ (.), p (habitat + season) | 4 | 2343.13 | 0.64 | 0.3607 |
| Hoary | | | | |
| Ψ (habitat), p (habitat) | 4 | 1990.24 | 0.00 | 0.3756 |
| Ψ (habitat + year), p (habitat + year) | 6 | 1990.48 | 0.24 | 0.3332 |
| Big brown | | | | |
| Ψ (habitat), p (habitat) | 4 | 1961.30 | 0.00 | 0.7888 |
| Little brown | | | | |
| Ψ (.), p (habitat + year) | 4 | 1028.32 | 0.00 | 0.6225 |
| Ψ (.), p (habitat + year + season) | 5 | 1029.83 | 1.51 | 0.2926 |
| Indiana bat | | | | |
| Ψ (.), p (habitat + year) | 4 | 998.16 | 0.00 | 0.6853 |
| Northern | | | | |
| Ψ (.), p (season) | 3 | 176.63 | 0.00 | 0.2318 |
| Ψ (.), p (.) | 2 | 177.26 | 0.63 | 0.1692 |
| Ψ (.), p (habitat + season) | 4 | 178.00 | 1.37 | 0.1169 |
| Ψ (.), p (habitat) | 3 | 178.57 | 1.94 | 0.0879 |
| Eastern small-footed | | | | |
| Ψ (.), p (year + season) | 4 | 84.92 | 0.00 | 0.2717 |
| Ψ (.), p (habitat + year + season) | 5 | 86.03 | 1.11 | 0.1560 |
| Ψ (habitat), p (habitat) | 4 | 86.60 | 1.68 | 0.1173 |
| Tri-colored | | | | |
| Ψ (habitat + year), p (habitat + year) | 6 | 191.16 | 0.00 | 0.3879 |
| Ψ (.), p (habitat + year) | 4 | 191.97 | 0.81 | 0.2587 |

occupancy modeling as a monitoring tool for assessing the effectiveness of a multiple-species conservation plan for bats in the Pacific Northwest. He found occupancy modeling that related species presence to habitat factors was most effective for common species but equivocal for rare species unless sampling effort is high. Those findings were consistent with ours for eastern small-footed and northern bats in that we were unable to model these species due to very low detection data.

Although previous research has focused on occupancy modeling for multiple bat species conservation and

management, no prior studies have assessed differences in multiple species detectability at various passive acoustic sampling arrays in the context of WNS-associated declines. Overall, detection probabilities were the highest for most species at the grid of detectors, regardless of year or time of the season. Each array type was represented by a unique value for percent-wet.

Therefore, our simple characterization of habitat did not have an impact on the probability of detecting a species at a particular array type, although it may have influenced the probability of detecting particular species at one array

Table 3. Occupancy (Ψ) models for determining impacts of passive acoustic sampling arrays on the detection probability (p) estimates of bat species at 40 sites at Fort Drum Military Installation, summers 2011 and 2012; “.” = constant, “array” = permanent, grid, transect, double transect, “habitat” = wet vs. dry sites, “year” = 2011 vs. 2012, “season” = 15 May-15 July/pre-volancy of young vs. 15 July-15 Aug/post-volancy of young.

| Bat species | Model |
|---------------|---|
| Silver-haired | Ψ (habitat), p (habitat + array) |
| Eastern red | Ψ (.), p (season + array) |
| Hoary | Ψ (.), p (array) |
| Big brown | Ψ (.), p (array) |
| Little brown | Ψ (.), p (habitat + year + array) |
| Indiana | Ψ (.), p (habitat + year + array) |
| Tri-colored | Ψ (habitat + year), p (year + array) |

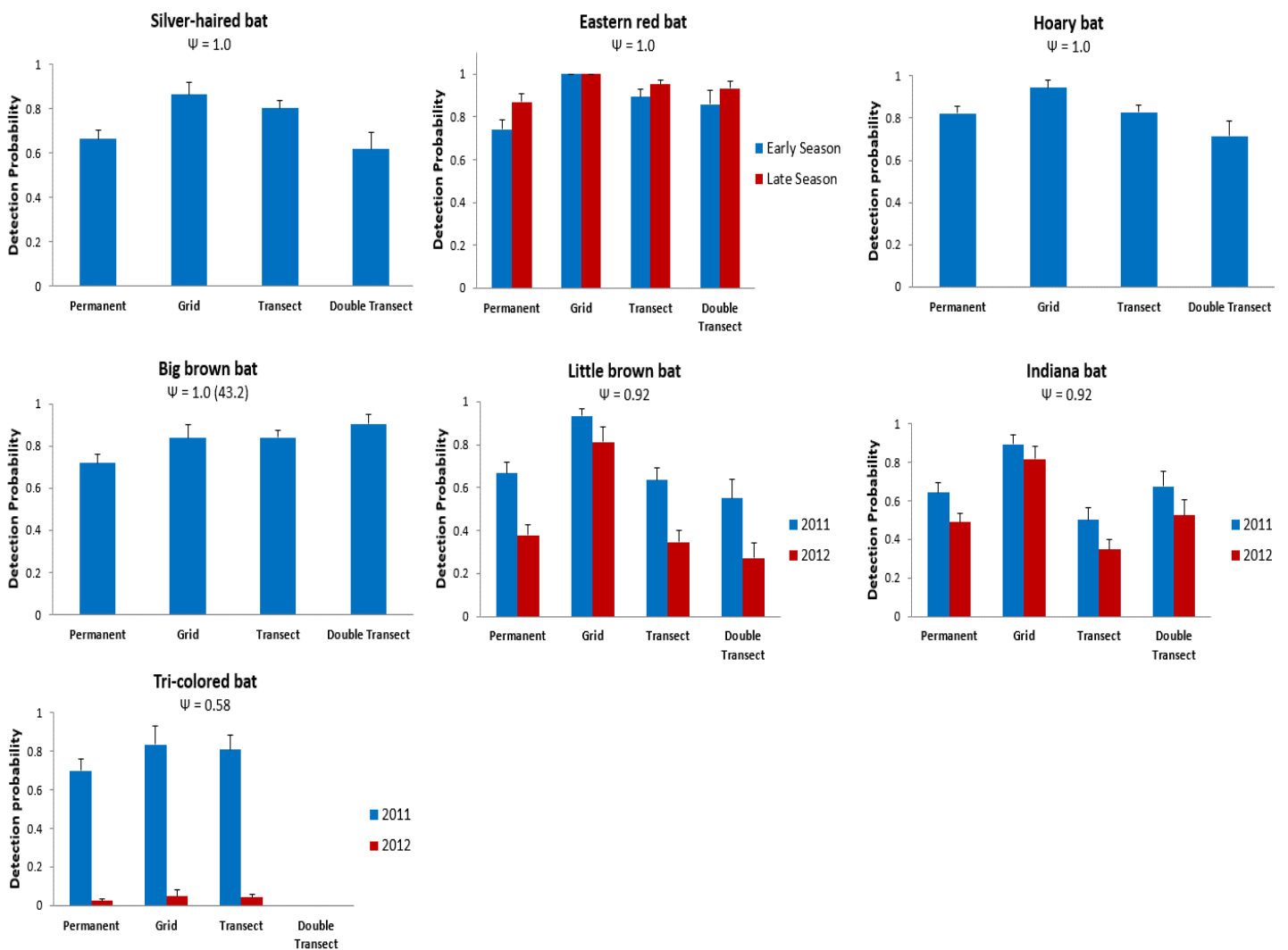


Figure 2. Detection probabilities of bats at various passive acoustic sampling arrays at Fort Drum Military Installation, summers 2011 and 2012. Time of season (early/late) and sampling year (2011/2012) included for important covariates affecting detection probability estimates from single species occupancy models; Ψ = proportion of sites occupied (naive estimate of occupancy).

type over another when included as a covariate. Indiana, little brown, and tri-colored bats were always detected at

the highest probabilities at the grid of detectors despite overall declines from 2011 to 2012. However, species not

impacted by WNS and the big brown bat were detected with high probabilities regardless of the sampling array, year, or time of season and should presently be expected to be detected under most sampling circumstances at Fort Drum.

Although the grid generally produced the highest detection probabilities, it is important to consider the varying levels of effort that were required for each array. Our grid placement required the highest number of available Anabat units relative to other arrays at 15 units, followed by the double transect requiring 10 units, and the permanent and transect sites that each required five units at a time. For many managers, limited fiscal resources may prohibit the use of large numbers of acoustical detectors simultaneously. In such cases, permanent stations or transects probably are the most feasible alternatives. However, it is important to note that MacKenzie and Royle (2005) suggested that the optimal strategy for detecting rare species is to focus effort at more sites at the expense of longer sampling duration at fewer sites. Relative to the endangered Hawaiian hoary bat, Gorresen et al. (2008) observed that reasonable precision of most parameter estimates of detection and occupancy were not achieved until 15 individual sites were sampled with acoustical detectors.

In our study, permanent sites did not detect little brown bats any better and only detected Indiana bats slightly better than did our transect sites. Permanent sites were never moved to different locations, whereas transect sites were placed repeatedly along four different streams at different time intervals. Of course, transect deployment and removal required much more effort than permanent stations, but batteries and storage cards did not need to be exchanged during recording cycles as occurred at permanent sites. More importantly, transects sampled a much greater area of the landscape. The flexibility in sampling a greater number of sites, with greater variation in habitat over a larger area in less time using transects probably makes this design the preferable method for reasonably high detectability for multiple species on most landscapes as compared to fixed units.

For focal monitoring of Indiana bats, benefits of long sampling durations and coverage over a wide area may both be necessary for optimally determining presence. As such, deploying detectors for greater than 6-8 days in transects along streams where the expectation of foraging use would be high probably is required. For example, based on the detection probabilities derived from our study, true absence of Indiana bats can be determined at the grid in 5-6 sampling nights or in 8-10 sampling nights using transects (Wintle et al., 2012). Similar patterns may be observed for determining probable absence of little brown bats. For tri-colored bats, the grid and the transect could both determine probable absence in 4-12 sampling nights relative to detectability of this species in 2011 and in 2012 using the grid, but a much longer sampling duration would be required based on detection estimates from transects in 2012 (Figure 3).

Although longer-duration transects may be necessary for detecting Indiana bats when equipment is limited, the double transect detected Indiana bats mostly as well as our grid design. Duchamp et al. (2006) suggested that having two detectors at a site increased the probability of detecting bat species in Indiana and Missouri, consistent with our findings for Indiana bats relative to other array types. However, for little brown bats, the double transect was no better than permanent stations or regular transects. This may indicate that although little brown bats are declining at Fort Drum (Ford et al., 2011), the spatial extent of their foraging may not be as limited or altered as other WNS-impacted species. For example, tri-colored bats were never detected at the double transect, despite easy detectability of this species at other arrays in 2011. However, even pre-WNS, tri-colored bats were scarcely observed in comparison to the once common little brown bat at Fort Drum, and it is uncertain whether they continue to occupy the landscape in stable numbers post-WNS.

As a species that was once found in colonies of hundreds to thousands of individuals in the summer across their range (Davis and Hitchcock, 1965) and at Fort Drum (Dobony et al., 2011), little brown bats are now infrequently observed in the Northeast, as many documented colonies have collapsed (Frick et al., 2010; Dzal et al., 2011; Turner et al., 2011). Indiana bats have also exhibited population declines regionally and locally as a result of WNS mortality (Ford et al., 2011; Ingersoll et al., 2013; USFWS, 2013) and were known to have a distribution restricted largely to the Cantonment area on the Fort Drum landscape even pre-WNS (Johnson et al., 2011a). Therefore, randomly selected locations are unlikely to be suitable for detecting these species at Fort Drum- and potentially elsewhere- when severe declines have been observed. Use of completely random sites may markedly increase survey effort required even over those shown here. Indiana bats and little brown bats were detected with high probabilities at all sampling arrays and at much higher detection rates than tri-colored, northern, and eastern small-footed bats, probably because efforts were focused on known historic maternity areas for these species in this study. On landscapes where historic capture information is not available, efforts can be focused at these species by focusing on areas with suitable habitat for these species or other target species.

Although the Indiana bat is the only species at Fort Drum that is presently subject to regulatory mandates of the Endangered Species Act (ESA 1973, as amended), northern bats have been proposed for endangered status by the USFWS (2013) and little brown bats are under review to assess their candidacy for federal listing as a result of WNS-associated declines (Kunz and Reichard, 2010). Furthermore, the tri-colored bat is believed to be one of the most severely impact species by WNS (Turner et al., 2011) and may also be subject to status reviews or listing proposals in the near future. With potential legal status changes probable, the ability to detect these bat

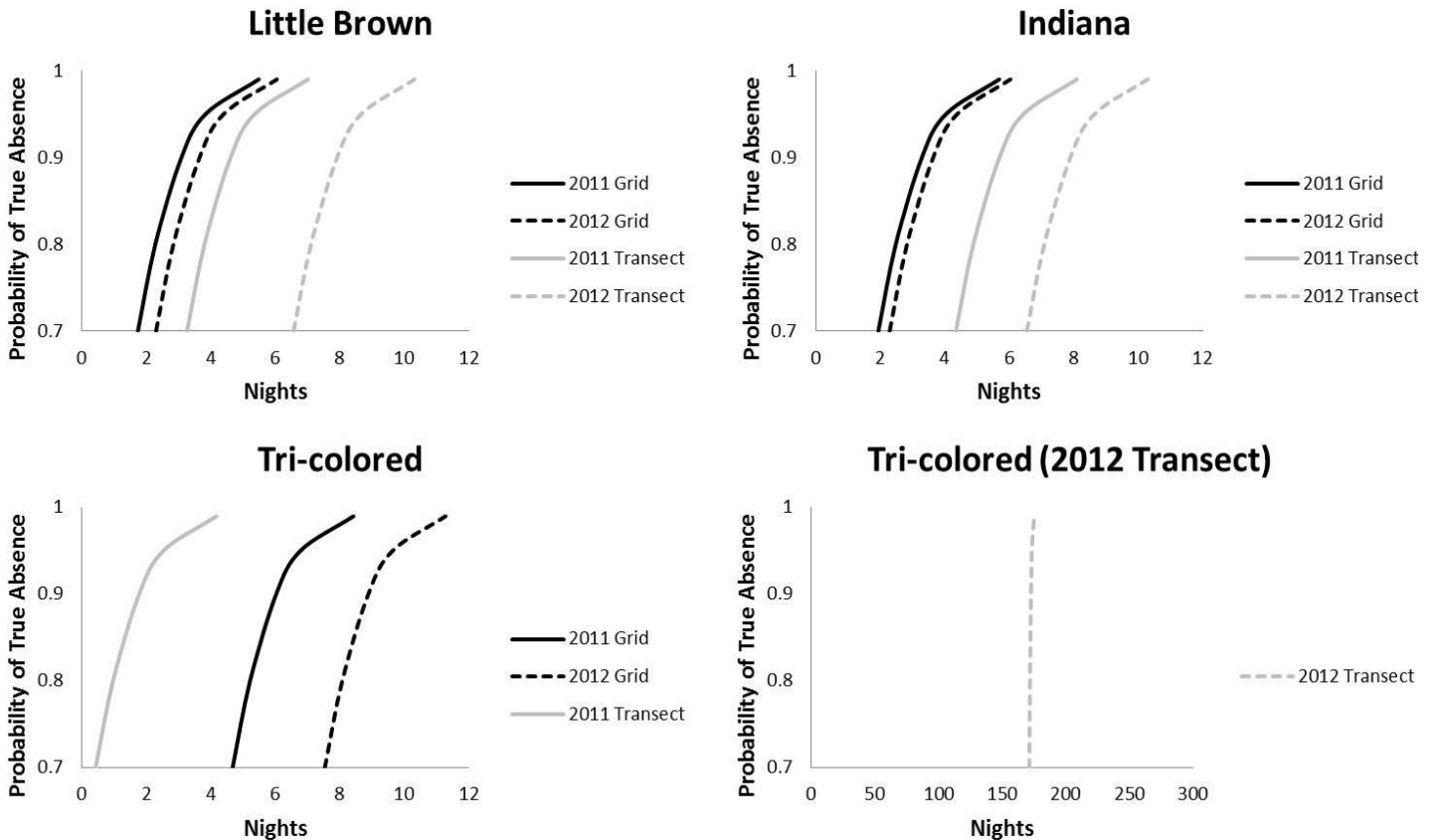


Figure 3. Suggested effort of passive acoustic sampling for determining true absence of Indiana (*M. sodalis*), little brown (*M. lucifugus*), and tri-colored (*Perimyotis subflavus*) bats using statistical probability according to the methods of Wintle et al. (2012). Detection probabilities derived from single species occupancy models at a grid of 15 Anabat detectors and four transects each of five Anabat detectors at Fort Drum Military Installation, New York, summers 2011 and 2012.

species when present will be critical from a regulatory perspective whereby managers can employ monitoring programs and mitigation activities to avoid or minimize potential take (ESA 1973, as amended). Following the suggestions of Weller (2008), future efforts to assess optimal sampling conditions for northern bats, eastern small-footed bats, and tri-colored bats are needed and perhaps should be focused on known historic areas of occupancy to determine if these species still occupy the Fort Drum landscape following WNS declines. For managers seeking an optimal strategy for determining presence or probable absence of Indiana bats or little brown bats, our study suggests that a grid of detectors in an expected area of use is most effective, possibly due to its inherent ability to survey a wider spatial area than the other methods presented here. However, in situations where high numbers of Anabats or other detectors are not available, the other sampling arrays presented here may be viable options for detecting the focal species in our study, that is, the Indiana and little brown bats, when placed in areas of anticipated use. However, deploying detectors across the widest area possible in areas of known previous use, expected use, or suitable habitat is

likely more effective than deploying detectors for very long periods at permanent stations or in random locations. Nonetheless, determining the best sampling design for additional WNS-impacted species will warrant further investigation, particularly for those species that may obtain federal listing in the near future.

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Full Length Research Paper

Endangering the endangered: Are protected areas save havens for threatened species in Cameroon? Case of Banyang-Mbo Wildlife Sanctuary, South Western Cameroon

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A hunting survey was conducted in the Banyang-Mbo Wildlife Sanctuary and support zones to estimate bushmeat off-take as a means to understand the current conservation status of protected species in that important area of biodiversity in Cameroon. A total of 756 protected animal carcasses with a total biomass of 6,815 kg, in six taxonomic groups constituted 24% of the total off-take of animals killed or captured by two adjacent ethnic groups of Banyang-Mbo Wildlife Sanctuary. Hunters caught more than 30 individuals of each of the red eared monkey (*Cercopithecus erythrotis camerunensis*), squirrel sp. (*Protexerus stangeri*, *Funisciurus pyrropus*), brush-tailed porcupine (*Atherurus africanus*), Water chevrotain (*Hyemoschus aquaticus*) African dwarf crocodile (*Osteolaemus tetraspis*), red river hog (*Potamoschus porcus*) and bay duiker (*Cephalophus dorsalis*) which together accounted for 75% of all protected species captures and 89% of the biomass. There was significant variation in the number of protected species exploited with the most captured taxonomic group, the rodents, comprising 37% of the kills or captures and 13% of the total biomass. The ungulates followed with 28% of the captures and 68% of total biomass. Proportionately, the much larger-bodied ungulates contributed more to biomass off-take than the comparatively smaller primates, reptiles and rodents. Carnivores and birds were least harvested taxonomic groups of protected species accounting for 9 and 4% of total protected species off-take respectively. Final results indicate that threatened animals do not have enough protection within BMWS and the conservation value of the sanctuary is highly compromised.

Key words: Threatened species, off-take, conservation status, Banyang-Mbo Wildlife Sanctuary, Cameroon.

INTRODUCTION

A protected area is defined as a geographical space, recognized, dedicated and managed, through legal or other effective means, to achieve the long-term conservation of nature with associated ecosystem services and cultural values (IUCN PACO, 2011a; Noss, 1998). Protected areas are essential for biodiversity conservation

and are the cornerstones of virtually all national and international conservation strategies. They are areas set aside to maintain functioning natural ecosystems, to act as refuges for species and to maintain ecological processes that cannot survive in most intensely managed landscapes and seascapes (Bennett and Robinson,

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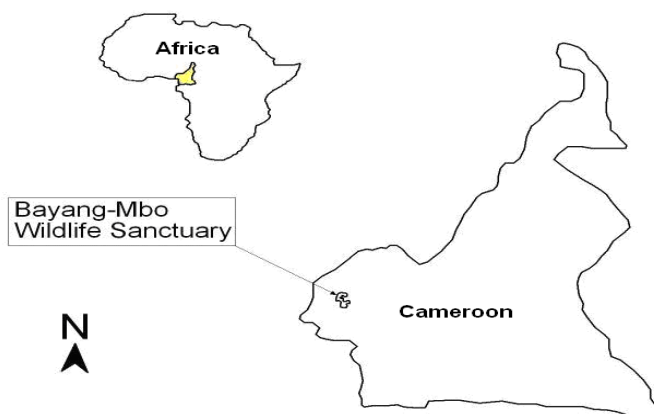


Figure 1. Maps of Africa and Cameroon showing location of BMWS adapted from Wilcox and Zouango (2000 unpublished).

2000; Wilkie et al., 1998b; Nasi et al., 2008; Slade et al., 1998). Protected areas act as benchmarks against which we understand human interactions with the natural world (Lahm, 1993; Cardillo et al., 2008; Wilkie et al., 1998a, b; Noss, 1998). Today, they are often the only hope we have of stopping many threatened or endangered species from becoming extinct (Robinson and Redford, 1991; Slade et al., 1998; Swanson and Barbier, 1992; Robinson and Bennett, 2000a). They are understood to be those areas in which human occupation or at least the exploitation of resources is limited (Cannon, 2003; Wilkie et al., 2005; Robinson and Bennett, 2000a, b).

Though protected areas are designated with the objective of conserving biodiversity and providing an indicator for that conservation's progress, the extent to which they defend resources and ecosystem dynamics from degradation are slightly more complex (Robinson and Redford, 1991; Bowen-Jones and Pendry, 1999; Fa and Yuste, 2001; Milner-Gulland and Akçakaya, 2001; Van Vliet and Nasi, 2008a; Ajonina et al., 2012 unpublished). Enforcing protected area boundaries is a costly and labour-intensive endeavour, particularly if the allocation of a new protected region places new restrictions on the use of resources by the native people which may lead to their subsequent displacement. This has troubled relationships between conservationists and rural communities in many protected regions the Banyang-Mbo Wildlife Sanctuary not being an exception in this regard and is often why many Wildlife Reserves and National Parks face the human threat of poaching for the illegal bushmeat or trophy trades, which are resorted to as an alternative form of subsistence (Hill et al., 2003; Ngandjui and Blanc, 2000; de Brooks et al., 2009; Wilkie et al., 2005; Foguekem et al., 2010; IUCN PACO, 2011a; IUCN, 2012). At the Banyang-Mbo Wildlife Sanctuary, large mammals have been surveyed and those of conservation interest include the leopard (*Panthera pardus*), African elephant (*Loxodonta africana*), giant pangolin (*Smutsia gigantea*), chimpanzee (*Pan troglodytes*), drill (*Mandrillus*

leucophaeus), water chevrotain (*Hyemoschus aquaticus*), forest buffalo (*Syncerus caffer nanus*), the dwarf, long-snouted and Nile crocodiles (*Osteolaemus tetraspis*, *Crocodylus cataphractus*, *Crocodylus niloticus*), and all tortoises (Chelonia) (Hart and Upoki, 1997; IUCN PACO, 2011a; MINFOF, 2012; Wilcox and Diangha, 2007; Ajonina, 2009). Hunting is one of the major causes of the decline of species of conservation concern in Afrotropical rainforest areas. The BMWS and its support zone have been listed as a center of primate endemism and one fifth of all African primate species live in this area. These species however, may be locally threatened by over hunting or continuous encroachment (WCS, 2012). According to law N°94/01 of 20th January 1994 establishing the regime for forests, wildlife and fisheries in Cameroon, class "A" are species with the highest degree of protection which includes endangered or extinct species as the black rhino, chimpanzee and the drill. The ministerial Order N°649/MINFOF of 18/12/2006 gives the repartition of wildlife species into groups and the extent of slaughter corresponding to each type of hunting license (MINFOF, 2012). Whereas class "B" are partially protected and may also not be killed without hunting license.

There is increasing and justifiable pressure to take proper account of human needs when setting up protected areas and these sometimes have to be "traded off" against conservation needs. Whereas in the past governments often made decisions about protected areas and informed local people afterwards, today the emphasis is shifting towards greater discussions with stakeholders and joint decisions about how such lands should be set aside and managed. Such negotiations are never easy but usually produce stronger and longer-lasting results for both conservation and people. The populations of many endangered species within protected areas in Cameroon are fast declining due to anthropogenic activities and accurate, current information on the status of these populations is essential for the design of effective conservation strategies within a human-dominated landscape.

Site description

The BMWS and the surrounding area covers an area of about 662 km², between 05°08' and 05°34' N and 09°30' and 09°47' E (Figure 1). It spans an altitudinal range of 120-1760 m. The mean annual rainfall is 4082.7±486 mm with heavy rains in July-September. The mean annual maximum temperature is 30.2°C and the minimum 23.7°C (Nchanji and Plumpre, 2001). The northern zone of the sanctuary is characterized by flat areas and gentle hills. The southern zone is hillier, with steep slopes and narrow ridges. A network of streams, some seasonal, and rivers that originate from the south, drain the forest (Wilcox and Zouango, 2000; Wilcox and Diangha, 2007; Ajonina, 2009).

The BMWS is located within the northwestern part of the Lower Guinea forest; this forest formation is one of the world's greatest lowland rainforest in terms of both

extent and biodiversity (Myers et al., 2000). According to reports by Philips and Miller (2002), the sanctuary possesses the highest plant diversity anywhere in Central Africa. A total of 543 species (trees, herbs, shrubs), representing 75 taxonomic families, have been recorded so far at this site. Of this total, 405 species of 53 taxonomic families are tree species with a diameter greater than 10 cm. This total includes 13 species, which have IUCN Red Data categories ascribed to them. Three plant species new to science and only known from the Sanctuary have recently been described (*Rothmania ebamutensis*, *Aulacocalyx mapiana* and *Tricalysia lejolyana* (Rubiaceae) (Sonke et al., 2002; Sonké, 2000).

The BMWS is also classified as an Important Bird Area (IBA) (Fotso et al., 2001). The avifauna is extremely rich with 325 species recorded and includes Mount Kupe Bush Shrike (*Telophorus kupeensis*), a rare endemic species at risk of extinction (Ajonina et al., 2012 unpublished; Tchamba and Elkan, 1995). This bird is only known from three contiguous sites: Banyang-Mbo, Mount Kupe, and the Bakossi mountains, of which the BMWS is the only area currently with a clearly defined protection status (IUCN PACO 2011a; IUCN 2012). Two of the six species that make up the "Cameroon Gabon Lowlands Endemic Bird Area (EBA)" have been recorded at this site (Rachel's Malimbe *Malimbus racheliae* and the grey-necked Picathartes *Picathartes oreas*) (Ajonina et al., 2012 unpublished). In addition, 11 of the 27 species that make up the Cameroon montane EBA have been recorded at this site and 20 of the 44 species characteristic of the Afrotropical Highlands biome that occur in Cameroon have been recorded in the Sanctuary.

The BMWS gazetted in 1996 was designed specifically to protect its populations of large mammals, particularly of forest elephant (*L. cyclotis*) where numbers recorded are higher than anywhere else in Western Cameroon (Blanc et al., 2003). The sanctuary is home to at least 38 species of large and medium-sized mammals (average adult body mass ≥ 1 kg), including the chimpanzee (*Pan troglodytes vellerosus*) and the drill (*Mandrillus leucophaeus*), both of which are endemic to SE Nigeria and SW Cameroon and are endangered species according to the IUCN criteria. The forest buffalo (*S. caffer nanus*), yellow-backed duiker (*Cephalophus sylvicultor*) and the water chevrotain (*Hyemoschus aquaticus*), all recorded as threatened species, are also present within and around the sanctuary (Table 1). However, it is feared that the leopard (*Panthera pardus*), and the giant pangolin (*Manis gigantean*), formerly known from the area may have already been locally extirpated through indiscriminate hunting (Wilcox and Zouango, 2000; Forbosoh et al., 2005; Wilcox and Diangha, 2007; Redford 1992, Ajonina, 2009).

Historically, there have been periods of significant human population within the BMWS area. These date back to the late iron-age period of the 9th to 18th Centuries AD, and evidence suggests that these settlements strongly

influenced the current landscape (Oslisy et al., 2000). Today, there are 54 villages situated within 10 km from the BMWS boundaries, with a total population estimate of about 50,000 people partitioned into four major ethnic groups; the Banyangi, Bakossi, Bassosi and Mbo (MINFOF, 2012). Like elsewhere in Cameroon, communities adjacent to the BMWS, rely on forest resources for their livelihoods. This dependence has led to the development of cultural values with strong affinities to the forest which, over many generations, have regulated access to essential natural resources. For example, the Banyangi ethnic group's *ekpé* traditional society which is based around a "leopard dance" is held in high esteem as the institution plays a cardinal role in conflict resolution (Noss, 1998; Bruner et al., 2001; Robinson and Bennett, 2004; Forbosoh et al., 2005 unpublished). The traditional costume of the dance group includes the skin of a leopard, but the catastrophic decline in the local leopard population obliged the institution to switch to genet pelt. Similarly, elephant parts, notably the tusk and tail, constitute an integral part of the traditional costume of many institutions in the Mbo area and beyond. Moreover, belief in "totem" animals, especially among the Mbo and Banyangi ethnic groups is rife. But faced with the challenges of globalization and the associated collapse of traditional social structures, respect for the forest and other natural resources has deteriorated, and unsustainable harvesting of forest products is spreading fast as people are struggling to meet their growing livelihood needs. Where there are no alternative income generating activities, people turn to the sale of bushmeat, as it is easy to preserve through drying and smoking and can easily be carried long distances to suitable markets. Indeed the hunting of wildlife remains a common livelihood activity to all ethnic groups and the decree conferring the legal status of the sanctuary recognizes and maintains the usufruct rights of the adjacent population, particularly the right to hunt wildlife species that are not covered by National and International legislation. Typically, bushmeat is sold in local weekly markets, principally to non-resident intermediaries from major consumption centers in the adjacent Littoral and West Provinces (WCS, 2011, 2012; Fitzgibbon et al., 2000). Occasionally, the urban intermediaries go to the villages few days before the market day and pile-up bushmeat to be transported on the market day, taking advantage of the availability of vehicle transport. At the urban centers, they sell the meat directly to consumers or restaurant operators. Ordinarily, the hunter deals directly with local residents before the market day.

MATERIALS AND METHODS

Data collection

The BMWS and its support zone have been listed as a center of primate endemism and one fifth of all African primate species live in this area. At the Banyang-Mbo Wildlife Sanctuary, large mammals have been surveyed and those of conservation interest include the leopard (*P. pardus*), African elephant (*L. africana*), giant pangolin

Table 1. List of species known to live in the Banyang-Mbo Wildlife Sanctuary.

| Common name | Scientific name | Legal status | Keyang name | Mbo name | Pidgin name |
|---------------------------|---------------------------------|--------------|--------------|-------------|---------------------|
| Primates | Primates | | Nstik | Nkem | Monkey |
| Greater white nose monkey | <i>Cercopithecus nictitan</i> | | Ekai | seeh | White nose |
| White -collared mangabey | <i>Cercocebus torquatus</i> | | Ekasso | Nkakum | Gendarme |
| Crown red guenon | <i>Cercopithecus pogonia</i> | | | Souboum | |
| Mona monkey | <i>Cercopithecus mona</i> | | Ngai | Pouing | |
| Red-eared monkey | <i>Cercopithecus erythrotis</i> | Protected | Nchwei | Sunkw uon | |
| Drill | <i>Papio leucophaeus</i> | Protected | Nsongnya | Sunkwuon | Sumbu |
| Chimpanzee | <i>Pan troglodytes</i> | Protected | Ekirikak | Mebange | Chimpanzee |
| Golden potto | <i>Arctocebus catabarensis</i> | Protected | Ebow | Medubambe | Bush baby |
| Allen's galago | <i>Galago alleni</i> | | Ebow | Oboah | Bush baby |
| Preuss'monkey | <i>Cercopithecus preussi</i> | Protected | | | |
| Artiodactyla | Artiodactyla | | | | Deer |
| Blue duiker | <i>Cephalophus monticola</i> | | Reteh | Mehsen | |
| Bay duiker | <i>Cephalophus dorsalis</i> | Protected | Ngukenow | Dembin | Frotambo |
| Yellow-black duiker | <i>Cephalophus sylvicultor</i> | Protected | Nkongho | Nzii | Sleeping deer |
| Ogilby's duiker | <i>Cephalophus ogilby</i> | | Nso | Mbin | Bush deer |
| Red river hog (Bush pig) | <i>Potamoschus porcus</i> | Protected | Njiwi | Ngwuo | Bush swine |
| Water chevrotain | <i>Hyemoschus aquaticus</i> | Protected | Seku | Sukum | Water beef |
| Bush buck | <i>Tragelaphus scriptus</i> | Protected | Mfon | Nya'a | Antelope |
| Forest buffalo | <i>Syncerus caffer nanus</i> | Protected | | | |
| Pholidota | Pholidota | | | | Catah beef |
| Tree pangolin | <i>Manis tricuspis</i> | | Njie | Saih | Catah beef |
| Long -tailed pangolin | <i>Manis tetradactyla</i> | | Njie | Saih | Catah beef |
| Giant pangolin | <i>Manis gigantean</i> | Protected | Njie Nsok | Saih nzo | |
| Rodentia | Rodentia | | | | |
| Brush- tailed porcupine | <i>Atherurus africana</i> | Protected | Nnyok | Gwuon | Tchuku- Tchuku beef |
| Cane rat | <i>Thryonomis swinderianus</i> | | Njuinok | Nzibi | Cutting grass |
| Giant rat | <i>Cricetomys gambianus</i> | | Ngumbok | Kwenyam | Grumbeef |
| Brown rat | <i>Crecetomys spp.</i> | | | Pou'oh | Brown rat |
| African giant squirrel | <i>Protexerus stangeri</i> | Protected | Mbarichkat | Mbouyam | Squirrel |
| Red footed squirrel | <i>Funisciurus pyrropus</i> | | | Mekwa'a | Squirrel |
| Fying squirrel | <i>Anomalurus derbianus</i> | | | ngwing | Fying squirrel |
| Carnivore | Carnivore | | | | |
| African palm Civet | <i>Nandinia binotata</i> | Protected | Mbay | Mbpeh | Bush pussi |
| African civet | <i>Viverra civetta</i> | Protected | Resem | Eswuah | Bush dog |
| Forest genet | <i>Genetta spp.</i> | Protected | | Meshing | Bush pussi |
| Dark mongoose | <i>Crossarchus obscurus</i> | | Sebioh | Mezuing | 20 in line |
| Marsh mongoose | <i>Atilax paludinosus</i> | | Mbak | Ebubuh | |
| Black-legged mongoose | <i>Bdeogale nigripes</i> | | Mbak | mbunyam | |
| Otter sp. | | | | Ebohlong | |
| Leopard | <i>Panthera pardus</i> | Protected | Nkwoh | Ngwo'o | |
| Hyracoidea | Hyracoidea | | | | |
| Tree hyrax | | | | kehnyam | |
| Reptiles | | | | | |
| Gabon viper | <i>Bitis gabonica</i> | | | Ejung | Viper |

Table 1. Contd.

| | | | | | |
|-------------------------|------------------------------------|-----------|--------|-----------|-------------|
| Cobra | <i>Naja spp.</i> | | Meri | Ehubi | Black snake |
| Green mamba | <i>Dendroaspis jamensoni</i> | | Mbam | Nkonwe'eh | Green snake |
| African rock python | <i>Python sebae</i> | Protected | Ngem | Ngem | Python |
| Nile monitor lizard | <i>Varanus niloticus</i> | | Remak | Nguanweh | Ngombe |
| Nile crocodile | <i>Crocodylus spp.</i> | Protected | Nyong | Ngan | Crocodile |
| African dwarf crocodile | <i>Osteolaemus tetraspis</i> | Protected | Ebu | Nkwbrin | Alligator |
| Tortoise sp. | | protected | Rewen | Kubah | Trokey |
| Birds | | | | | |
| Palm -nut Vulture | <i>Gypohierax angolensis</i> | Protected | | Jung | Eagle |
| Guinea fowl | <i>Gutera plumifera</i> | | Ehang | Kehngweh | Bush fowl |
| Black casqued hornbill | <i>Ceratogymna atrata</i> | Protected | Ngond | Nkwon | Hornbill |
| Parrots | <i>Parrots erythacus</i> | Protected | Euneng | Quing | parrot |
| Red king fisher | | | | Ugne'eh | king fisher |
| Francolin | Francolin | | | | |
| Green turaco | <i>Turaco persa</i> | Protected | | | |
| Chiroptera | | | | | |
| Bats sp. | Bats | | Waku | Njame | Blind bat |
| Proboscidea | | | | | |
| Elephant | <i>Loxodonta africana cyclotis</i> | Protected | Nsok | Nzo | Elephant |
| Hyracoidea | | | | | |
| Tree hyrax | <i>Dendrohyrax dorsalis</i> | Protected | | Kehnyam | Stone beef |
| Rock hyrax | <i>Procavia capensis</i> | | | | |

Adapted from WCS (1999/ 2000) and Ajonina et al. (2012).

(*S. gigantean*), chimpanzee (*P. troglodytes*), drill (*M. leucophaeus*), water chevrotain (*H. aquaticus*), forest buffalo (*S. caffer nanus*), the dwarf, long-snouted, and Nile crocodiles (*O. tetraspis*, *C. cataphractus*, *C. niloticus*), and all tortoises (Chelonia).

From November 2006 to June 2007, we recorded off-take and hunting practices in 10 of the 14 villages originally earmarked for the survey (Figure 2) located < 10 km from the BMWS. We also used a semi-structured questionnaire to interview 84 hunters about their hunting techniques, the species hunted, and the spatial and temporal distribution of their hunting activities (Appendix 1). Apprehension about probable policy implications of the research outcomes restricted the survey to the most receptive villages. Generally, the study was conducted in five Banyangi (Bara, Ebeagwa, Tinto 1 and 2, Tembang and Akribah) and five Mbo (Elumba, Etodi, Nzoa, Tangang and Ndom) villages, respectively.

Within each village we identified hunters through village council and negotiated a collective agreement on keeping all registered data anonymous and effectively motivating them to take all animal carcasses to specified locations for examination. Prior to the commencement of the survey, research assistants received intensive training on species identification, weighing carcasses and record keeping using the Kingdon (1997) nomenclature. Off-take was recorded for all 84 hunters in the study villages every other day. We documented: the hunting location (name of the camp or river); the time spent hunting; the hunting technique used; and the species hunted, including the sex, estimated age and type of forest in which the individual was killed or captured and information on the disposition of the carcass. The investigator regularly inspected the

data collected in order to ensure consistency and continuity. This information was used to generate data on: the proportion of gun and snare hunters, the proportion of protected species in harvested, trends of monthly harvest, proportion of game used for self-consumption or sold for income, the number of active hunters and the biomass extracted each month, and the preferred habitat types for hunting activities. A participatory mapping exercise was conducted in two of the most notorious hunting villages to map the hunting territory, including hunting trails and camps. Several field visits allowed us to reference the trails and camps using a Global Positioning System. The map was then used to position all hunting trips from the village. This allowed us to assess the spatial heterogeneity of hunting pressure in these villages by comparing hunting pressure at different distances from the village and calculating an index of hunting pressure for each habitat as the ratio of kills per habitat to the availability of each habitat:

$$I_h = (n_h/N)p_h$$

Where n_h is the number of kills in habitat h ; N is the total number of kills; and p_h is the proportion of each habitat within the hunted area, calculated using MapInfo 6.5.

We used two different approaches to assess the impact of hunting on mammal species. First, assuming that as the impact of hunting increases, mammal species are killed farther away from the village, we measured kills in relation to the distance from the village. The index of kills was calculated as the ratio of kills per protected species at various distance classes to the proportion of hunting trips

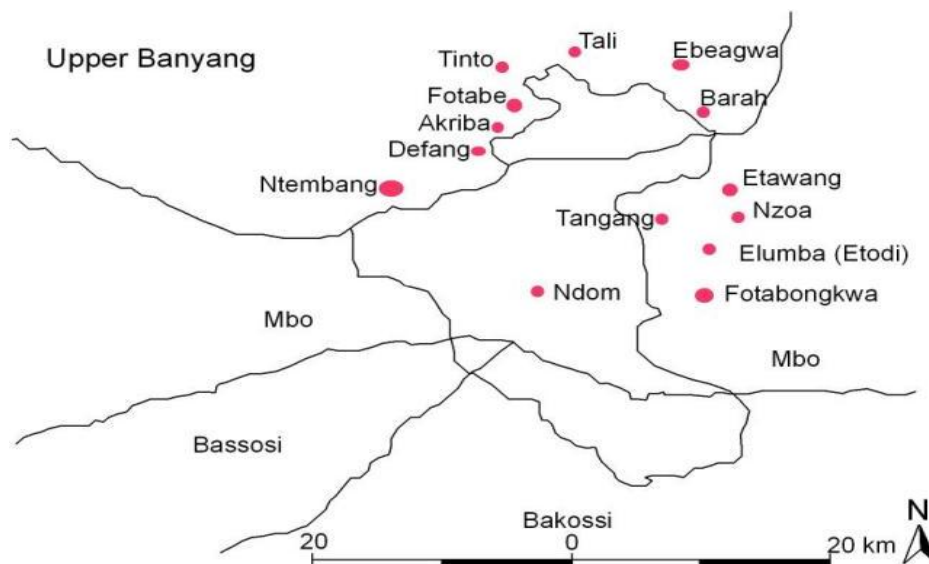


Figure 2. Study area within BMWS and adjacent villages.

that occurred at each distance class:

$$I_d = (n_d/N)/(t_d)$$

Data analysis

Qualitative interview data were analyzed using a textual approach focusing on words and meanings (Dey, 1993). Responses were thematically grouped and within each group the data were categorized. Qualitative categories were eventually quantified and input as nominal or ordinal data into SPSS version 17, along with quantitative demographic variables for each respondent. Basic frequencies were calculated for all categorical data and appropriate charts produced. Further analysis using cross tabulations and clustered bar graphs was conducted in an attempt to identify confounding variables. Continuous data were analyzed in SPSS or Microsoft Excel and descriptive statistics calculated. The nonparametric Kruskal-Wallis test was used to determine the significance of differences between villages and the Mann-Whitney U-test was used to investigate differences between hunters and trappers. Differences between nominal variables were tested using Chi square and the likelihood ratio (Field, 2005). Significance was set at $p < 0.05$. Associations between ranked and other scale variables were correlated using Spearman's correlation coefficient. One-sampled t-tests and ANOVA were used to analyze variance of means between datasets.

RESULTS

Off-take of protected species

During the eight month period of data collection in 2006/2007, a total of 756 protected animal carcasses with a total biomass of 6,815 kg, in six taxonomic groups constituted 24% of the total off-take of animals killed or captured by two adjacent ethnic groups of Banyang-Mbo Wildlife Sanctuary (Appendix 1). Hunters caught more than 30 individuals of each of the red eared monkey

(*Cercopithecus erythrotis camerunensis*), squirrel sp. (*Protexerus stangeri*, *Funisciurus pyrropus*), brush-tailed porcupine (*Atherurus africanus*), Water chevrotain (*Hyemoschus aquaticus*) African dwarf crocodile (*Osteolaemus tetraspis*), red river hog (*Potamoschus porcus*), and bay duiker (*Cephalophus dorsalis*) which together accounted for 75% of all captures and 89% of the biomass. There was significant variation in the number of protected species exploited with the most captured taxonomic group, the rodents, comprising 37% of the kills or captures and 13% of the total biomass. The artiodactyls followed with 28% of the captures and 68% of the biomass. Proportionately, the much larger-bodied artiodactyls contributed more to biomass off-take than the comparatively smaller primates, reptiles and rodents. The carnivores and birds were the least harvested taxonomic groups of protected species accounting for 9 and 4% of the total protected species off-take, respectively (Figure 2).

The red-eared monkey (*Cercopithecus erythrotis camerunensis*) accounted for 61% of all illegal primates off-take, the bay duiker (*Cephalophus dorsalis*) constituted 40% of artiodactyls harvest, the African dwarf crocodile (*Osteolaemus tetraspis*) 50% of reptiles, the African civet (*Civettictis civetta*) 41% of carnivores and the African grey parrots (*Psittacus erithacus*) 64% of illegally killed or captured birds.

Off-take distribution by biomass

Our results indicated that though ungulates constituted 28% of total off-take of protected species recorded killed, they constituted the highest percentage of off-take by biomass contributing 68% of the total biomass of threatened

Percentage off-take of protected species at BMWS

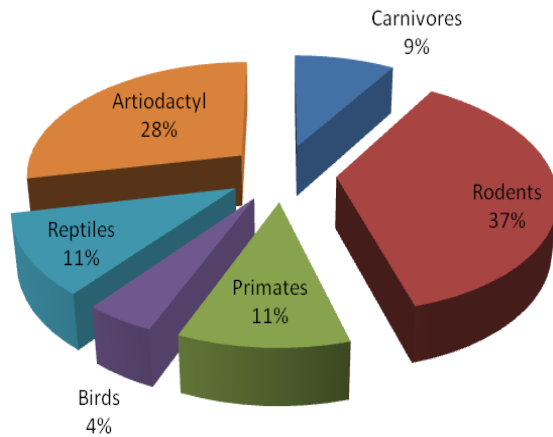


Figure 3. Percentage off-take of protected species at BMWS.

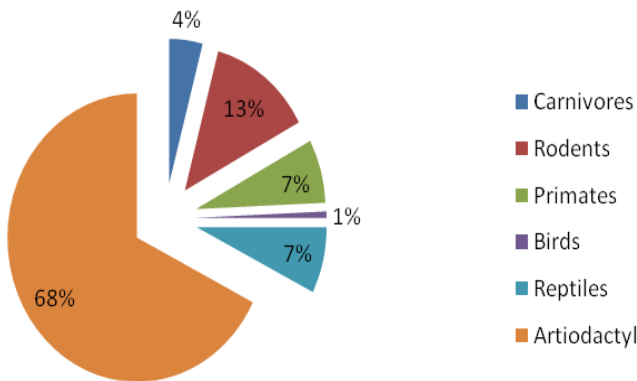


Figure 4. Biomass distribution of hunted protected species at BMWS.

animals killed. This was followed by rodents: constituting 13% of the biomass though had the highest number of off-take of protected species. The least contribution in terms of biomass came from the birds which constituted only 1% of total biomass off-take of protected species (Figure 3). Most of weight for ungulates came from the bay duiker.

Seasonality of off-take of protected species

Hunting occurs as an all-year-round activity although hunting pressures are more intense in the rainy season than the dry season (Figure 5). Peak harvest of most protected species during this study occurred in February and April. Generally, off-take was low in the month of January for all the species. Most hunters revealed that hunting at this time of the year was not very successful because the leaves are dry and animals can see far and

are always on alert when they hear any noise. It was gathered from hunters that peak harvest occurs in the months of July to October when rains are heavy. Generally, rodents constituted the most killed or capture protected species in all months of the study followed by artiodactyls whereas birds were the least killed or captured species.

Hunting methods

Generally, the gun and wire snare were weapons of choice for both the Banyangi and Mbo hunters during this study. Firearms (mostly locally fabricated shotguns) accounted for 61% of recorded killed primates and 53% of artiodactyls whereas wire snare accounted for 51% of carnivores kills or captures and 38% of reptile kills or captures. Most of the protected birds (39%) were captured or killed using other methods. Hunters used the two methods against all the major animal groups recorded. Indeed, we recorded no significant difference in the composition of captures by both methods. Adult animals accounted for at least 72% of cable snare captures of the three most hunted species (Figure 6).

Off-take by ethnic group

The Mbo ethnic group killed or captured primates 54%, reptiles 56%, rodents 60% and ungulates 55% in the illegal kills or capture of protected animals whereas the Banyangi hunters accounted for carnivores 59% and birds 55% in the total off-take of protected birds and carnivores (Figure 7). Generally, hunting is more intense in the Mbo section of the sanctuary where most people take hunting as a primary occupation.

Spatial distribution of hunting

The total hunting territory of BMWS was estimated at 15.5 km². However, hunting pressure was only exerted along hunting trails. Therefore, small and medium-sized territorial protected mammals were directly affected only if their territory overlapped one or more hunting trails. At < 10 km from the village, most families used own main and secondary trails however, at > 10 km from the village, trails and camps were used commonly by all hunters. A hunter changed hunting trails according to his or her perception of the decrease in the catch per unit effort and the scarcity of mammal signs. Trails that were located along rivers were only used during the dry season because of difficulty of access. Hunting pressure was not constant throughout the year. In July, August, September and October, > 20% of hunting activities was practiced at > 10 km from the village. During the rest of the year, hunting was mainly practiced close to the village. In February, March and April, at least 50% of hunting activities occurred at < 2 km from the village.

Also, hunting pressure differed among the forest types.

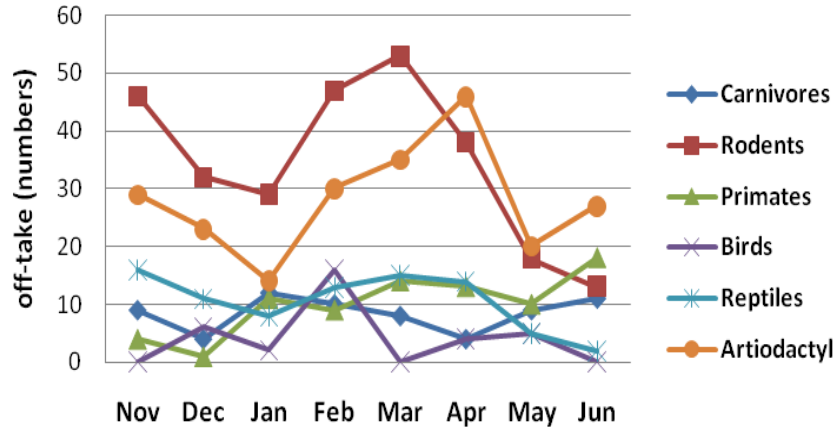


Figure 5. Monthly off-take of species by taxonomic groups in the BMWS and adjacent areas.

Off-take of protected species by weapon type

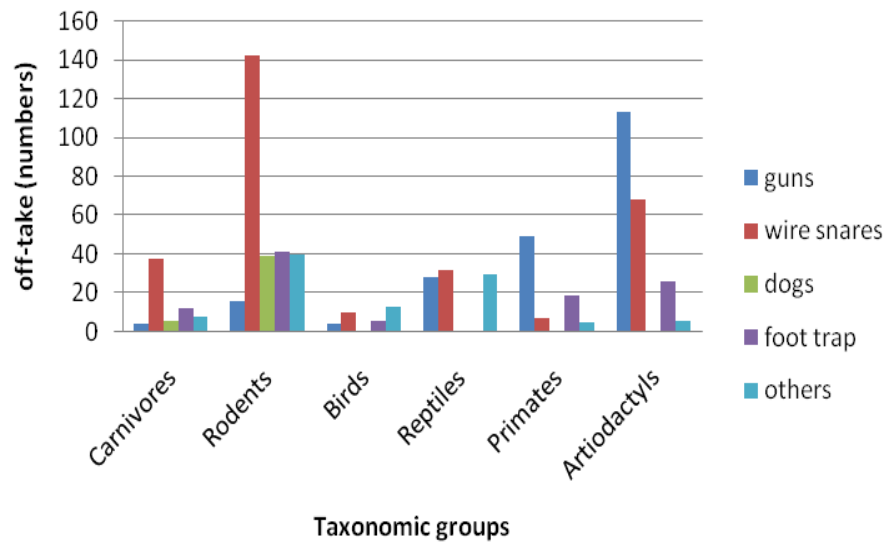


Figure 6. Susceptibility of taxonomic groups to weapon types.

Off-take records revealed that the primates particularly the red eared guenon (*Cercopithecus erythrotis*) dominated the primates' off-take in all the zones of hunting with most kills at distance more than 10 km. Protected birds, rodents, carnivores and reptiles kills or captures decreased steadily with distance from the village and the number decreased with distance from the village. Kills of ungulates increased with distance from the village. The index of kills (I_d) shows that rodents, carnivores, birds and reptiles were more likely killed at < 2 km and never at > 10 km from the villages (Figure 8). Primates were killed at all distances from the village and ungulates were more likely killed at 5 to 10 km and > 10km from the village.

DISCUSSION

Human responses to a species' perceived to be rare can have wide-ranging impacts on its population dynamics and the benefits arising from increased protection and habitat preservation are well documented. An eight month off-take study was conducted at the BMWS with the aim to understand the current status of protected species in that important area of biodiversity in Cameroon. Off-take reported amounted to a minimum of 1.14 protected animals per km² per year, which is much lower than the annual off-take from Monte Mitra, Equatoria Guinea (Fa and Yuste, 2001), Arabuko-Sokoke Forest, Kenya

Off-take of protected species by ethnic group

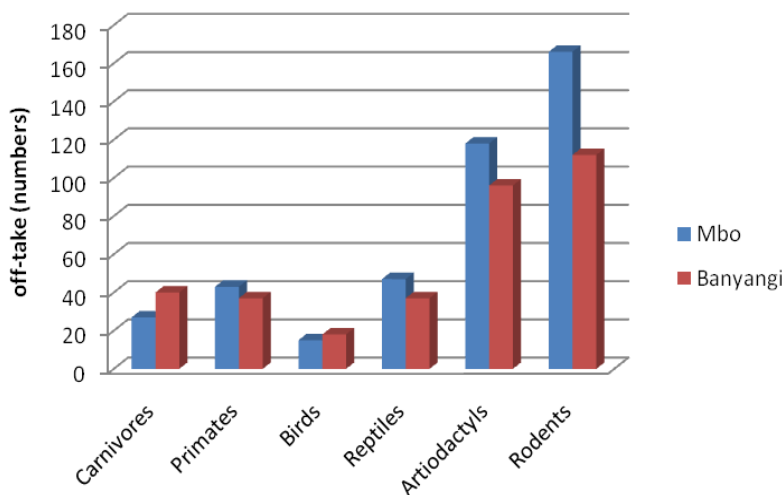


Figure 7. Distribution of animal off-take by ethnic group in BMWS area.

(Fitzgibbon et al., 2000) and the Central African Republic (Noss, 1998). The corresponding minimum biomass is 10 kg of bushmeat per km² per year, is consistent with results from the Monte Mitra, Equatorial Guinea, that show an extraction rate of 10 kg of bushmeat per km² per year (Fa and Yuste, 2001). With mean annual rainfall in excess of 4000 mm, the BMWS falls under the “wet forest” category of Holdridge (1967) cited in Forboseh et al., (2005) for which Robinson and Bennett (2000b) suggest a mammalian standing biomass of about 3,000 kg/km². Assuming that 10% of the standing biomass is available for human harvest as suggested by Robinson and Bennett (2004), it means that BMWS potentially could produce a maximum of 300 kg/km²/year. The paucity of data on biological productivity precludes formal assessment of the ecological sustainability at the BMWS. Moreover, the extraction rate of 10 kg/km²/year refers to off-take by 10 villages investigated, and therefore represents the minimum rate. If the nearly 50,000 people adjacent to the sanctuary were to depend exclusively on wildlife for meat it would require about 45000 kg of protected species per year following the US recommended daily amount of protein for a 70 kg man of 0.28 kg of meat per person per day (Robinson and Bennett, 2000b). Clearly, the annual requirement of the adjacent population alone far exceeds the theoretical maximum sustainable off-take of 198,600 kg per year from the 662 km² Sanctuary. Our comparison of previous (Forboseh et al., 2005 unpublished) and current off-take of protected species indicates that the proportion of birds remained stable, whereas the proportions of ungulates, rodents and primates are significantly higher in this study than in 2002 (ungulates: $\chi^2 = 20.116$, $p < 0.0001$; rodents $\chi^2 = 5.624$, p

= 0.009; primates: $\chi^2 = 6.28$, $p = 0.006$. Off-take of reptiles; $\chi^2 = 6.532$, $p = 0.01$) and carnivores; $\chi^2 = 14.6$, $p < 0.0001$) were significantly lower in this study than in the previous study.

There is growing consensus that even the most productive tropical forests cannot support much more than one person per km² where humans depend exclusively on wildlife for meat (Bennett and Robinson, 2000a). Evidently, off-take at the BMWS is highly unlikely to be socio-economically sustainable. These results point to the high degree of threat on the sanctuary wildlife especially protected species and the urgent need for more tangible management strategies required for its conservation. Already, the extraction pattern in and around the sanctuary is already indicative of declines in wildlife populations where the large to medium body-size animals are initially hunted to local extirpation or to such scarcity that small rodents account for most of the remnant animal off-take as is the case reported for the Amazon forest (Carrillo et al., 2000; Alvard et al., 1997; Maisels et al., 2001). Large and medium-sized ungulates, rodents and primates accounted for at least 75% of captures in the BMWS in 1999-2000 (Willcox and Nzouango, 2000). Between 1999 and 2003, blue duikers, the brush-tailed porcupine and red duikers consistently were the most captured species recorded in the area. In particular, duikers made up the majority of the catch in BMWS as in Liberia (Bruner et al., 2001; Buck et al., 2007), Equatorial Guinea (Fa and Yuste 2000; Brooks et al., 2009; Cardillo et al., 2008) and the Lobéké region of Cameroon (Fimbel et al., 2000). Consistent with models of optimal foraging, hunters prefer large-bodied animals that maximize the yield per unit effort. Indeed, large - bodied animals are

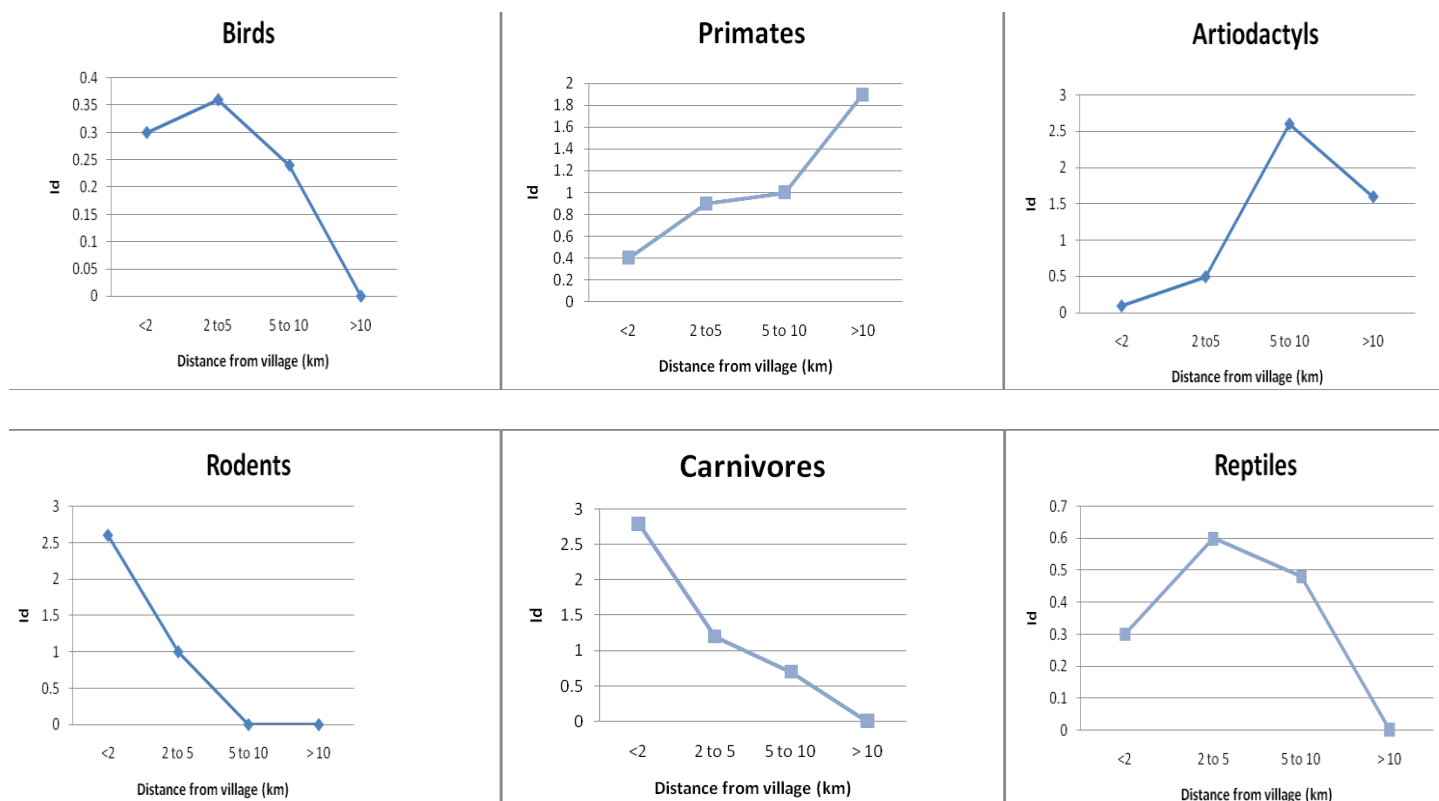


Figure 8. Index of kills (Id) at various classes of distance from a village for six species.

generally more profitable to hunters than smaller animals, though smaller sized species become vulnerable with increasing hunting pressure as found in this study and a similar study by Newing (2001). The preference for large-bodied mammals does not preclude hunting of small animals. Despite this, it is encouraging that the off-take of the most hunted species comprised a high proportion of adult animals, suggesting that the sanctuary still holds large populations of breeding adults. Maintaining a breeding population of adults is critically important for the long-term productivity of the sanctuary's wildlife.

The BMWS data amount to an off-take rate of at least 0.10 water chevrotains per km² per year and 0.001 chimpanzees per km² per year in this study is low; in a similar study, Forbosoh et al. (2005) reported off-take of 0.05 for water chevrotain and 0.02 for chimpanzee. The chimpanzee off-take rate is similar to that obtained in the Motaba region of north-east Congo (Kano and Asato, 1994), where it represents about 7% of the standing biomass and exceeds the annual population increase (Bowen-Jones and Pendry, 1999). There are no standing biomass figures of the BMWS mammals, but for all the species reported above, off-take is likely to be ecologically unsustainable. There is a general tendency of local hunters to under-report kills of protected species as evidenced by the absence of some threatened species like the elephants in the list of killed animals. Unlike in Monte Mitra, Equatorial Guinea (Fa and Yuste, 2001),

only a small proportion of captures in the BMWS enters the market. It seems that hunting for subsistence is more prevalent in the area, and bushmeat is the primary source of dietary protein.

Firearms and cable snares are the most preferred hunting weapons, collectively accounting for 92% of the 2009 recorded animal kills. In Cameroon, firearm owners are required to carry a permit, and a hunting license that clearly specifies species and season, if hunting is the intended use. However, the hunters identified as part of our survey possess neither permits nor licenses and by implication should not hunt. Similarly, the wire snare is outlawed throughout much of Africa, including Cameroon, because it is indiscriminate and wasteful, especially when irregularly inspected (Noss, 1998; Bruner et al., 2001; Robinson and Bennett, 2004). Clearly, our hunters are benefiting from an ambiguous and weak law enforcement regime. Because snare hunting was more common, hunters go hunting every 2 days probably to maximize kills and minimize losses. Similar observations were made by Lahm (1993), however, with the prominence of guns, the rhythm of hunting is very variable, with periods during which hunters go to the forest every day and long periods during which they are occupied by other activities. The widespread adoption of hunting with guns could have a serious implications for the nature of off-take, giving increased opportunities to hunt larger protected animals, mainly red river hog (mean weight: 40 kg) and bay duiker

(mean weight: 35 kg), and arboreal animals such as apes and small diurnal monkeys. In contrast, off-take of rodents such as African brush-tailed porcupine would decrease, most likely with the decrease in snare hunting. Based on our combined approach applied to different sites and contexts, we envision a revision of the Cameroonian forest, wildlife and fisheries law which currently lacks technical and social legitimacy. For example, the current law forbids hunting at night with guns and wire snare hunting. Therefore, 100% of the animals killed in this study are hunted illegally using illegal methods and in banned periods. Giving the dependence of local population on bushmeat as a source of protein and the quasi-technical impossibility to replace wild meat with domestic meat, such law that equate to blanket interdiction have no chance of implementation. The law is also flawed from an ecological and conservation viewpoint because species like brush tail porcupine and some small diurnal monkeys are partially protected, although they are known to be vulnerable and thus need specific hunting regulations for their protection.

Analysis of the seasonal data indicates that hunting is a year-round activity that necessitates a year-round anti-poaching campaign and other legal interventions with emphasis from July to October that stand out as peak hunting months. The protected red-eared monkey, African brush-tail porcupine and water chevrotain are vulnerable throughout the year, while the Cameroon-Nigeria chimpanzee appears most vulnerable between April-June, with the onset of the rains. All class A and B species cannot be hunted or trapped. However, this law is rarely enforced and the absence of organized government involvement has led to the presence of many of such animals in off-take during our survey and those of Wilcox and Zouango (2000), Forbosoh et al. (2005) and Wilcox and Diangha (2007). Bushmeat, including protected species have been reportedly sold openly throughout the country by hunters and middle men, in market stalls, and in restaurants. A law that is not enforced undermines the authority of the government, and a law that can only be enforced at great cost and difficulty might need to be revised. There is much work to be done in order to tackle this issue in most protected areas in Cameroon.

Our results show that hunting in villages adjacent to BMWS is practiced both for local consumption (60%) and for income (40%) to cover basic family expenses. In 2000 and 2007 Wilcox and Zouango and Wilcox and Diangha made similar observations whereby 70% of the villagers consumed their killed animals, sold 20% and gave out 10% as gifts. There is therefore no clear trend for a shift from subsistence to commercial hunting as has been demonstrated for other regions of Africa (de Merode et al., 2004).

Differences in hunting pressure in different habitat types are explained by hunters' preferences, as well as the spatial distribution of habitats and the ease of access. Despite hunters' preferences for mature forest, disturbed

forest had higher hunting pressure because this habitat surrounds the villages, whereas mature forest is only present at > 10 km from villages. Rivers are particularly used during the dry season, when water level is down. Habitat preferences by hunters have also been described for net hunters in Central African Republic (Noss, 1998). Degraded forest such as secondary regrowth supplied 47% of the animals killed and the greatest diversity of species at short distances from the villages. Mature forest supplied species with the greatest commercial value, e.g., red eared monkey, red river hog and bay duikers and is a convenient source of meat for traditional ceremonies. Therefore, the conservation of such undisturbed or lightly disturbed habitat is essential to meet local economic and cultural needs.

Our results indicate that the forests of BMWS are still able to offer large-bodied species to hunters at reasonable distances from villages. Despite off-take being below the MSY, bay duiker and drill were mostly killed at > 5 km from the villages. Van Vliet and Nasi (2007) also found that bay duiker is very vulnerable to hunting. Hunting pressure on small diurnal monkeys and red river hog has increased significantly in the last 20 years, and these species are now mainly found at >10 km from the villages.

Government's policy in conserving wildlife in Cameroon is to ensure that the mortality rate does not exceed the natural rate of increase through setting up conservation areas to safeguard most wildlife species. Law enforcement, though unpopular among the local communities adjacent this protected wildlife area, is one of the most effective means of controlling unsustainable use of wildlife resources. Due to lack of significant conservation and protection measures, many wildlife species including the drill and chimpanzee, still remain strongly threatened by combined effects of several factors such as: poaching, traffic in young animals, deforestation, fragmentation of the habitat and poverty. These illegal activities are aggravated by ineffective and inadequate protection of BMWS resulting from weak or inexistent management structures and law enforcement. Very little resources have been set aside by government for sanctuary security patrols and wildlife protection. Similar studies by Foguekem et al. (2010) and Omondi et al. (2008) indicate that lack of motivation, infrastructure and equipment further prevents efficiency. If the current trends continue, the long term viability of numerous wildlife species in this protected area may be seriously endangered. Without local effort, this sanctuary is sure to become so degraded that it will no longer require protection and will no longer be listed as a protected area.

A number of protected reptile species found in BMWS are consumed locally. These include the tortoises (*Kinixys erosa* and *Kinixys homeana*), the python (*Python sebae*), the forest cobra (*Naja melanoleuca*), and the green mamba (*Dendroaspis jamesoni*). Despite the efforts of WCS encouraging people not to hunt protected animals, villagers are still heavily exploiting some species.

This is especially the case for tortoises of the genus *Kinixy*; *K. erosa*, Serrated Hinge-back Tortoise and *K. homeana* Forest Hinge-back Tortoise that are listed as an Appendix I species under the Convention on International Trade in Endangered Species (CITES) and vulnerable (VU A2cd) in the IUCN Red Book, due to a suspected decline in population size caused by habitat loss and over exploitation a similar observation was made at the Waza National Park by (IUCN, 2012). The dependence of these species on primary forest, the late onset of sexual maturity and the low reproduction rate render them especially prone to overexploitation. Of the three species of African crocodiles, the dwarf crocodile, *Osteolaemus tetraspis*, was by far the most heavily hunted, a similar result was obtained at the Waza National Park in northern Cameroon (WCS, 2012). This preference is influenced by its small size and relatively non-aggressive nature, which facilitates easy capture, and further, it, stays alive while being transported to markets. Newing (2001) stated that the African dwarf crocodile is one of the most critically endangered crocodylians in the world. Many officially protected areas, in Cameroon like the BMWS where animals should be safe from depredation by humans, are not actually safe havens for those protected species. There is lack of protection inside and around BMWS and it was found that often more protected species are found per day in bush meat off-take than reported by Wilcox and Zuoango (2000). Conservation should be within the realms of politics, economics, ecology and social problems at a local level.

Currently, no accurate data are available on the status of protected species and habitat for this ecosystem. At present, the Banyang-Mbo Wildlife Sanctuary is seriously compromised, and of uncertain conservation value. However, the lowland sanctuary is still of high biodiversity value. The reptiles reported above are now regarded as rare throughout this ecoregion; however, there is insufficient data to adequately determine the population size of these protected reptiles within BMWS. Further study is clearly warranted to determine population size and threats to those populations. Once this information has been collected, a better picture of the ecological status of these animals can be determined, the effects of hunting and deforestation can be better assessed and with additional data, the implementation of species survival plans for these animals could be improved. The actual population status of most or all the threatened species within the BMWS is unknown, even though hundreds are captured annually (Ajonina, 2009 unpublished). A major institution, actively taking part in the conservation of endangered animals is Convention on International Trade in Endangered Species (CITES). The CITES laws have made the most strict regulations involving the international transport of animals. In the case of the chimpanzee, gorilla, African elephant tusk, African dwarf crocodile, etc, merely the regulation on international transportation is not enough.

Conclusion and recommendation

Cameroon faces major conservation challenges and despite the country's natural riches, several wildlife species are threatened in protected areas within the country. Our results indicate that protected species at Banyang-Mbo Wildlife Sanctuary are further threatened with high population declines due to human population growth, illegal hunting and habitat destruction which have negatively affected the protected species and other wildlife in the Banyang-Mbo wildlife sanctuary.

Despite this findings there is need to further research on status surveys, the identification and protection of important populations and habitat; the enhancement of the conservation and management capacities of national authorities; the development of national management plans for endangered species conservation; captive breeding and restocking programs and the development of economic incentives for protected species conservation through well-regulated sustainable use in protected areas in Cameroon.

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Appendix 1. Questions administered to hunters during study.

1. What really made you to start hunting?
2. How many times per week do you go for hunting?
3. Are you a specialized hunter?
4. Do you hunt for your own consumption or to earn income? If both, which is the most important motivation for hunting?
5. Do you hunt on a year round basis? If no, are there any periods during the year when you particularly hunt? If yes why?
6. Which season is most suitable for hunting and why?
7. Do you go hunting alone or in hunting groups and why?
8. Can a hunter check or remove a dead animal from another hunter's trap if it is getting rotten?
9. Are there periods when hunting is easier? why?
10. How far from the village do you usually go hunting (a) < 2 km, (b) 2 to 5 km, (c) between 5 and 10 km, (d) >10 km any reason for your answer?
11. Which weapon type do you use most for hunting and why?
12. In which forest type do you use your technique? What types of animals are killed most?
13. If snare hunting (a) how many snares do you currently set?
(b) How do you choose where and when to place snares? In what type of forest?
(c) At what time of the day do you set snares? How often do you return to check the snares?
(d) How long does a snare line stay in place? What makes you decide to change the location of your snare line?
(e) How much does a snare cable cost?
(f) Imagine that you set 100 snares. How many would have caught an animal by the first day that you returned to check them?
14. If hunting with a gun (a) was it easy to buy a gun? Where? How much?
(b) Is it easy to find cartridges? Where? How much?
(c) Can you estimate how many missed shots you have on each hunting expedition?
(d) What species are easiest to shoot?
(e) Do you use calls to attract animals? When? Where? What animals?
15. Do you understand the meaning of protected animals?
16. Can you identify a protected animal during hunting in the forest?
17. How many times have you been confronted by a wildlife officer for killing carrying a certain species of animal?
18. Does the presence of the Ministry of MINFOF in your village have any impact on your hunting activities?

Full Length Research Paper

Overhead shading and growth of young longleaf pine

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A study to determine the effects of environmental conditions on the growth of longleaf pine (*Pinus palustris* Mill.) was initiated in 1969 on the Escambia Experimental Forest near Brewton, Alabama, USA. This study sample consisted of forty young naturally regenerated, even aged longleaf pine seedlings evenly divided between two soil types. At the beginning of the study, the seedlings were 14 years from seed and ranged in height from 0.8 to 1.5 m. From 1969 to 1970, height and diameter measurements were recorded once to four times weekly during the growing seasons and once a month during the dormant seasons. To test the effects of shading on growth, cheesecloth was suspended over 10 randomly selected seedlings from each soil type only during the first growing season, from March 28 to September 24, 1969. This study provides data from the only known in-field shading experiment with longleaf pine seedlings of this size. The effects of the shading treatment and soil type were evaluated for height and diameter growth. The shading treatment did not have a significant effect on either height or diameter growth, but soil type had a significant effect on diameter growth.

Key words: *Pinus palustris*, longleaf pine, shade, soil type, shade intolerance.

INTRODUCTION

Trees, like all terrestrial plants, are dependent on sunlight, carbon dioxide, oxygen, water, and nutrients to survive and grow. Growth on the most basic scale can be defined as an irreversible increase in the size or number of cells (Kramer and Boyer, 1995). Growth and maintenance are the main purposes of a tree's metabolism and essential to a tree's survival (Kramer and Boyer, 1995). However, growth is not achieved without resistance from environmental stresses. Changes in the environment result in changes in a tree's internal physiological processes like photosynthesis, respiration, and absorption of water and minerals, which in turn affect growth (Kozlowski and Pallardy, 1997). At the genetic level, these changes or stresses in the environment can cause plants to modify morphology and physiology over time through phenotypic plasticity, which has many implications for forest management (Bradshaw, 1965;

Schlichting, 1986; Chambel et al., 2005). The desire to identify and understand the many relationships that exist between the environment and tree growth has influenced numerous research efforts, but often the answers found lead to more questions.

The first European settlers in what is now the southeastern United States were confronted with an upland forest that was dominated by this one tree species—longleaf pine (*Pinus palustris* Mill.). Stretching from the coastal plains of Virginia across a broad belt of the South Atlantic and Gulf Coasts into eastern Texas, longleaf historically occurred on over 36 million hectares and is thought to have been predominant on over 24 million hectares, but the ecosystem now only occupies fragments of the range (Frost, 1993). From southeastern Virginia to eastern Texas, it historically dominated the Coastal Plain but also extended into the Cumberland Plateau,

Ridge and Valley, Blue Ridge, and Piedmont physiographic provinces (Boyer 1990). Throughout the literature, longleaf pine is referred to as an intolerant species (Schwarz, 1907; Wahlenberg, 1946; Boyer, 1990). From observations of virgin stands of longleaf pine, Schwarz (1907) believed that light was the most critical factor in the development of longleaf pine regeneration. He believed light determined not only the time and place, but also the manner in which succession occurred. The light required for a new generation of trees was admitted through openings in the crown cover. Small openings were caused by the fall of one or several large trees as a result of old age, disease, lightning, insects or windstorms. Larger openings were similarly caused by hurricanes and severe storms. Schwarz (1907) noted smaller openings in the forest would be filled by a dense grove of young trees, locally known as "sapling thickets." If the opening did not fill with longleaf pine then oaks and other species would take over. Longleaf pine has a grass-stage, a unique growth phase where there is no above-ground height growth, but the seedling is putting on root and ground-line diameter growth (Chapman, 1932; Wahlenberg, 1946; Brown, 1964; Boyer, 1990). Seedlings will remain in this grass stage until a sufficient root system has developed, and height growth is initiated (Wahlenberg, 1946; Boyer, 1990; Outcalt, 2000). With the commencement of height growth, a seedling can grow about 30.5 to 100 cm a year (Wahlenberg, 1946). Long-leaf pine's adaptations as a climax species to fire like the grass stage, timing of height growth initiation, needle architecture, and branching structure makes it unique among the other southern pines and tree species around the world (Chapman, 1932).

Researchers in the 1930's began to look at how the growth of longleaf pine was related to environmental variations. Lodewick (1930) found positive correlations between diameter growth and precipitation. Diameter growth was found to be positively correlated with precipitation and negatively related to temperature (Coile 1936). Pessin (1938) using field studies showed tree stocking levels and ground cover plants had negative effects on seedling growth and noted the importance of light intensity for longleaf pine seedlings. Niinemets and Valladares (2006) found an inverse relationship between tolerances for shade and drought for a range of species, which shows tolerances can change depending on the existing factors. Manipulating light availability is very important since longleaf pine is commonly considered shade intolerant (Schwarz, 1907; Wahlenberg, 1946; Boyer, 1990). McGuire et al. (2001) found a positive correlation between the amount of light available and longleaf pine seedling growth in an in-field experiment with seedlings that had not initiated height growth. Barnett (1989) found significant differences between the shading treatments and a full sunlight control in a nursery setting. However, he did not see significant differences between the 30 and 50% shading treatments. Palik et al. (1997) found an upward curvilinear relationship between the growth rates of the selected trees and the variations in nitrogen and sunlight levels

from 40 to 80% of full sunlight. Jose et al. (2003) evaluated the effects of light, water, and nitrogen on grass stage longleaf pine seedling growth. They concluded that seedling growth in a greenhouse with a shade treatment seemed to be more influenced by water and nitrogen than light, but they did find a significant response in root and stem biomass with interactions between light and soil resources (Jose et al., 2003). Climent et al. (2006) also found significant effects in biomass with reductions in light for Canary Island pine (*Pinus canariensis*) seedlings. They also found that light and water effects were significant and created an interaction where shoot elongation almost stopped. Variations might be related to the in-field conditions in comparison with controlled greenhouse studies, but variations might also be related to an age and growing conditions of microsites. Cavender-Bares and Bazzaz (2000) found environmental components and ontogenetic components played an equal role in photosynthetic capacity if water was not a limiting factor between red oak (*Quercus rubra*) seedlings and mature trees.

In 1969, the U.S. Forest Service, studied a young longleaf pine stand in southern Alabama, USA to explore the relationships between the growth of young longleaf pine that had initiated height growth and the surrounding environment including an in-field shading treatment. This study was a follow-up to work done with loblolly pine in North Carolina (Boyer, 1970, 1976). Due to time and money constraints, the data were never analyzed until work began on it in 2003 as part of a master's thesis (Gilbert, 2007). This article focuses on the study of the in-field shading treatment.

METHODS

This project was installed on the Escambia Experimental Forest near Brewton, Alabama, USA in 1969. Data from a weather station located near the study site showed average precipitation which was 176.78 cm in 1969 and 195.83 cm in 1970. The average maximum temperature was 25°C and the average minimum temperature was 11°C for both years. The study sample consisted of young naturally regenerated, even aged longleaf pines, which were a product of the 1955 seed crop. All of the overstory had been removed prior to the study, which eliminated all overhead competition. Forty longleaf pine seedlings ranging from 0.8 to 1.5 m in height were selected for the study. Twenty were selected on each of the 2 distinct soil types that were present in the stand.

The two soil types that separated the seedlings were Lucy loamy sand (Lucy site) and Wagram loamy sand (Wagram site). The taxonomic class for both soils is: loamy, kaolinitic, thermic Arenic Kandiodults (Soil Survey Division, NRCS, 2003). The seedlings on the Lucy site were located on the crest of a ridge, and the seedlings on the Wagram site were located on a slope at the base of the ridge. Mattox et al. (1975) reported soil composition, average site index and depths of the A-horizon for both soil types. The soils are very similar with an average site index of 20.4 m (base age 50). The depth of the A-horizon for the Lucy soil varies from 55.88 to 101.6 cm, while the depth of the A-horizon varies from 50.8 to 68.58 cm for Wagram soils. The Lucy loamy sand also has higher clay content at shallower depths than the Wagram loamy sand. Soil samples were taken for this study in coordination with the collection of soil moisture at neutron probe tubes across the study site.

Shading treatment

Half of the seedlings in each of the two soil types were randomly selected for artificial shading. These seedlings were shaded with cheesecloth for six months during the growing season of the first year of the study. The shading treatment was installed on March 28, 1969 and removed September 24, 1969. The cheesecloth was stretched across a one-meter-square frame that was structured to keep the cheesecloth at least one meter above the growing tip of the seedling. The structures were periodically checked, adjusted, and maintained at one meter above the growing tip. The cheese cloth prevented the growing tips of the shaded seedlings from receiving direct sunlight during the peak of the diurnal cycle. A pyrhelograph was used to measure solar radiation in langley's (cal/cm^2) for both shaded and unshaded areas. The mean percent difference in solar radiation between non-shaded and shaded seedlings was 38.5%.

Growth measurements

Initial heights were measured from the ground level to the base of the bud on the terminal growing shoot of each seedling on January 27, 1969. The length of the terminal bud was also measured. Separate records were maintained for each new leader. Terminal shoots were measured from 2 to 4 times weekly from March to October of 1969 and 1970. During the dormant season of both years, heights were measured every two weeks or at least once a month.

Monthly growth intervals covered 22 months from March 1969 through December 1970. Each monthly interval was 28 ± 1 day. The biweekly intervals included 22 measurements in 1969 ranging from March to December and 24 measurements in 1970 ranging from January to December. Biweekly growth intervals were 14 ± 1 day in length. Weekly intervals included 34 measurements in 1969 ranging from March to October and 38 measurements in 1970 ranging from March to November. The weekly intervals were 7 ± 1 day in length.

Diameters of each seedling were measured in centimeters at 10 cm above the ground line with the use of dendrometer bands. The 10 cm height was set because all the seedlings had not reached dbh (diameter at breast height of 1.37 m). Diameter measurements were taken weekly from March to October of 1969 and 1970. From the end of October to March, measurements were taken every two weeks to a month during 1969 and 1970. These measurements can again be divided into monthly, biweekly, and weekly growth intervals using the same interval lengths and durations as outlined for height growth.

Statistical procedures

Statistical procedures were executed in Statistical Analysis System (SAS) software version 9.1 (SAS, 2003). All analyses were conducted at the 0.05 level of significance. Analyses of variances were conducted to test for the effects of shade treatments and location over various growth rates using PROC GLM and PROC MIXED in SAS (SAS 2003). PROC MIXED was used to conduct a repeated measures analysis using seedling as the random factor (SAS, 2003). The repeated measures analyses were conducted with the measurements from each interval and with consecutive growth from the initial measurement. PROC MIXED was chosen because it allows for missing data and an unbalanced data structure (Littell et al., 1996). Interactions were tested further using Tukey's Studentized Range (HSD) and least significant difference (LSD) multiple comparison procedures and within group t-tests (Ramsey and Schafer, 2002; SAS, 2003).

All statistical tests were evaluated to confirm any potential violations from assumptions. To evaluate normality of residuals, a normal probability plot or a normal QQ plot was used from PROC UNIVARIATE (Neter et al., 1996; Ramsey and Schafer, 2002; SAS, 2000-2004). The Anderson-Darling test from PROC UNIVARIATE was also used to determine normality of the residuals (SAS, 2000-2004). Results from the QQ plot and from PROC UNIVARIATE were compared to determine if normality was suspected. Unless otherwise stated, the assumptions were not violated.

RESULTS

Height growth: 1969 to 1970

Evaluating differences in height growth over the different intervals is necessary to see if the recorded environmental conditions and treatments affect height growth. Each interval (monthly, biweekly, and weekly) covers different portions of the growing seasons. Patterns of growth over the growing seasons show that seedlings do not grow at the same rates during the entire growing season. Figure 1 displays average monthly height measurements illustrating how patterns of growth changed over the study period. Growth rate tends to vary over the growing season which could raise questions about how relationships between growth and the environment change over the same period. Figure 2 displays initial height measurements. Initial height measurements ranged from 0.8 to 1.5 m. The measurements were tested to look for any significant differences before the treatments were applied. Initial heights were not significantly different for site, shading treatments, or an interaction for the site and shading treatment combinations.

Table 1 shows mean height growth for 1969 and 1970 individually. Overall height growth was first tested over the entire monthly interval. There were no significant differences between shade treatments or site location, but there was a significant interaction between the site and shade variables. Within group comparisons were then used to evaluate the significant interaction. Monthly height growth was also explored for potential significant differences using the repeated measurements design. Only time related variables and the shade/site interaction were significantly different. Testing the biweekly interval for height growth resulted in no significant differences between shade treatments or site location, but the shade/site interaction was significant as for monthly intervals. Biweekly intervals were also tested using repeated measures. Again, only time related variables and the shade/site interaction were significantly different. Weekly intervals were tested for both 1969 and 1970 because the measurements were not consecutive. Only the time variable was significantly different in 1969, and no significant differences were observed in 1970 except the shade/site interaction and time related variables. This shows that the soil/shade interaction occurred in the 1970 growth period, as shown in Table 1.

Neither the shade treatment nor site location was statistically significant with respect to height growth over

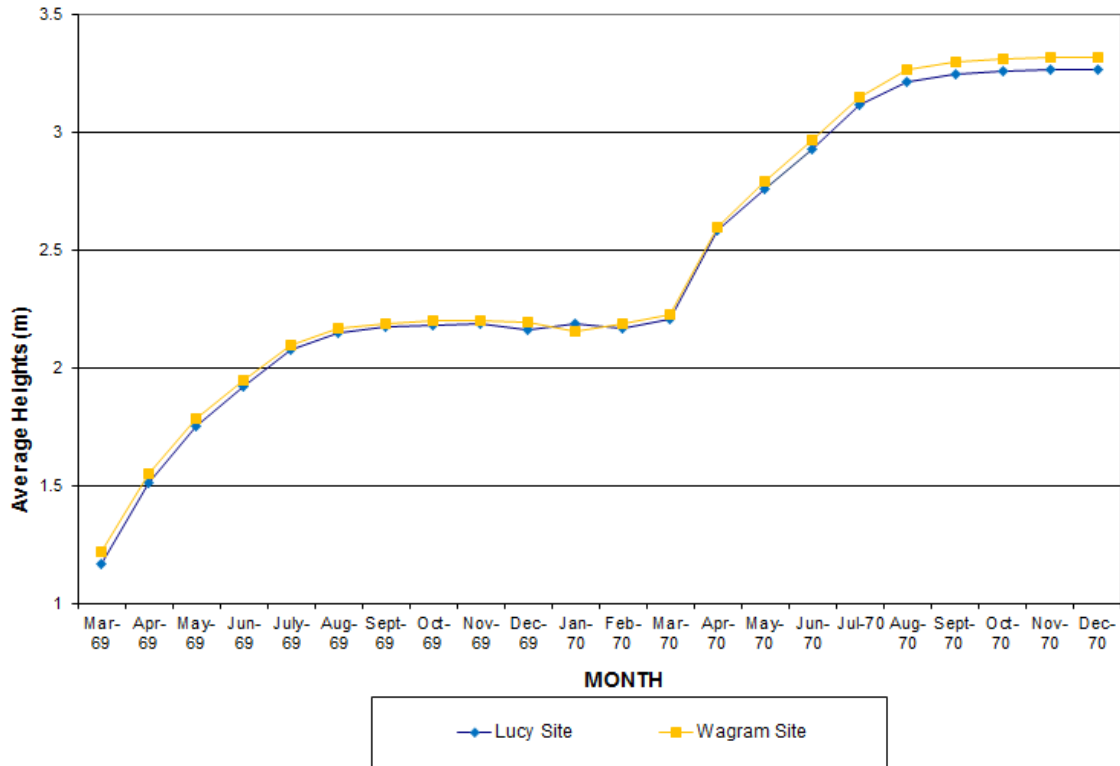


Figure 1. Average monthly height measurements illustrating a similar pattern of height growth on the Lucy and Wagram sites for seedlings sampled on the Escambia Experimental Forest, near Brewton, Alabama, USA.

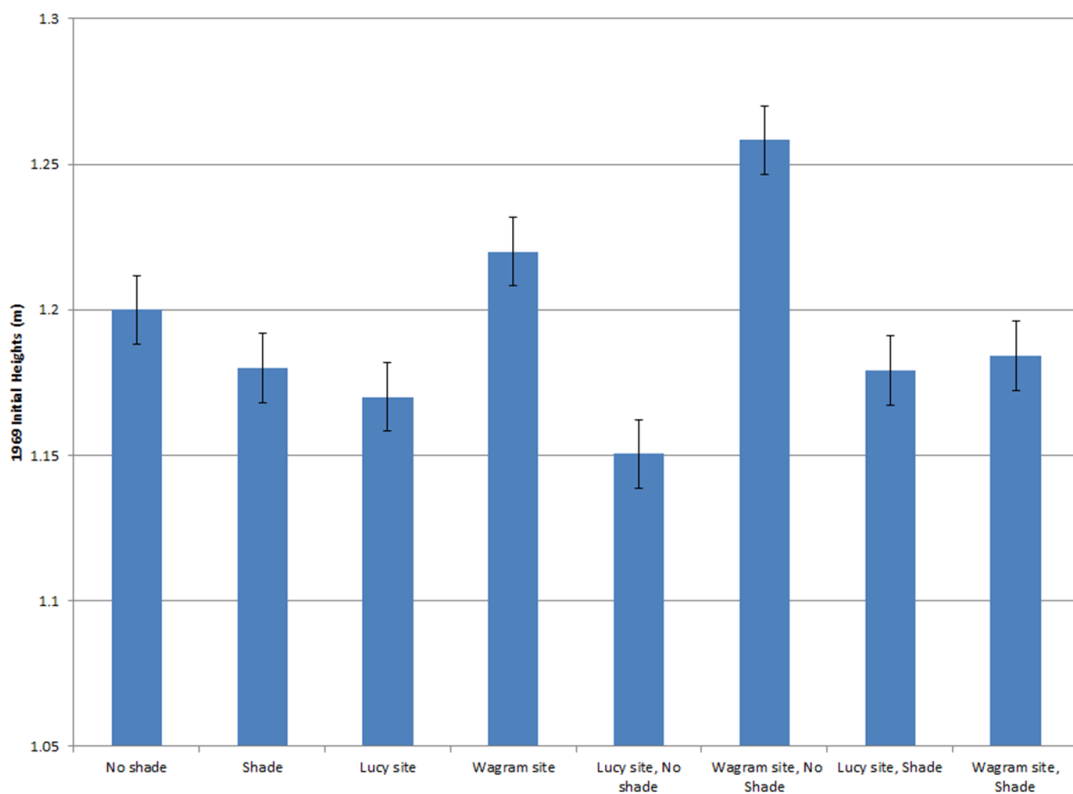


Figure 2. Average initial heights (m) in for seedlings sampled on the Escambia Experimental Forest, near Brewton, Alabama, USA.

Table 1. Test of average height growth and within group comparisons during the 1969 and 1970 intervals for seedlings sampled on the Escambia Experimental Forest, near Brewton, Alabama, USA.

| Year | Site type | Average height growth (m) |
|------|-------------------------|---------------------------|
| 1969 | Lucy | 1.01a |
| | Wagram | 0.99a |
| 1970 | Lucy | 1.09b |
| | Wagram | 1.12b |
| Year | Shade treatment | Average height growth (m) |
| 1969 | Not Shaded | 0.99c |
| | Shaded | 1.01c |
| 1970 | Not Shaded | 1.09d |
| | Shaded | 1.12d |
| Year | Shade/site combination | Average height growth (m) |
| 1969 | NW (Not Shaded, Wagram) | 1.02e |
| | SL (Shaded, Lucy) | 1.06e |
| | SW (Shaded, Wagram) | 0.96e |
| | NL (Not Shaded, Lucy) | 0.97e |
| 1970 | NW (Not Shaded, Wagram) | 1.20f |
| | SL (Shaded, Lucy) | 1.20f |
| | SW (Shaded, Wagram) | 1.04fg |
| | NL (Not Shaded, Lucy) | 0.98g |

Means with the same letter are not significantly different ($\alpha = 0.05$); NL = no shade, Lucy site; NW = no shade, Wagram site; SL = shade, Lucysite; SW = shade, Wagram site.

the intervals, in overall tests, or in years evaluated. An interaction between the shade treatment and site location was significant. The interaction was not significant for the 1969 growth interval, but it was for 1970. When isolating 1970 growth, the same general interactions occurred, but mean height growth for seedlings on the Lucy site with no shade treatment was significantly lower than on the Wagram site with no shade treatment and seedlings on the Lucy site with the shade treatment. The significant interaction for the total growth over the 2 years showed that mean height growth for seedlings on the Wagram site with no shade and seedlings on the Lucy site with the shade treatment was significantly greater than mean height growth for seedlings on the Wagram site with the shading treatment. The within group interactions for 1970 and overall height growth were significant for LSD means but not for HSD means. Figure 3 displays mean height growth over the two year measurement period from March of 1969 through December 1970 for the shade treatment, site and shade/site combinations, which illustrates the findings from the statistical examinations.

Diameter growth: 1969 to 1970

Patterns of growth over the growing seasons show that seedlings diameters do not grow at the same rates during

the entire growing season. Figure 4 displays average monthly diameter measurements illustrating how patterns of growth changed over the study period. There was a loss of diameter growth as shown in Figure 4 during January and February of 1970, which were the coldest months of the study with average minimum temperatures of 1 and 0°C, respectively. The initial diameter measurements ranged from 2.97 to 4.64 cm and were not significantly different for site location or shade treatment at the beginning of the study. There was a significant shade/site interaction. For site locations, Levene's test for homogeneity of variance was significant, but Barlett's was not. Testing within group interactions for initial diameters showed that diameters between shade treatments on the Wagram site were significant for LSD means but not for HSD. Figure 5 displays average initial diameters for shade treatments, site locations and shade/site locations at the beginning of the study.

Monthly diameters were measured from March 5 to December 24, 1969. When testing overall diameter growth for 1969, a significant difference of mean diameter growth between seedlings on the Lucy site and the Wagram site was observed (Table 2). Diameters were measured from January to December 1970 (Table 2). Testing within group differences, site location was significant, and mean diameter growth was non-significantly larger for the seedlings

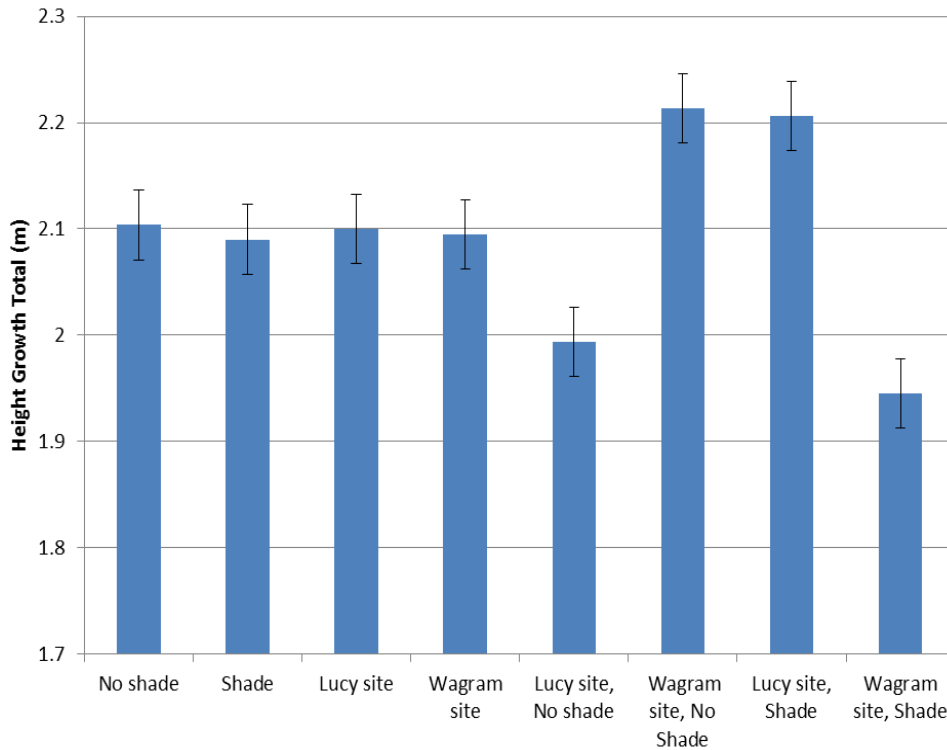


Figure 3. Average height growth and within group comparisons during the 1969 and 1970 interval from March 1969 to December 1970 for seedlings sampled on the Escambia Experimental Forest, near Brewton, Alabama, USA.

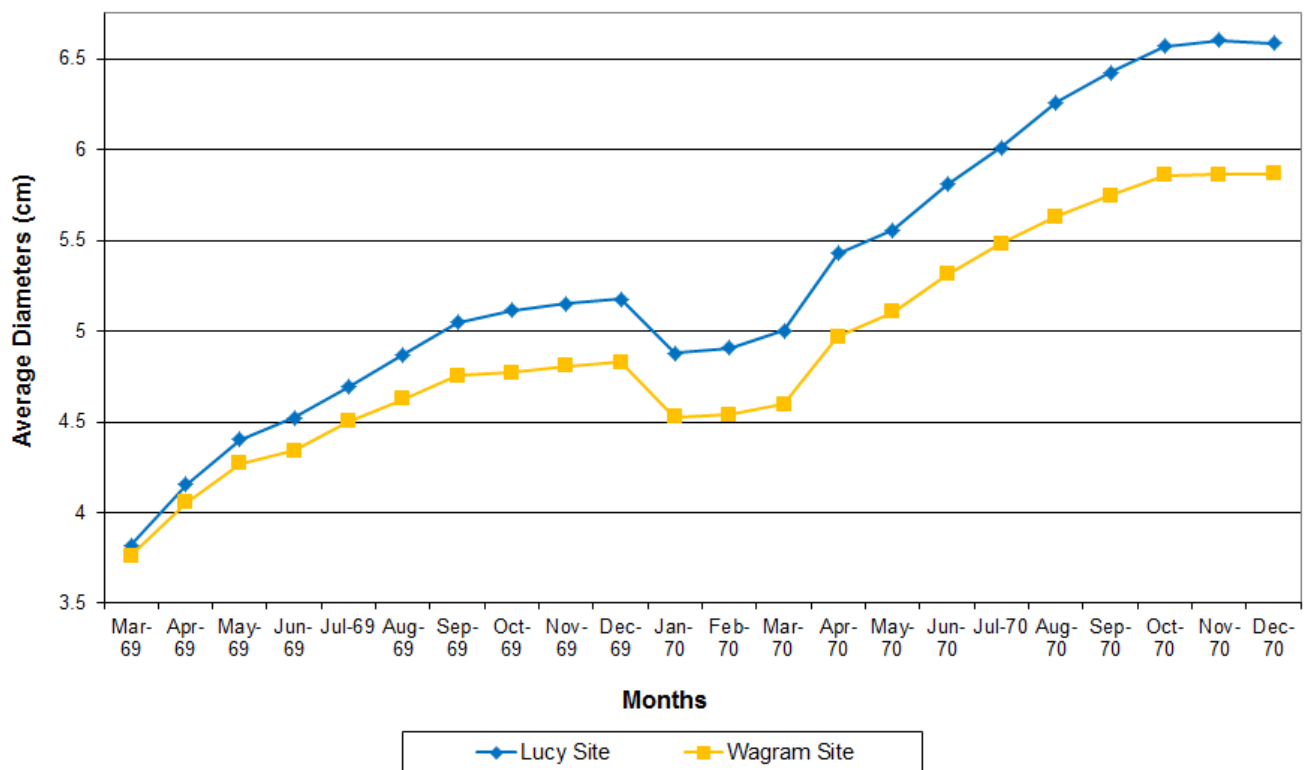


Figure 4. Average monthly diameter measurements illustrating greater diameter growth on the Lucy site for seedlings sampled on the Escambia Experimental Forest, near Brewton, Alabama, USA.

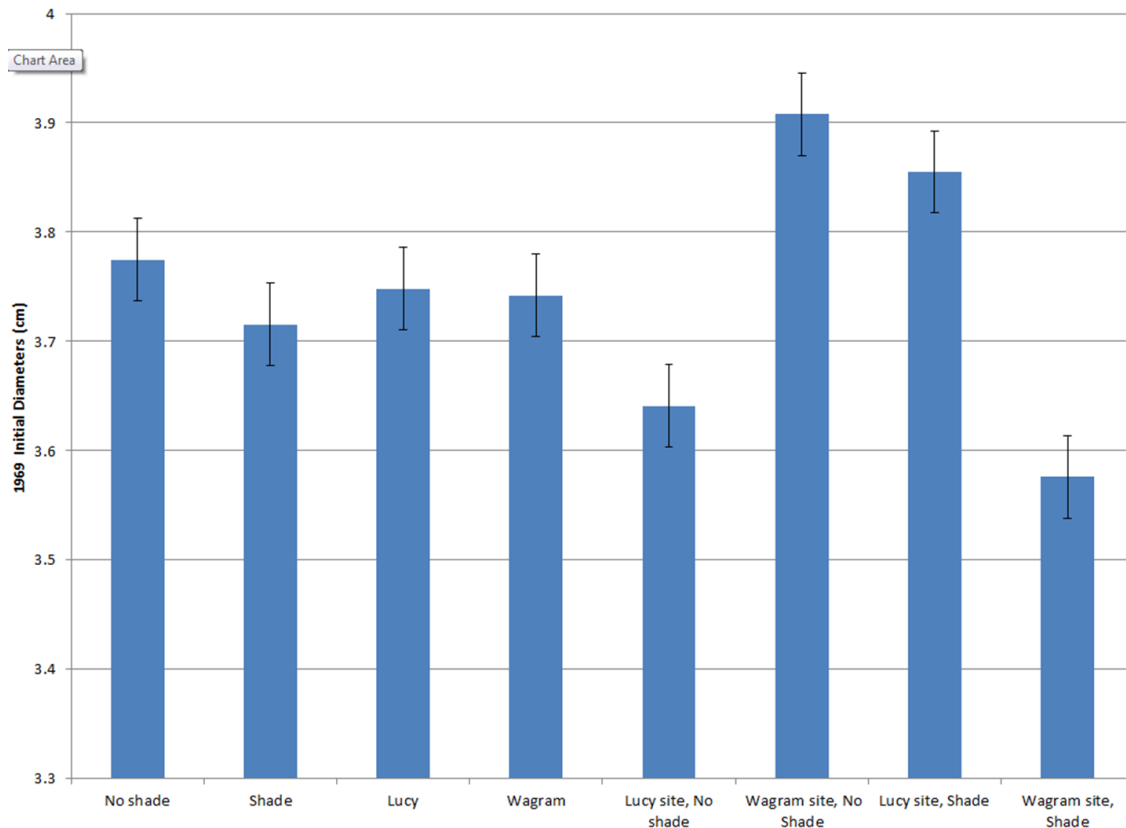


Figure 5. Average initial diameters (cm) in for seedlings sampled on the Escambia Experimental Forest, near Brewton, Alabama, USA.

with the shade treatment on both sites. A significant difference between seedlings on Lucy site and the Wagram site was observed for overall growth during the 1970 monthly interval and total growth over the two year period. Figure 6 displays mean diameter growth over the two year measurement period from March of 1969 to December 1970 for the shade treatment, site and shade / site combinations, which illustrates the findings from the statistical examinations.

Monthly diameter growth was also explored for potential significant differences using the repeated measures design. The results of the monthly measures analysis over both years showed a significant difference for site location, shade treatment and significant interactions between time and shade and time and site. Repeated measures analyses also showed a significant shade treatment for diameter growth over the monthly interval in 1970 and the weekly intervals in 1969 and 1970. The weekly analyses showed significant interactions between soil conditions and time and the shade treatment and time, but there was no significant interaction between the site location and shade treatment. However, adjusting weekly interval tests for autocorrelation did affect the analyses, and the normality assumption was suspect for the weekly 1970 test. Calculating the overall means for each year, the shade treatment was not significant, but

testing growth within each site location during 1969 showed that shaded seedlings grew less than the non-shaded seedlings. The opposite was seen in 1970. The biweekly intervals from 1969 and 1970 were also tested, but there were no significant differences or interactions in diameter growth with respect to site location or shade treatment for the 1969 interval. The 1970 biweekly interval showed a significant shade/site interaction, but neither the shade treatment nor site location was statistically significant.

DISCUSSION

The desire to identify and develop a better understanding on the many relationships that exist between the environment and seedling growth was the main purpose of this research work. To determine this, height and diameter growth were evaluated over numerous intervals during various periods of the growing and dormant seasons. It was necessary to examine seasonal patterns of growth to understand how seedlings grow before attempting to determine what is affecting growth. Figures 1 and 4 show average monthly height and diameter measurements over the two year measurement period, which displays the seasonal patterns of growth for the sampled longleaf pine seedlings. The six month shade treatment was installed

Table 2. Test of average diameter growth and within group comparisons during the 1969 and 1970 intervals for seedlings sampled on the Escambia Experimental Forest, near Brewton, Alabama, USA.

| Treatment and site | Average diameter growth (cm) | |
|-------------------------------|------------------------------|---------------------------------|
| | 1969 | 1970 |
| No shade | 1.31a | 1.49a |
| Shade | 1.21a | 1.56a |
| Lucy | 1.43b | 1.71b |
| Wagram | 1.09c | 1.34c |
| Shade/site combination | | |
| NL | 1.50d | 1.67d |
| SL | 1.36de | 1.76d |
| NW | 1.11fe | 1.30e |
| SW | 1.06f | 1.38e |
| Analysis of variance | DF | Probability > F-value |
| Site | 1 | 0.0004* |
| Shade | 1 | 0.2674 |
| Site-Shade interaction | 1 | 0.5903 |
| Error means square | 36 | 0.9436 |

Means with the same letter are not significantly different ($\alpha=0.05$); df – degrees of freedom; NW = no shade, Wagram site; SW = shade, Wagram site; NL = no shade, Lucy site; and SL = shade, Lucy site.

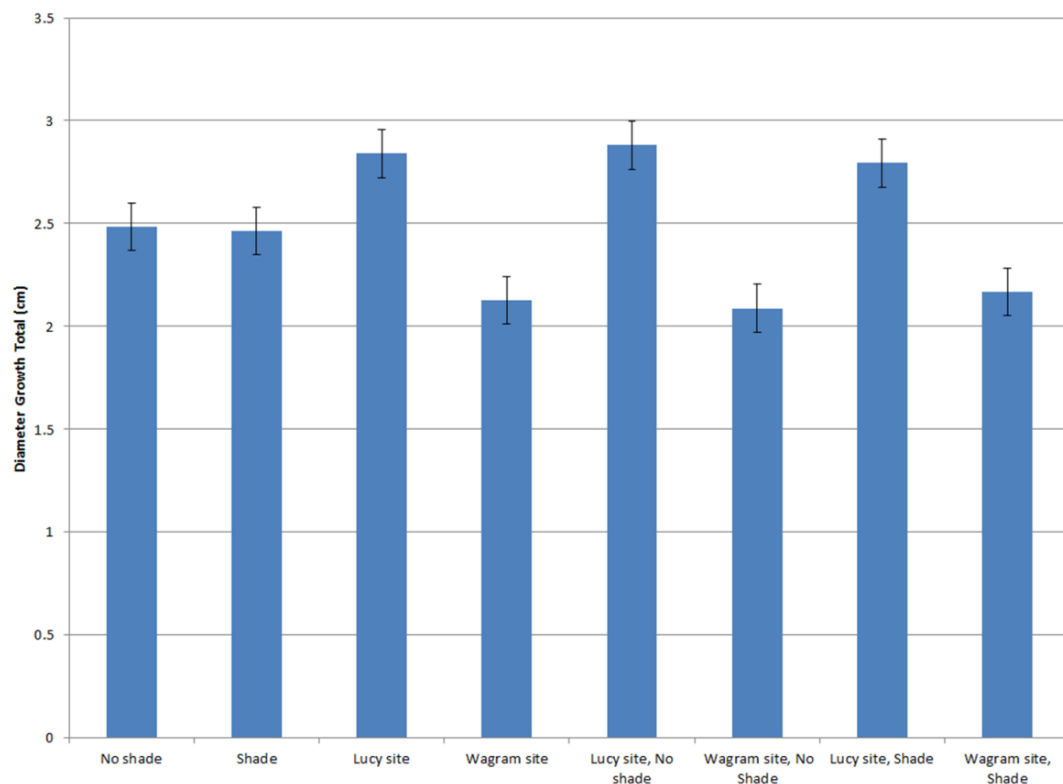


Figure 6. Average diameter growth and within group comparisons during the 1969 and 1970 interval from March 1969 to December 1970 for seedlings sampled on the Escambia Experimental Forest, near Brewton, Alabama, USA.

during the peak of the growing season.

Longleaf pine is commonly known to be very shade intolerant and drought resistant. To determine how much light intensity affects the growth of longleaf pine seedlings, a shading treatment was randomly installed to half of the sampled seedlings. Six months of shading caused a percent difference in direct solar radiation of about 38% during the 1969 growing season. The treatment seemed to have no significant effect on height growth of young longleaf pine over any of the intervals analyzed except for a larger average height growth for seedlings on the Wagram site when looking at growth over the two-year period. For the Lucy site there was an opposite trend for 1970 growth where shaded seedlings on average grew significantly better than non-shaded seedlings (Table 1). This could be a residual effect from the shading treatment because weekly diameter growth of both 1969 and 1970 was significant with respect to the shade treatment. In this study, the shade treatment did not affect diameter growth in either year. Mean diameter growth was also lower, however not significant, for shaded seedlings on both sites, and it was not significant on an annual basis.

The lack of an overall effect on height growth potentially occurred because of several reasons. One reason might be the percent of shading caused by the cheesecloth, or the height it was placed. The cheesecloth was suspended and maintained at 1 m above the terminal bud, which allowed the seedling to receive lateral rays of sunlight during the early morning and late afternoon. Schwarz (1907) notes that areas of dense seedlings and saplings were able to survive in virgin stands by receiving light during the morning and afternoon from certain angles where light entered low in the forest. Duration is another key factor. The treatment was only applied in the 1969 growing season. It may take a longer duration of shading to see an effect on growth. This along with the low percent of sunlight reduced by the cheesecloth did not seem to change the growth rates of the shaded seedlings in comparison with those that were not shaded at all, although overhead shade levels of 50% reduced diameter growth of 20-week-old longleaf pine seedlings growing in a trusshouse with shade cloth (Barnett, 1989). In contrast, a greenhouse study with 16 months of continuous shade of over 60% did not have a significant effect on height growth or root collar diameter of longleaf pine seedlings growing in a greenhouse, but they did find a significant response in root and stem biomass with interactions between light and soil resources (Jose et al., 2003). Climent et al. (2006) also found significant effects in biomass with reductions in light for Canary Island pine seedlings. Testing for significant effects of shade and site on above and below ground biomass for different age classes of seedlings might provide interesting results for the different stages of longleaf pine growth.

Longleaf pine is commonly thought of as a species that is intolerant to shade. This concept may not be completely understood. This study provides data from the only

known in-field shading experiment with longleaf pine seedlings that had initiated height growth and were initially up to 1.5 m tall, which are very different from grass stage seedlings or controlled studies. Cavender-Bares and Bazzaz (2000) found that age classes and environmental components played any equal role in photosynthetic capacity if water was not a limiting factor between red oak seedlings and mature trees. The intolerance of longleaf pine to shade may depend more on the age class or stage of growth like grass stage or after height growth is initiated, location of the tree in the stand, and the duration of the shading.

Site location was another key factor that needed to be evaluated in determining a possible influence on growth. Seedlings were on different sites and the two measures of growth were affected in different ways. Site location did not seem to have a significant effect on seedling height growth, but it did for diameter growth. Site location significantly affected diameter growth over all measurement periods during the two-year study including the dormant season measurements. Diameter growth for seedlings on the Lucy site was significantly greater than diameter growth for seedlings on the Wagram site across all intervals except for the biweekly datasets. Diameter growth varied by location in both years as seen in Table 2. Figure 4 shows average monthly diameter measurements over the two year measurements, which displays greater diameter growth on the Lucy site for seedlings sampled. The difference in growth seemed to increase as the seedlings got older. One explanation could be that longleaf pine grows better on drier ridge top site with Lucy soils due to potentially less ground layer competition than Wagram soil on the lower side slope site. If the root systems were deep enough to reach the B-horizon, there might be a moisture gradient caused by the higher clay content at shallower depths than at the bottom of the ridge. The significant differences in diameter growth by site location may also be due to competition of surrounding seedlings or other vegetation instead of soil differences, but without more detailed data about surrounding seedlings and vegetation this concept cannot be further explored. Another explanation might be due to different levels of non-pine competition. Longleaf pine often grows slower on sites with more ground-layer competition (Pessin, 1938) and it is possible that competition was greater at the Wagram site. The growth difference may also be due to differences in soil characteristics. Without more detailed data on surrounding seedlings and vegetation, we can only consider the primary cause of better diameter growth on the Lucy site.

Conclusions

This study provides an interesting look at the growth of longleaf pine over two years and provides insight about the question of the shade tolerance of longleaf pine. This concept may not be completely understood and needs to

be explored in detail. With the resurgence of interest in longleaf pine and restoration, shade tolerance plays a large role in the stand dynamics of restoration and conservation efforts. This study provides data from the only known in-field shading experiment with longleaf pine seedlings that had initiated height growth and were initially up to 1.5 m tall at 14 years from seed, which are very different from grass stage seedlings or controlled studies. This makes the results difficult to compare with other studies. The intolerance of longleaf pine to shade may depend more on the stage of growth like grass stage or after height growth is initiated, location of the tree in the stand, and the duration of the shading. There are still many unanswered questions about influences of the environment on the growth of young longleaf pine seedlings. Future field based studies can build upon this research and explore the shade tolerance, environmental interactions, and phenotypic plasticity for seedlings of this size and utilize current technology to help answer more questions. More understanding about relationships between seedling growth and the environment is very important in elucidating patterns of seedling growth.

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Full Length Research Paper

Fine-scale spatial distribution of biomass using satellite images

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Spatial information on the distribution of biomass is an important issue for monitoring and managing the environment. It is a prerequisite for successful forest fire management and for predicting fire intensity and fire behaviour, but estimates of biomass are time consuming and expensive and need to be done depending on size classes. We propose a method that takes into account the contemporary use of an allometric approach and remote sensed data from Very High Resolution (VHR) satellite images to obtain distribution maps of biomass subdivided into different components while keeping plant-destructive collection of data to a minimum. To test the feasibility of distributing biomass into classes, we subdivided biomass into two size classes according to the size of leaves (thickness) and branches (diameter). This is an approach that can be adapted to both fuel classes or to estimation of the ligneous component. We haphazardly selected eight areas, within the Site of Community Importance "Monteferrato e Monte Iavello" (Italy), where easy-to-measure characteristics (height, diameter, cover) of vascular plants were collected. Regression equations between easy-to-measure vegetation characteristics and biomass values were derived to estimate the biomass of each area in the two size classes. Then, we evaluated the relationships between the normalized difference vegetation index (NDVI) and the estimated biomass values for each area using regression equations and size class. The equations that resulted from the regression analysis were used to create maps of biomass using NDVI map. Such a procedure allows the identification of features otherwise lost when the vegetation is represented only by vegetation class labels. This includes the orientation of vegetation lines which may favor the spread of fire in a given direction; information that may be useful for hazard management and prevention.

Key words: Allometric equation, *Pinus pinaster*, *Erica scoparia*, biomass size class, Very High Resolution (VHR) satellite imagery.

INTRODUCTION

It is known that anthropogenic climate change and increasing human impact will lead to increased pressure on the environment, including natural and semi natural vege-

tation. Monitoring is considered essential and can be frequent and even cheap if satellite images are used. Indices relative to the biomass (such as greenness index and

NDVI) are often used for evaluating biomass amount and vegetation health. This information is not enough and it is often necessary to have the distribution of biomass subdivided following some criteria. A common one, probably the most useful, is based on size classes which can help in estimating the amount of photosynthesizing biomass, or the amount of easy and quick to burn material, or the amount of ligneous material, which, once estimated at different time interval, can help the estimation of dead branches and trunk (this also important for fire hazard evaluation). In the case of biomass as fuel in wildfires biomass size, classes are important. Fires are predicted to potentially be more widespread and more frequent (Parry et al., 2007; Marlon et al., 2009) as the climate continues to be warm. Several studies indicate that the increase in the frequency of wildfires and the longer duration of the fire season are linked to increased spring and summer temperatures (Westerling et al., 2006), changes in the pattern of precipitation (Flannigan et al., 2000), and development of high fuel loads associated with long-term fire suppression (Schoennagel et al., 2004).

In the light of the scientific world's diagnosis of new climatic scenarios, wildfires in the Mediterranean basin represent a serious worry, considering that here wildfires are a common, and mostly human-induced feature where they have long played an important role in modifying vegetation patterns (Montenegro et al., 2004). Large-scale summer wildfires throughout the region have dramatically increased in the last few decades (Nunes et al., 2005; Bajocco and Ricotta, 2008; Catry et al., 2009; Ricotta et al., 2012), mainly as a consequence of rapid land-use changes (marginal rural land abandonment increasing fuel accumulation), socio-economic conflicts and competing interests, in conjunction with climatic warming which is reducing fuel humidity (Pausas and Vallejo, 1999).

In this context, spatial information on the distribution of fuel load (biomass weight) is a prerequisite for successful forest fire management and for predicting fire intensity and fire behaviour (Rothermel, 1972; Gray and Reinhardt, 2003). The fuel load determines the potential amount of heat that can be released during a fire, whereas the type and distribution of fuel elements affect their combustibility. Fine fuels burn more readily than coarse ones. Fine fuels react faster to weather changes, particularly if these fuels are dead, and they play a major role in the initial stages of all fires (Baeza et al., 2002). Fuel models considering the different types of biomass (fine and coarse) are an important factor that should be taken into consideration for fire planning, assessing fire risk, and improving fire prevention since fuel types may present completely different fire propagation rates and fire behavior.

The feasibility of modeling fuel/biomass distribution by remote sensing data has been frequently discussed in several studies and estimates of fire hazard and distribution maps of fuel have been provided (Vidal et al., 1994; Vidal and Devaux-Ros, 1995; Burgan et al., 1996; Riano et al., 2002; Rollins et al., 2004; Lasaponara and

Lanorte, 2007). Nevertheless, the use of remote sensing images has been based on the analysis of medium- to high-resolution sensors, such as Landsat TM data without the subdivision of biomass into size classes.

Generally, satellite data are expressed in the form of spectral indices that attempt to enhance the spectral contribution of different features distributed over a surface. One of the most promising applications of satellite data is the estimation of biomass or primary productivity over time and space through satellite derived vegetation indices (Cihlar et al., 1991; Todd, 1998; Pettorelli et al., 2005). To be effective biomass estimators, spectral indices must be able to differentiate vegetation features; the characteristic reflectance pattern for green vegetation is low reflecting in the visible portion of the spectrum (particularly red) with a sharp increase in reflectance in the near-infrared portion. Vegetation indices respond to these expected differences in near-infrared and red reflectance. These broad-band vegetation indices have shown to be well correlated with canopy parameters related to chlorophyll and biomass abundance. For example, the normalized difference vegetation index (NDVI) is calculated as the difference between near-infrared and red reflectance values divided by the sum of near-infrared and red reflectance values. The Normalized Difference NDVI is a widely used surrogate of the amount (as green biomass) and vigor of vegetation at the surface (Tucker, 1979; Richardson et al., 1983; Christensen and Goudriaan, 1993). Previous studies have related NDVI values to different vegetation attributes such as plant biomass (Tucker et al., 1985; Persson et al., 1993; Hobbs, 1995), leaf area index (LAI) (Waring, 1983; Tucker et al., 1986; Gilabert et al., 1996), net primary production (NPP) (Tucker et al., 1981, 1983; Paruelo et al., 1997) and percentage of absorbed photosynthetically active radiations (APAR) (Asrar et al., 1984; Sellers et al., 1992).

In particular, the use of correlations between NDVI and biomass has been found to be unstable (Richardson et al., 1983; Tucker et al., 1983; Christensen and Goudriaan, 1993). This is because the reflection coefficients are primarily determined by green foliage biomass and not the amount of dry matter (Christensen and Goudriaan, 1993). Thus, in order to assess the efficacy of NDVI in estimating biomass size classes and their spatial distribution, field-based quantitative estimates of available living or dead vegetation weights are needed (Roussopoulos and Loomis, 1979; Mikaelian and Korzukhin, 1997; Sah et al., 2004).

An attractive means for estimating forest biomass is through the use of empirical allometric relationships (Whittaker and Woodwell, 1968; Usó et al., 1997). Allometry describes relations or mutual proportions between different plant organs or in general structural characteristics. Since measuring plant biomass in field conditions is laborious and extremely time consuming, empirical relationships or models are used to estimate the biomass from in-field, easily measurable, biometric variables such as

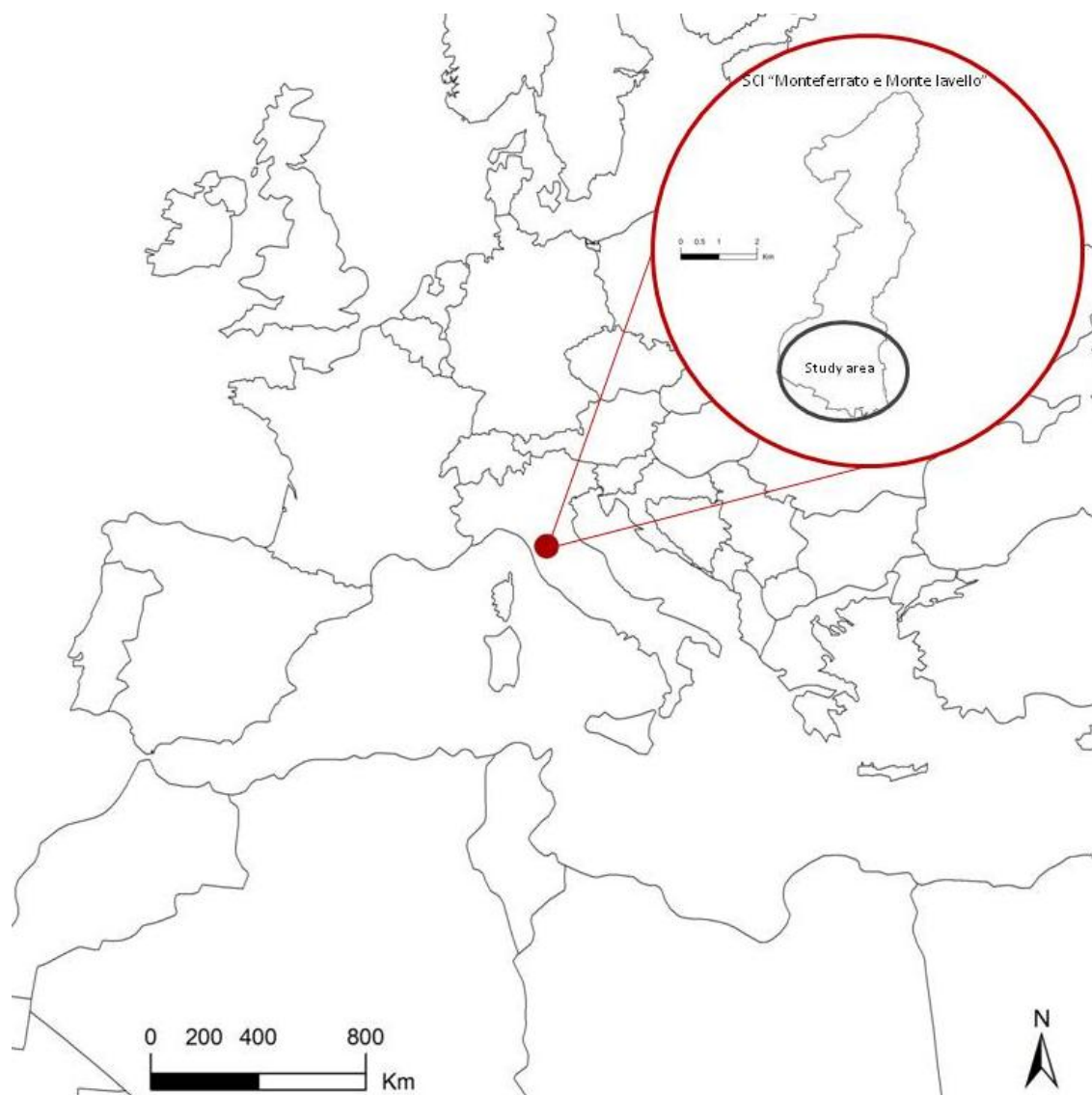


Figure 1. Location of the SCI “Monteferrato e Monte Iavello” (IT5140007) and the study area.

stem diameter, height, crown diameters or volume and crown cover. They have been used to estimate the biomass of trees, shrubs and herbaceous species in several environments (Assaeed, 1997; Ares and Fownes, 2000; Hierro et al., 2000; Zianis and Mencuccini, 2004; Northup et al., 2005; Pilli et al., 2006; Pokorný and Tomášková, 2007; Oñatibia et al., 2010).

The basic objective of this work was to develop a method for estimating biomass when subdivided into size classes and obtaining their distribution maps starting with the collection of simple vegetation characteristics and keeping the vegetation-destructive part of the sampling to a minimum. More specifically, this study aimed at: i) estimating biomass through the use of an allometric model based on relationships between biomass and vegetation characteristics directly measured in the field in some sample areas, ii) evaluating the potential of Very High Resolution (VHR) images (like QuickBird) for estimating

biomass/fuel, iii) assessing the reliability of NDVI index to spatialise values of biomass and fuel throughout the study area.

MATERIALS AND METHODS

Study area

The study was carried out within the NATURA2000 site of community importance (SCI) “Monteferrato e Monte Iavello” (IT5140007; UTM32 667237E, 4867255N), located west of Florence, Tuscany (Figure 1). The SCI is approximately 1375 ha in area and ranges in elevation from 60 to 936 m a.s.l., with south slope exposure. Within this area, there are 530 ha of outcrops of ultramafic rocks with serpentine where we focused our observations. Here elevation ranges between 61 and 420 m a.s.l. Mean annual rainfall is approximately 1037 mm, October is the wettest month with 140 mm and July is the driest month with 31 mm. Mean annual temperature is 14.4°C. The average temperature of the coldest month (January) is 6°C and the average temperature of the warmest month (July) is 24.1°C (LaMMA and CSN, 2001). In the southern part of the SCI, there is



Figure 2. The four photographs illustrate different types of vegetation cover present in the study area.

the Centre of Natural Science (CSN), which hosts an important volunteer group of fire control and prevention.

The study area is characterized by evergreen mediterranean shrubland and woodland with dominance of the maritime pine (*Pinus pinaster*) (Figure 2), introduced from the 1st half of the 19th century; some individuals of cypress (*Cupressus sempervirens*), and oak (*Quercus ilex*). Heather (predominantly *Erica scoparia*) dominates the shrub layer, with laurel (*Laurus nobilis*), rock rose (*Cistus* spp) and myrtle (*Myrtus communis*). The herbaceous vegetation is characterized by perennial grasses and forbs such as *Brachypodium rupestre* and *B. pinnatum* which grow under the tree and shrub canopy, and *Helichrysum italicum*, *Alyssum bertolonii*, *Bromus erectus*, *Festuca* spp., which are found in more xeric situations. Furthermore, exposed stones are common in places and ferns, lichens and mosses are also typically present (Figure 2).

Field biomass sampling

Ground data were collected to extract the allometric relationships between dry weight and easily measurable plant parameters (EMPP: such as diameter, height, crown diameters, crown cover) in shrubs and herbaceous layers during the summer of 2011.

In the study area, a series of shrubs (*Erica scoparia*) were haphazardly selected to encompass a range of height, diameters, and crown forms, in order to obtain a sample of different size shrubs varying from smallest to largest. For each shrub, the following parameters were measured: i) two maximum crown diameters (taken at right angles to each other across the canopy of the shrub); ii) mean height obtained by measurements taken at many points, along transects running parallel or orthogonal to the major diameter (minimum number of observations was 10 values over 2 transects)

(Figure 3). Afterwards, shrubs were cut at ground level and the fresh biomass was weighed with a portable scale.

A series of 60 x 60 cm plots were distributed with systematic sampling in the study area to collect herbaceous layer. For each sample, the percentage cover of herbaceous layer was estimated; all plant species were cut at ground level and weighed with a portable scale (1 g resolution).

In order to correctly evaluate the amount of biomass, the vegetation mass was arbitrarily divided into two size classes using the value of 1 cm as the delimiter between fine and coarse biomass. The size group <1cm of diameter (shrub leaves and fine parts of branches and sprouts, and herbaceous species) includes material that can ignite quickly and burn completely in a short time and the size group >1 cm (shrub coarse material) is mainly made of large small to large branches which will burn for a longer period but will ignite less quickly (Deeming et al., 1972). Chopped shrub and herbaceous species were placed in nylon bags, labeled, and transferred to the laboratory for calculating dry weight (oven-dried at 100°C until constant weight was reached). All components were then weighted separately, in order to calculate fuel load and fuel moisture for each class.

Then, for each sample of shrub and herbaceous species, we obtain: total biomass in g per each of the chosen dimension classes and their moisture content (water % of dry weight), the mean shrub height (m), two shrub diameters, area and volume of each shrub, percentage cover of herbaceous species.

For the tree layer, allometric equations were found to exist for similar pine forests (*Pinus pinaster*) during the literature review (Giovannini et al., 2001) and were used during this study, assuming that they were applied here too. Giovannini et al. (2001) related DBH and tree dry weight, dividing the dry weight in three fractions: total tree, fine wood (diameter <5 cm) and leaves.

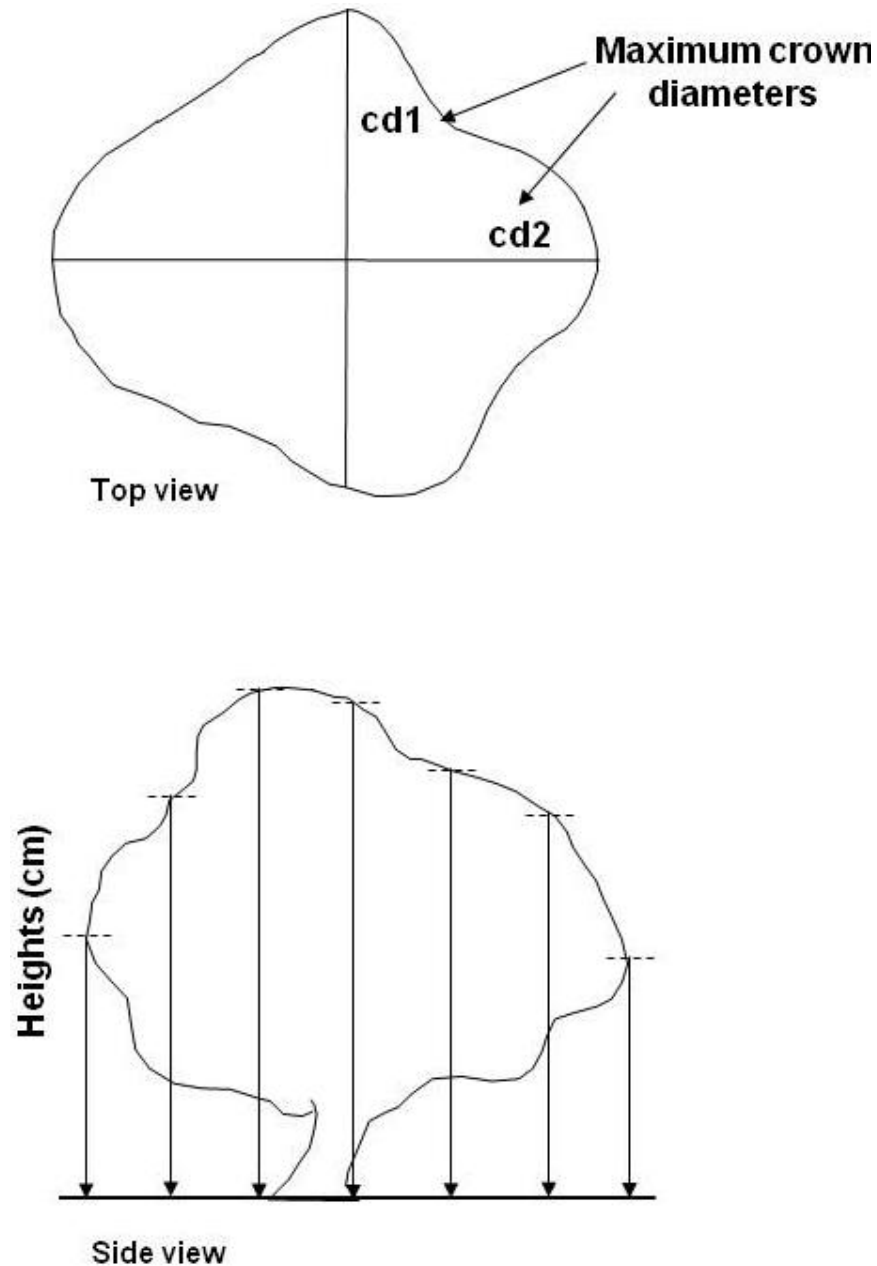


Figure 3. Top and side view of a typical shrub showing measurement technique used.

Spectral operations for estimating biomass patterns

A QuickBird multispectral image (spatial resolution 2.4 m; spectral resolution from 450 nm to 890 nm: four bands), acquired on 9 February 2009 (Figure 4), was pre-processed for both radiometric calibration and geometric corrections to ensure that each pixel in the image faithfully records the same type of measurement at the same geographic location over time (Kennedy et al., 2009) and to minimize the signal-to-noise ratio. Radiometric calibration included a linear transformation of the Image Digital Numbers (DNs) into Top of Atmosphere Radiance (TOARD) and then a non-linear transformation of TOARD into TOA Reflectance (TOARF) values ranging into [0,1], with these considered a parent class of surface reflectance values in an ideal (atmospheric-noise free) case (Baraldi et

al., 2010). This assumption is made when ancillary data for atmospheric correction are not available. A geometric correction was performed in order to assign satellite images to their correct position on the Earth's surface (georeferencing) using Ground Control Points (GCP) as known points with a polynomial warping function and a nearest neighbor resampling to the Datum WGS84/UTM coordinate system.

From calibrated images, spectral indices can be extracted with the aim of enhancing the spectral contribution of vegetation while minimizing that of the background. The normalized difference vegetation index (NDVI) is one of the most widely used vegetation indexes and its contribution in satellite assessment and monitoring of global vegetation cover has been well demonstrated over the past two decades. It is defined as:



Figure 4. A true color image-map of the QuickBird scene acquired on 9 February 2009.

$$\text{NDVI} = (R_{\text{NIR}} - R_{\text{RED}}) / (R_{\text{NIR}} + R_{\text{RED}})$$

Where, R_{RED} , and R_{NIR} represent surface reflectance averaged over visible ($\lambda \sim 0.6 \mu\text{m}$) and NIR (near infrared) ($\lambda \sim 0.8 \mu\text{m}$) regions of the spectrum, respectively. The NDVI is correlated with certain biophysical properties of the vegetation canopy, such as fractional vegetation cover, vegetation condition, and biomass.

Assessing biomass in georeferenced areas using NDVI

Once the allometric relationships between easy-to-measure plant parameters (EMPP) and biomass (dry weight) were established, we used them to estimate the biomass/fuel in georeferenced areas (polygons) with uniform vegetation types in terms of density. For each georeferenced area, we analysed the relationships between mean NDVI and estimated biomass values (fine and coarse) per unit area (t/ha) using linear regression models.

Within each stand, two areas were randomly selected. Understorey vegetation (herbaceous and shrub layers) in each area was collected in square plots (1 x 1 m) 2 m spaced along belt transects. The number of plots was proportional to extent area. For all shrubs rooted in the plots, the following easy-to-measure plant parameters (EMPP) were determined: the longest crown diameter, the crown diameter perpendicular to the longest, total plant height the crown height of live foliage and the basal diameter (stem diameter just above ground level). Percent coverage of species in the herbaceous layer was recorded. Within each area, we measured the DBH (trunk diameter at breast height) and the height for each tree.

The regression models between EMPP and biomass values, allowed us to estimate the total biomass (fine and coarse) for shrub and herbaceous layers for each area through the application of the

regression equations. For the tree layer, the total pine dry weight, the fine wood fraction and coarse fraction were estimated by DBH measurements using allometric models developed for similar pine forest in Mediterranean area by Giovannini et al. (2001). The biomass proportions were summed to give the tree biomass.

Statistical approaches for estimating biomass

Regression analyses were performed to determine the relationship between biomass and EMPP. The independent variables used were crown diameters (cd1, cd2), mean height (H), crown area (ca) of shrub plants and percentage cover of herbaceous plants. The dependent variables were fine, coarse and total biomass and were tested for normality of distribution using the Shapiro-Wilk test (Shapiro and Wilk, 1965), and if necessary transformation was made.

Linear ($Y = \beta_0 + \beta_1 X_1$) or non linear regression models ($Y = \text{dry weight of biomass in grams}, X_1 \dots X_n$ are the respective explanatory variables in each model) were used to assess the choice of independent variables and predictive equations selected based on adjusted R^2 values and the significance (p-value) of the regression coefficients. GIS analyses were performed with GRASS (GRASS Development Team, 2011) and statistical analyses using R 2.13.1 for Windows (R Development Core Team, 2011).

RESULTS

The allometric models based on regression equations models all resulted as statistically significant and explained between 66 ($R^2 = 0.66$) and 93% ($R^2 = 0.93$) of the variability in individual biomass (fine and coarse). In parti-

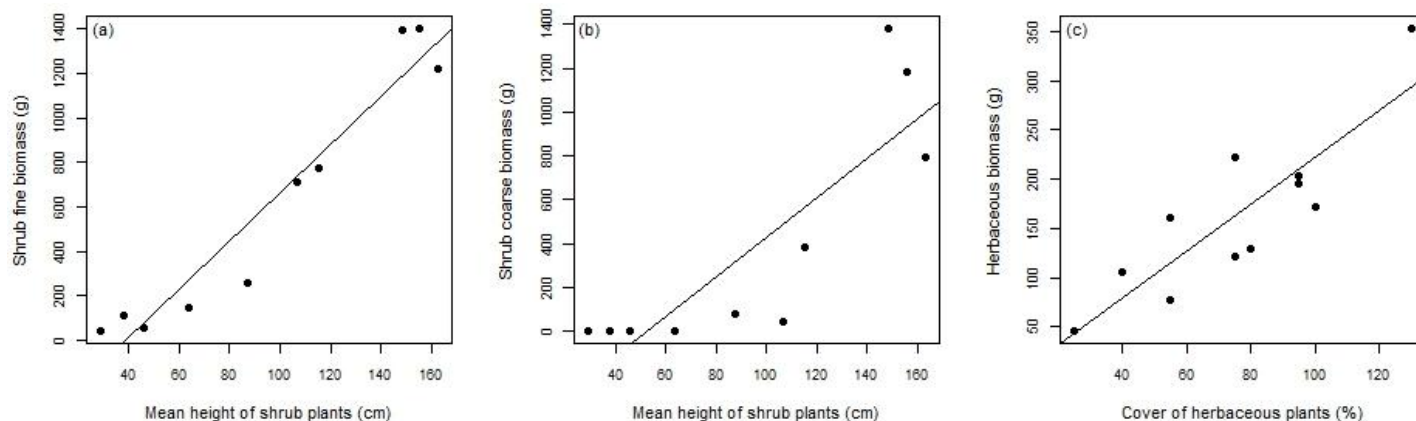


Figure 5. Scatter plots between shrub fine, shrub coarse and mean high of shrub and between herbaceous biomass and percentage of herbaceous plants cover.

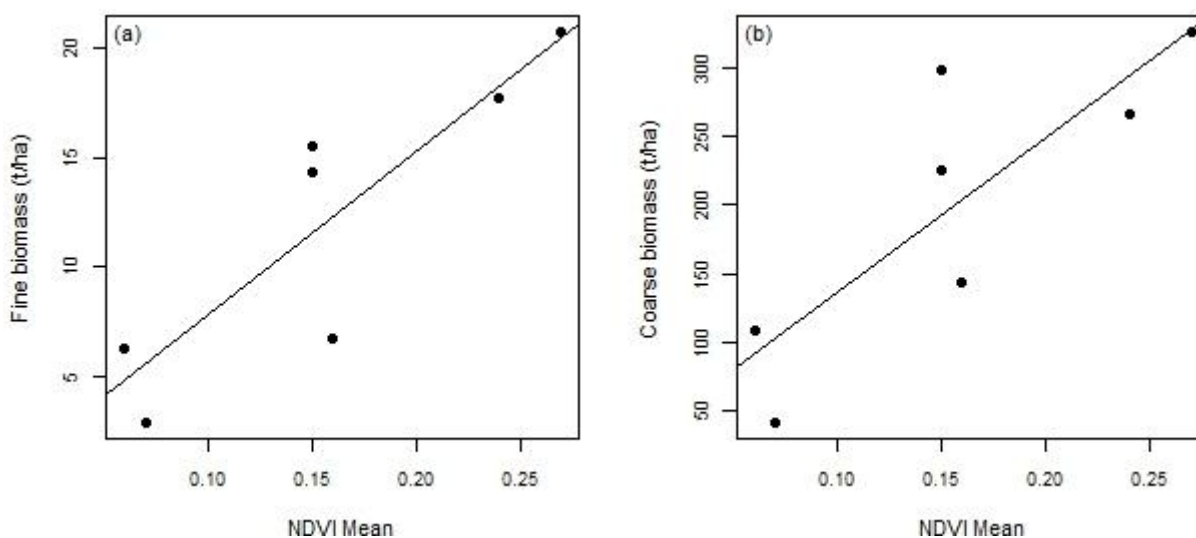


Figure 6. Scatter plot between NDVI and fine and coarse biomass.

cular, results of the regression analysis indicate that the mean height was the variable that best predicted fine and coarse shrub biomass (Table 1). The linear relationships with R^2 above 0.7 were chosen as the best fitting equations to the predicted fine and coarse fuel biomass (Table 1). Figure 5 shows the scatter plot of best relationships obtained by regression analyses: mean height of shrub plants plotted against fine and coarse shrub biomass displayed a positive trend as scatter is significant. Likewise, the scatter plot between percent cover of herbaceous plants and herbaceous biomass indicated a good relationship. These scatter plots can give an idea of the effectiveness of EMPP in investigating its relationship with biomass.

Allometric analysis showed that the percentage of coverage of herbaceous species is an effective allometric measure for predicting the biomass in the herbaceous vegetation layer, since the regression result was statis-

tically significant (Table 1). The NDVI index, performed on QuickBird images with Very High Resolution, confirmed itself as a good predictor of fine and coarse dry weights for estimating biomass/fuel in this area, as reported in Table 2. Figure 6 shows mean NDVI plotted against fine and coarse biomass.

The coefficients results obtained from the regression analysis were used to distribute the fuel values on the area of interest and to create a fine-fuel map and coarse-fuel map (Figure 7a, b). The fine fuel ranged from 0.02 to 20 t/ha and the coarse fuel ranged from 0 to 320 t/ha.

DISCUSSION

The knowledge of how the distribution of natural biomass weights subdivide into size classes is important for improving current fire prevention and fire behavior modeling,

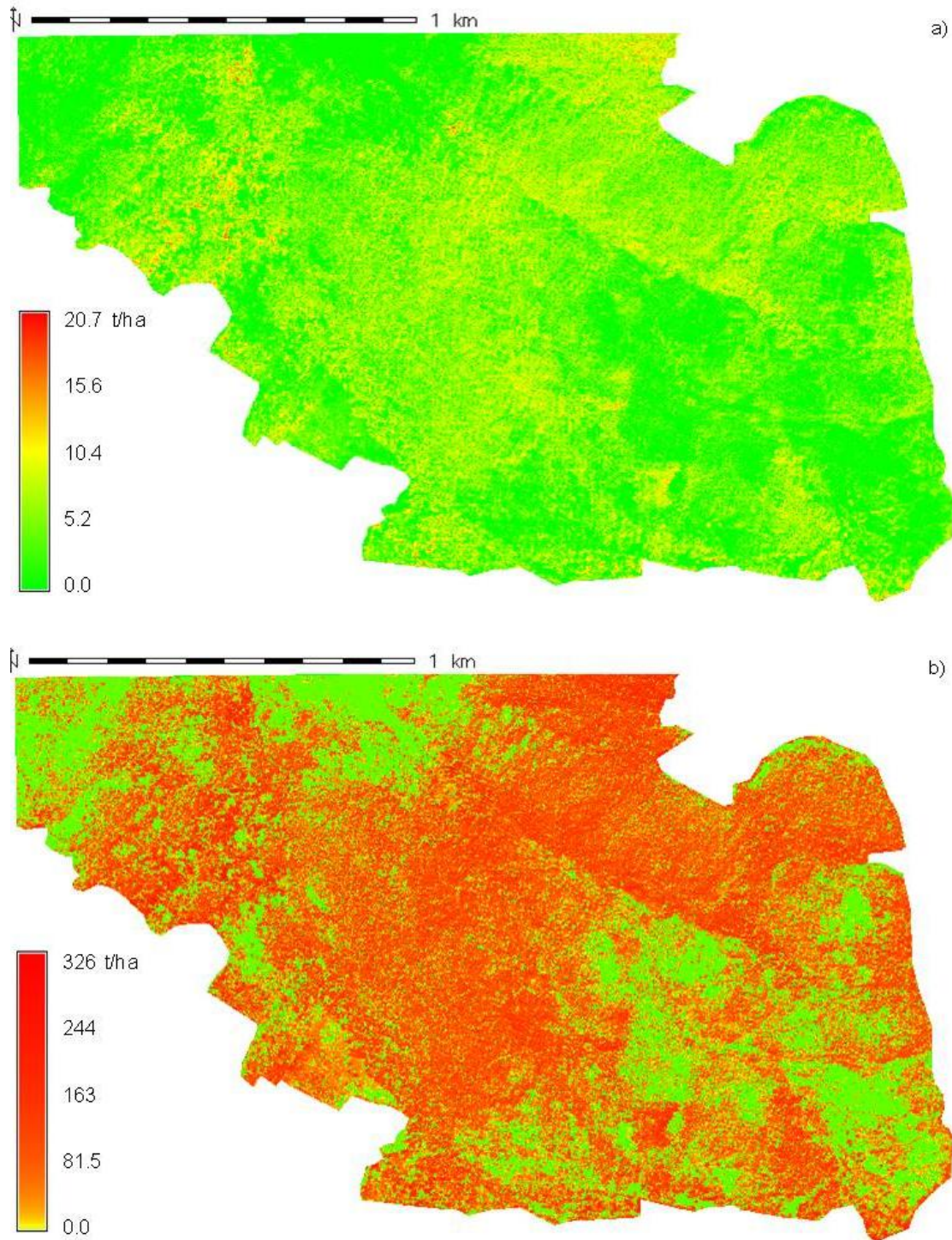


Figure 7. Distribution of fine (A) and coarse (B) biomass (t/ha) in the study area.

which can alleviate the negative impacts of fire on the ecosystem. It may also be important in order to follow the production of vegetation dead mass and other issues where leaves need to be separated by more ligneous components. Information on the spatial distribution of

biomass weights in terms of fine and coarse types is essential for understanding where fuel is denser or where it is discontinuous and the different fire propagation rates and fire behavior. For example, a considerable part of the study area was depleted of vegetation, hence, following

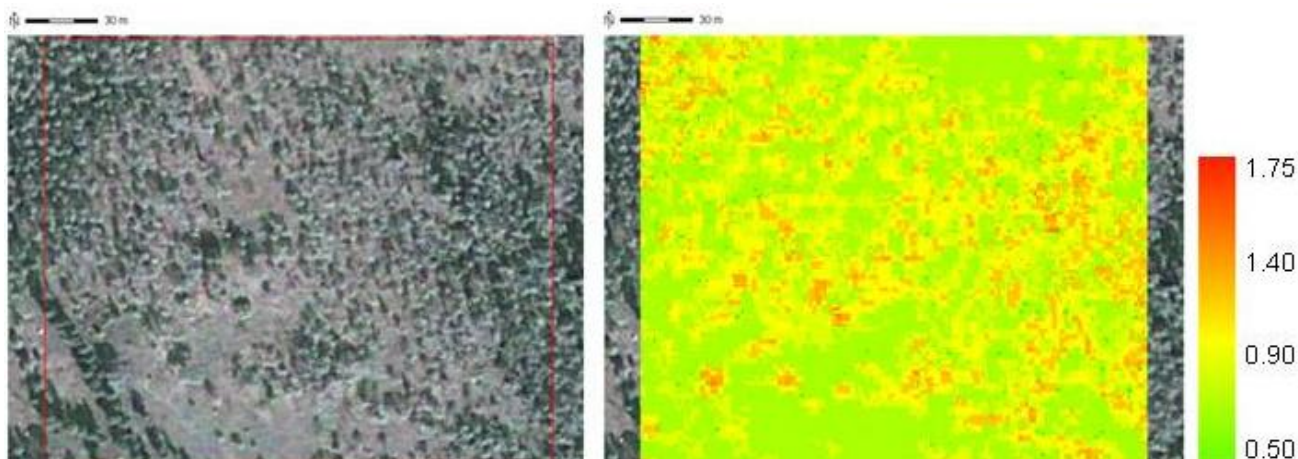


Figure 8. Detail of fine fuel map.

CORINE land cover classification or even a classification based on two-stage hierarchical system (Baraldi et al., 2010; Blonda et al., 2011), it was classified as bare soil where no fuel would have been allocated. With this type of fuel distribution, it is clear that this underestimates the fine particles, the part of the vegetation that contributes most in the propagation of the forest fire. Furthermore, the model developed in this paper, allows the identification of features otherwise lost when the vegetation is represented only by vegetation class labels. This includes orientation of vegetation lines which may favor the spread of fire in a given direction, information that may be useful for hazard management and prevention (Figure 8).

Estimates of biomass and fuel loadings are necessary for many applications in the fields of fire management, ecology, biomass, and bioenergy research. However, the use of destructive sampling to provide these estimates is time consuming and expensive, and furthermore, in protected sites, there are strong restrictions on cutting vegetation, making difficult the acquisition of fuel distribution. In this paper, statistical regression models were used to derive allometric equations for biomass by fuel size categories. Numerous studies have assessed the relationship between vegetation structure and fuel biomass and among which some authors have reported that total biomass in shrubland ecosystem was closely related to height and vegetation cover (Ohmann et al., 1976; Buech and Rugg, 1995; Fogarty and Pearce, 2000; Sağlam, 2008). The correlations of vegetation height provided in our study also coincide with that described by Fernandes (1998) in Portuguese shrubland and by Rittenhouse and Sneva (1977) of Wyoming big sagebrush.

Available fuel biomasses are very important for the spreading and intensity of fire. In this study, average fine biomass was 3.8 t/ha, while average coarse biomass was 47.5 t/ha and average total biomass was 38 t/ha. Similar results were found in some Mediterranean shrublands (Specht, 1969; Icona, 1993; Dimitrakopoulos, 2002; Sağlam, 2008).

A close relationship was found in this study between NDVI and biomass, and consequently the vegetation index gave an indicator of fuel biomass. Numerous studies are now using NDVI as a proxy of vegetation productivity instead of performing direct vegetation assessments (Kerr and Ostrovsky, 2003; Pettorelli et al., 2007; Witemyer et al., 2007). Studies have revealed some discrepancies regarding the shape of these relationships (quadratic, log-linear, linear relationships between NDVI and vegetation biomass (Hobbs, 1995; Gilbert et al., 1996; Schino et al., 2003), respectively). However, due to the variety of statistical approaches used, it remains unclear whether these discrepancies reveal true biological differences, such as differences in plant community characteristics, or methodological concerns.

In this study, the relationships found between NDVI and biomass was used to distribute the biomass values on the area for creating maps of fine and coarse biomass components. Maps, in particular, are essential for computing fire hazard spatially and for assessing fire risk by their use in models simulating fire growth and intensity across a landscape (Keane et al., 2001). Biomass distribution maps account for structural characteristics of vegetation related to fire behavior and fire propagation. Remote-sensing data is becoming the primary method in fuel classification and mapping efforts. Satellite sensors provide digital information that can easily be tied into other spatial databases using GIS analysis, which can be imported into fire behavior and growth models.

Knowing the amount of biomass and other fuel characteristics across a landscape is becoming increasingly important to fire managers as a new generation of fuel and fire management decision support systems come on line. With accurate fuel information, fire managers should be able to make better-informed decisions about ongoing wildland fires and fuel treatments.

The originality of our study resides in the presentation of a new modeling approach that uses NDVI and field vegetation data to create distribution maps of biomass. It

Table 1. Regression models for estimation of shrub biomass by Cd1 (first max diameter of crown shrub), Ca (Area shrubs), Ch (mean height of shrub), Hcover (herbaceous cover).

| Dependent Variables | Predictor variables | Constant and coefficients | F | R ² Adj |
|---------------------------------|---------------------|---------------------------|-------|--------------------|
| Shrub coarse biomass | Cd1 | a: 3.49 b: -11.37 | 18.71 | 0.639** |
| | Ca | a: 1.72 b: -10.83 | 20.53 | 0.661** |
| | Ch | a: 0.01 b: -0.512 | 17.19 | 0.789** |
| Shrub fine biomass | Cd1 | a: 1.59 b: -0.94 | 50.11 | 0.830*** |
| | Ca | a: 0.72 b: -0.18 | 25.83 | 0.713*** |
| | Ch | a: 2.22 b: -4.06 | 150 | 0.937*** |
| Herbaceous biomass ^b | Hcover | a: 0.09 b: -1.07 | 21.29 | 0.833** |

Significance code: ***, P<0.001; **, P<0.01.

Table 2. Regression models for estimation of biomass by NDVI.

| Dependent variables | Predictor variables | Constant and coefficients | F | R ² Adj |
|---------------------|---------------------|---------------------------|------|--------------------|
| Fine biomass | NDVI Mean | a: 3.36 b: -0.24 | 17.4 | 0.701** |
| Coarse biomass | NDVI Mean | a: 51.07 b: -4.04 | 9.01 | 0.612* |

Significance code: **, P<0.01; *, P<0.05)

demonstrates that it is possible to spatially distribute several biomass characteristics by: 1) locally determining allometric relationships, 2) using them in a series of geo-referenced polygons for evaluating local values of the chosen biomass characteristics, 3) determining relationships between radiometric indices and the chosen biomass characteristics using biomass and radiometric characteristics calculated in each polygon, and finally 4) using the radiometric indices to spread the biomass characteristics values in the landscape.

Particularly, in this study, the relationships found between NDVI and biomass were used to distribute the biomass values on the area for creating maps of fine and coarse biomass components. Our model does not require the use of ancillary variables (Riaño et al., 2002) and is based on relationships between fine and coarse biomass and NDVI. Other authors used supervised classification techniques on low resolution imagery (such as Landsat TM) to generate the fuel maps and did not take into

account fine and coarse fuel (Vidal et al., 1994; Vidal and Devaux-Ros, 1995; Burgan et al., 1996; Riano et al., 2002; Rollins et al., 2004; Lasaponara and Lanorte, 2007).

In conclusion, in this study carried out in *Pinus pinaster* dominated sites, we developed a series of regression equations for predicting fine and coarse fuel biomass of species common in a Mediterranean region, mainly *Erica scoparia*. The regression models developed herein are suitable for predicting fuel biomass in similar shrub areas. Local and site-specific fuel biomass data should be used for more reliable fire behavior predictions. Given the range of the data on which the relationships were based, this study provides a valuable contribution to biomass research in general. However, it should be kept in mind that the range of fuel characteristics on which the relationships were based represents the range of conditions under which it is possible to use the relationships generated through this study.

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Short Communication

Differential Resource allocation of black mustard plants (*Brassica nigra* L.) with proximity to black walnut trees (*Juglans californica* L.) in a Southern California Riparian ecosystem

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The invasive forb *Brassica nigra* (black mustard) and the native tree *Juglans californica* (black walnut) are allelopathic species that suppress the growth of neighboring plants. This study evaluates how allocation to root, reproductive, and photosynthetic biomass in *B. nigra* was affected with proximity to *J. californica*. We hypothesized that a joint suppressive effect would lead to lower root biomass allocation in *B. nigra* near *J. californica* due to reduced interspecific competition. Our results indicate that *B. nigra* plants growing near *J. californica* had significantly lower root : total biomass ratios, and provide insights into how to effectively control this invasive species.

Key words: Allelopathy, competition, invasive species biology, principle of allocation, Santa Monica Mountains.

INTRODUCTION

Riparian ecosystems have a history of invasion from alien plants (Richardson et al., 2007). Black mustard (*Brassica nigra* L.) is an r-selected plant species native to Europe and the Mediterranean that has been introduced into the riparian, annual grassland vegetation of Southern California (Bell and Muller, 1973). California black walnut (*Juglans californica* L.) is a low growing hardwood tree endemic to Southern California (Keeley 1990). Both *B. nigra* and *J. californica* are allelopathic plant species that release chemical compounds into the environment which inhibit the growth of other surrounding plant species in the local area (Muller, 1969). The allelopathic properties of *B. nigra* have facilitated invasions of other plant species by inhibiting the annual grassland species of riparian ecosystems in Southern California (Bell and Muller, 1973). Success of invasive annuals can rely on resource allocation by reducing the investment into some

organs in favor of others when interspecific competition for resources is reduced (Cheplick, 2006). Plant resource allocation can be observed by comparing the proportions of total biomass of the reproductive effort (flowers), photosynthetic processes (leaves), and nutrient acquisition (roots).

Understanding ecological variables, including the proximity to another allelopathic species, which might change the patterns of resource allocation of *B. nigra* may lead to effective solutions for controlling the spread of this invasive species (Weston, 2005). The objective of our study was to investigate the changes in resource allocation of an invasive allelopathic plant species, *B. nigra*, within different proximities to a native allelopathic plant species, *J. californica*, by analyzing the biomass ratios of different plant organs. We hypothesized that (1) *B. nigra* allocates fewer resources to nutrient acquisition (roots) within closer

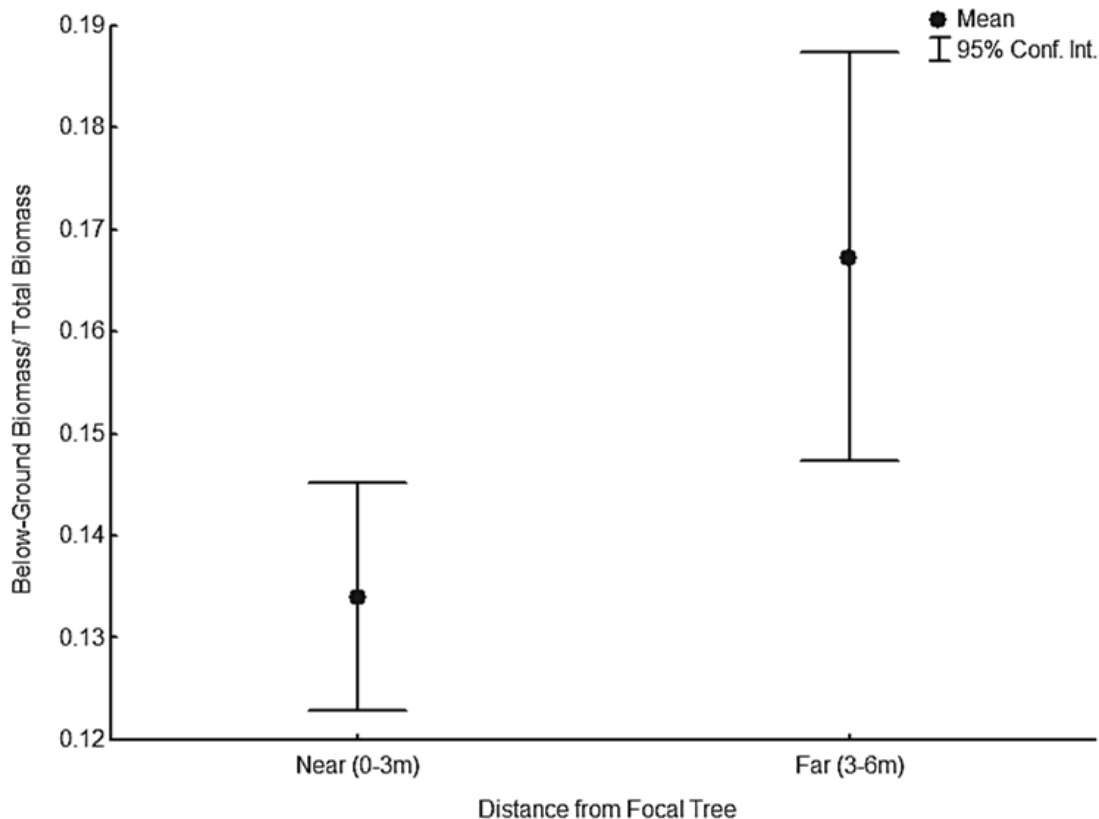


Figure 1. The root biomass ratio of *B. nigra* growing in proximities near and far from focal *J. californica*.

proximity to *J. californica*; (2) *B. nigra* resource allocation to reproductive effort or photosynthetic processes does not change with proximity to *J. californica*, and (3) *B. nigra* total biomass does not change with proximity to *J. californica*, because the combined suppressive effect of both allelopathic species would serve to reduce nutrient competition with other plant species and only impact root biomass without affecting total biomass or favoring any other organ. Alternatively, changes in *B. nigra* total biomass or allocation to other organs would suggest allelopathic suppression by *J. californica*.

MATERIALS AND METHODS

Samples of *B. nigra* were uprooted at Wilacre Park in the Santa Monica Mountains of Southern California on 11 April 2013. Eight focal *J. californica* trees were selected; and across all sites, *B. nigra* was a dominant element of the understory vegetation. We sampled a total of six plants, *B. nigra*, at each focal tree: three plants near (0–3 m) and three plants far (3–6 m). The *B. nigra* tissues were separated into flowers, stems, leaves, and roots before placement into a Labconco 4.5 L Freeze Dryer for desiccation. The distribution of biomass ratios for each of four components (total biomass, root biomass ratio, photosynthetic biomass ratio, and reproductive biomass ratio) of *B. nigra* were evaluated using a Shapiro-Wilks test for normality. Total plant biomass, root biomass ratio, and photosynthetic biomass ratio were compared using a Mann-Whitney U test. Reproductive biomass ratio was compared using a t-test. Sample variances of root biomass ratio were tested using an F-test.

All statistical analyses were conducted using the software package STATISTICA v9.1 (Statsoft, 2010).

RESULTS

The total plant biomass ($W=0.6058$, $P<0.05$), root biomass ratio ($W=0.5734$, $P<0.05$), and photosynthetic biomass ratio were not normally distributed ($W=0.9466$, $P<0.05$). The reproductive biomass ratio was normally distributed ($W=0.9887$, $P>0.05$). The total plant biomass ($U=282.0$, $P=0.9097$), photosynthetic biomass ratio ($U=228.0$, $P=0.2199$), and reproductive biomass ratio ($t=0.4048$, $P=0.6875$) did not differ significantly with distance from the focal *J. californica*. The root biomass ratio ($U=144.0$, $P=0.0031$) differed significantly with distance from the focal *J. californica*. The mean root biomass ratio for near plants ($\bar{X}=0.1340$) was less than for far plants ($\bar{X}=0.1674$, Figure 1). The variance of the root biomass ratio differed significantly with distance from the focal *J. californica* ($F=3.172$, $P=0.0076$, Figure 1). The root biomass ratio of far *B. nigra* had greater variance.

DISCUSSION

The root biomass ratios of *B. nigra* differed significantly with distance to the *J. californica*. There was more root biomass in far plants than in near plants. There was signi-

ificantly greater variation in root biomass ratio in far *B. nigra* than in plants near *J. californica*. The results suggest that *B. nigra* may allocate its resources differently when near and far from the allelopathic *J. californica*. Changes in allocation to *B. nigra* roots with proximity to *J. californica* favor neither flowers (reproductive) nor leaves (photosynthetic), and do not affect total biomass. These results further suggest that while *B. nigra* belowground allocation was impacted, *B. nigra* total biomass and allocation to aboveground tissues was not suppressed by *J. californica* allelopathy.

Allelopathic invasive species invest less in nutrient acquisition in close proximity to other allelopathic species because of overall reduction in competition (Callaway and Ridenour, 2004). Field studies also show effective use of allelopathy in the control of invasive species in non-native habitats (Weston, 2005). We propose that further studies be done to investigate (1) the use of allelopathy to control weedy (r-selected) non-allelopathic invasive species, (2) plants unaffected by allelopathy to control the invasive allelopath *B. nigra*, and (3) the density patterns of *B. nigra* with respect to the impacts of other allelopathic species like *J. californica*.

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Journal of Ecology and The Natural Environment

The background image shows a waterfront town with a canal. On the left, there are colorful buildings with red, blue, and green roofs. A wooden pier runs along the canal, with several boats docked. In the foreground, there is a blue metal bike rack on a paved path next to the canal. A large bush of pink flowers is visible on the right side of the path.

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