Research Article

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The Selection, Testing, and Application of Ecological Bioindicator Birds: A Case Study of the Bale Mountains, Southeast Ethiopia

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ABSTRACT

The interest in using ecological bioindicators - species or higher taxa, whose presence/absence or abundance reflect the abiotic or biotic state of an ecosystem - as cost-effective means of ecological monitoring has been globally increasing. The main aim of this study was to assess if such ecological bioindicator species could be identified within Afromontane grassland avifauna that would be used for monitoring the effects of livestock grazing on ecosystem in the Bale Mountains of Ethiopia. We collected data on birds and vegetation structure along 14 transects each in the light (protected) and heavy (unprotected) grazing sites in June 2014 (referred to as the first sampling period) and in December 2014 (the second sampling period). Then, we (i) initially identified potential bioindicator species for the light grazing site, based on data collected during the first sampling period; (ii) examined relationships between abundance of these bioindicators and level of grazing pressure; (iii) tested the consistency of those initially selected bioindicator species, based on independent data collected during the second sampling period. We used the Indicator Value (IndVal) Analysis method to identify bioindicator species for the light grazing site. Species with significant IndVal>60% were considered as potential bioindicator for the site compared to the heavy grazing site. Six species were initially identified as potential bioindicators from the first sampling period dataset, and four of these species were again consistently identified from the second sampling period. Furthermore, abundance of the bioindicators had significantly declined with increasing grazing pressure, but positively correlated with four habitat variables (i.e. heights of shrub, herb and grass, and cover of shrub). These findings suggest that those consistently identified four species represent suite of reliable bioindicators that can successfully be used for monitoring of changes in habitat structure in the site. Recommendations on how to apply these findings for ecological monitoring are provided.

Keywords: Bioindication, Conservation, Ecological, Monitoring, Afromontane Grassland, Grazing, Indicator Value.

1. INTRODUCTION

Bioindicators are species or group of species, or higher taxa, whose biological or ecological attributes (e.g. presence/absence, abundance, survival rate, reproductive success, etc) readily reflect the abiotic or biotic state of an environment (environmental bioindicators), an ecosystem (ecological bioindicators), or the diversity of taxa (biodiversity bioindicators) (McGeoch, 1998;

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Niemi and McDonald, 2004). Ecological bioindicators are primarily used either to assess the condition of (e.g., as an early-warning system) or to predict trends in state of an ecosystem (Dale and Beyeler, 2001). The premise to use bioindicators for ecological monitoring has come from the fact that most ecosystems are biologically and ecologically highly diverse and complex, making difficult to undertake surveys on tall taxa during monitoring (Noss, 1990). Further, most of such diverse and complex ecosystems are found in developing tropical countries like Ethiopia where there is often lack of resources (funding and expertise), making ecological monitoring activities more difficult (Addisu Asefa et al., 2015a). Consequently, application of the bioindication concept in conservation initiatives has been advocated to be used as a simple and cost-effective means of ecological monitoring to assess the current and predict the future healthiness of ecosystems (Noss, 1990; McGeoch, 1998; Carignan and Villard, 2002; Niemi and McDona, 2004).

Application of the bioindication concept using various biological taxa in conservation programmes have been reported by several authors. For example, Andersen et al. (2002) have used ants as bioindicators in land management; Davis (2001) has used Dung beetles as indicators of change in the forests of northern Borneo; and Kitching et al. (2000) have used moth assemblages as indicators of environmental quality in remnants of upland Australian rain forest. Birds have been also applied as indicators of environmental change (e.g., Morrison, 1986; Temple and Wiens, 1989). Similarly, Vilches et al. (2013) have used plant indicator species of broad-leaved oak forests in the eastern Iberian Peninsula. Despite the globally growing interest in studying and using of biological taxa as bioindicators to detect environmental changes and determine the causes and consequences of such changes on ecosystems (e.g. Kitching et al., 2000; Davis, 2001; Andersen et al., 2002; Vilches et al., 2013), inappropriate selection and application of bioindicators have put under question the utility of the bioindication concept as a conservation tool (Kremen, 1992; Landres et al., 1998; Carignan and Villard, 2002; Manne and Williams, 2003; Urban et al., 2012). Nonetheless, some authors (e.g. McGeoch, 1998) have provided a step-by-step procedure to be followed to select reliable bioindicator taxa that would be successfully applied for ecological monitoring. According to McGeoch (1998), the first step during bioindicator species identification is clearly defining the specific objectives—i.e. what is to be indicated by monitoring the bioindicators? Once the objectives are defined and potential bioindicator species are identified based on *a priori* suitability criteria (for detail on such criteria, see Kremen, 1992; Hiltyand Merelender, 2000; Manne and Williams, 2003), the presence of strong relationships should be confirmed between ecological attributes (e.g. abundance, presence/absence, or reproduction) of such bioindicators and the ecosystem stressors (e.g., grazing disturbances), as well with the ecological variables that they are supposed to indicate (e.g., vegetation cover, height, etc) (McGeoch, 1998). Then after, before recommending or using the candidate bioindicators for ecological monitoring, the consistency of initially selected potential bioindicators should be tested based on data independent from those used for initial identification, for example by re-sampling the same environment under different temporal or spatial conditions (Weaver, 1995; Majer and Nichols, 1998; McGeoch et al., 2002). Finally, to use information derived from bioindicator-based ecological monitoring for effective management decision making purposes, specific recommendations should be provided on how to apply the suite of reliable bioindicator species in ecological monitoring (McGeoch, 1998). In this study, we followed such step-by-step procedure to explore the potential use of birds as ecological bioindicators for monitoring the effects of livestock grazing on Afromontane grassland habitat in the Bale Mountains National Park (BMNP) of Ethiopia.

The Bale Mountains region is recognized as the centre of endemicity and evolution for several biological taxa (Williams et al., 2004; Addisu Asefa, 2011). It is also one of the 69 Important Bird Areas of Ethiopia (EWNHS, 2001). In the northern section of the BMNP is a montane grassland habitat which represents a critical habitat for the endangered Ethiopian endemic mountain nyala (*Tragelaphus buxtoni*), several other ungulates, and (near) endemic and globally threatened bird species such as the Abyssinian Long-claw (*Macronyx flavicollis*) and Rouget's Rail (*Rougetius rougetii*) (Yosef Mamo et al., 2014, Addisu Asefa et al., 2015b). Consequently, most conservation efforts, such as monitoring of illegal livestock grazing, in the Bale Mountains National Park (BMNP) have been concentrated to this area. However, this grassland ecosystem and its associated biodiversity have been threatened mainly by livestock grazing (Stephens et al., 2001; OARDB, 2007; Yosef Mamo et al., 2014). Grazing by livestock causes changes in the vertical and horizontal structural composition of vegetation through a combination of trampling, grazing/browsing, changes in nutrient fluxes and loss of recruitment (McIntyre et al., 2003), and facilitates encroachments of non-native species (Kimball and Schiffman, 2003). Results of similar studies undertaken in the montane grassland of the Bale Mountains have also demonstrated the deleterious effects of heavy grazing on vegetation and, consequently on birds (Yosef Mamo et al., 2014; Addisu Asefa et al., 2015b). Thus, monitoring the impact of livestock grazing, using bioindicators such as birds, on ecosystem of this grassland has been identified as key priority action by the BMNP management (OARDB, 2007). We chose birds in this study because they, among vertebrate groups of animals, have been a primary focus for most terrestrial applications of the bioindication concept (Mazerolle and Villard, 1999; Niemiand MacDonald, 2004). Overall, the reasons for choosing birds as bioindicators are: (a) relative ease of identification, (b) relative ease of measurement, (c) relatively large number of species with known responses to disturbance and (d) relatively low cost for monitoring (Morrison, 1986; Temple and Wiens, 1989; Mazerolle and Villard, 1999; Carignan and Villard, 2002; Niemi and MacDonald, 2004).

The specific objectives of this study were therefore to: (i) identify potential bioindicator bird species for the low grazing (protected) grassland site based on data collected during the first sampling period (in June 2014); (ii) test the responses of bird species that were initially identified as bioindicators of habitat change to both grazing pressure and grazing-induced changes in vegetation structure; (iii) test the consistency, and thus estimate the reliability, of initially selected potential bioindicator species using data collected in the area during the second sampling period (in November 2014); and, (iv) develop predictive models relating abundance (number of individuals of birds recorded along the sampling units) of bioindicators with habitat parameters which the bioindicators are supposed to be indicator for.

2. MATERIALS AND METHODS

2.1. Study Area

The Bale Mountains region is located in the south-eastern highlands of Ethiopia (Fig 1). It is part of the Eastern Afromontane Hotspot Biodiversity area designated by Conservation International (Williams et al., 2004). At the heart of these mountains is the Bale Mountains National Park (BMNP), which is located at about 400 Km southeast of the capital, Addis Ababa (OARDB, 2007). The national park covers an area of 2200 km^2 and ranges in altitude from 1500 – 4377m a.s.l. (OARDB, 2007). To date about 78 species of mammals and 278 bird species have been recorded from the Bale Mountains area; of which 17 mammals and 6 bird species are endemic to Ethiopia (Addisu Asefa, 2007, 2011). The Bale Mountains area is characterized by eight months (March-October) of rainy season and four months (November-February) of dry season (OARDB, 2007). The present study was carried out in the northern montane grassland area which occurs as a central broad flat valley (between altitudes of 3000 - 3150 m a.s.l.) between two mountainous ranges (Fig 1). This grassland has an area of c. 37 km^2, of which ~15 km^2 falls inside the BMNP boundary (hereafter referred to as light grazing site). This site is relatively well-protected from illegal livestock grazing. The remaining area of the grassland falls outside the park boundary and is being used as a communal livestock grazing land by the surrounding local community (hereafter referred to as heavy grazing site) (Fig 1; see also OABRD, 2007). On the average (mean ± S.D.) 1528 ± 86 heads of livestock (cattle and horses) has been reported to use this heavy grazing site every day (Yosef Mamo et al., 2014).

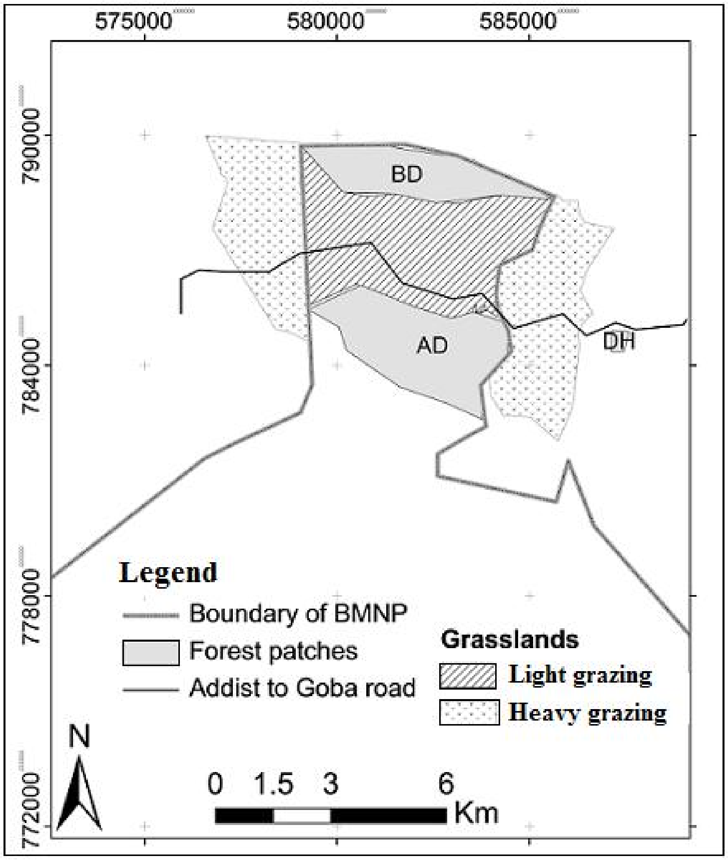

Figure 1. Map of the Bale Mountains National Park (only partly shown) and the light and heavy grazing grassland sites, and the three forest patches found around the grasslands. Abbreviations of the forest patches: DH = Dinsho Hill (BMNP HQs); AD = Adellay; and, BD = Boditti.

The vegetation in the montane grasslands of the Bale mountains is broadly classified in to three types: open grassland (areas covered by short grasses), marsh grassland (characterized by swamp grasses and sedges of *Cyperus* and *Scirpus* genera), and shrubland (covered by bushes of *Artemesia afra* and *Helichrysum splendidum*) (OARDB, 2007). The extent of the open grasslands is ~4 and 12 km^2 and of marsh grassland is 5 and 5 km^2, respectively, in the light and heavy grazing sites. Shrublands (~5 km^2) are completely destroyed in the heavy grazing site and currently occur only in the light grazing site (Hillman, 1986; Yosef Mamo et al., 2015).

2.2. Data Collection

We used our previously published data on abundance and occurrence data of birds in the study area (Addisu Asefa et al., 2015b) both for initial identification and subsequent testing of bioindicator species. Bird data were first collected in June 2014 during the wet season (hereafter referred to as the first sampling period) along systematically established 28 transects (14 each in the light grazing and heavy grazing sites and at a minimum distance of 300m apart) of each 1-km long. The start and end geographical coordinates of each transect were saved in Garmin GPS unit to ensure same transects were repeated during the dry season, which was undertakenin November 2014 (hereafter referred to the second sampling period). We undertook the second sampling work to test whether the species identified as bioindicators from the first sampling period would indeed be consistently appeared to fulfil the selection criteria. According to the recommendations of Weaver (1995), Majer and Nichols (1998), and McGeoch et al. (2002), the dataset to be used for such consistency testing should come from samples taken at different environmental conditions (e.g., sampling the same area during different seasons) compared to the samples taken for the initial bioindicator species identification. Thus, we collected the two datasets during different seasons based on this recommendation, as it would enable us to finally retain only subset of species, among initially identified potential ecological bioindicators, that would be effectively applied for the intended ecological monitoring (McGeoch et al., 2002). During both sampling periods, birds were counted within 50 m width on both sides of each transect. Transects were surveyed randomly and only one transect was surveyed per day. Bird surveys were undertaken early in the morning (between 07:00-10:00) when birds are thought to be more active, while slowly walking at speed of ~2 km hr^{-1}. Aerial feeders (raptors, swallows, and swifts) and wetland birds were not recorded as the primary objective of the study was on terrestrial birds. For list of species recoded in each site during each sampling period, see table 1 in Addisu Asefa et al. (2015b).

Data on six habitat parameters (heights and percentage covers of three plant functional forms [shrub, herb, and grass]) were also recorded within four 10 m × 10 m quadrates established along each transects at 200 m distance intervals (Addisu Asefa et al., 2015b).To determine heights of each plant functional form, four different measurements were taken at each quadrate (totalling to 16 measurements per transect) using a labelled measuring stick and cover was visually estimated (Newton, 2007).

2.3. Data Analysis

A given bird species was considered to be potential bioindicator of habitat condition and used for long-term monitoring of the impact of livestock grazing on vegetation structure in the light grazing site of our study area if it: (i) fulfils *a priori* suitability selection criteria, (ii) show clear response to disturbance, (iii) consistently fulfils again the *a priori* suitability selection criteria based on independent dataset collected from same sites during the second sampling period (in different season) and (iv) shows strong positive correlations with the habitat variables which it was supposed to be indicative(see also Kremen, 1992; McGeoch, 1998; Hilty and Merelender, 2000). We tested each of these assumptions in a step-by-step fashion as follows.

2.3.1. Initial Identification of Bioindicators

We initially identified potential bioindicator bird species for the light grazing sites, based on data collected during the first sampling period (i.e. wet season data), in two steps process. First, we used Dufrêene and Legendre's (1997) Indicator Value (IndVal) Analysis Method. This method assesses the degree (expressed as a percentage) to which each species fulfills the criteria of specificity (uniqueness to a particular site) and fidelity (frequency within that habitat type) in a site compared with the other sites (McGeoch and Chown, 1998) and provides indicator values (IndVal) of each species for each site compared. The formula for the IndVal is sated as follow: $\text{IndVal}_{ij} = (\text{Specificity}_{ij} \times \text{Fidelity}_{ij}) \times 100$,where IndVal_{ij} is the Indicator Value of species; '*i*' for site '*j*'; Specificity_{ij} is the proportion of the number of individuals (abundance) of species '*i*' that are in a '*j*' type of site; and Fidelity_{ij} is the proportion of sites of type '*j*' with species '*i*' (Dufrene and Legendre, 1997).

Species-specific indicator values were computed in IndVal software (Dufrêne and Legendre, 1997) using the species abundance matrix from each site as input. Dufrene and Legendre's (1997) random reallocation procedure of samples among sample groups was used to test the significance of the IndVal measures for each species. Different authors have been using varying extent of species' indicator values (IndVals) as a minimum threshold value for potential bioindicator species selection criteria. For example, Dufrene and Legendre (1997) and De Cáceres et al. (2010) used IndVal ≥25%, and van Rensburg et al. (1999) used IndVal >=70%. For the purpose of this study, we regarded those species with maximum significant IndVals >60% in a given site as potential bioindicator species for that site.

Then, we used additional *a priori* suitability criteria to refine the selection processbecause the IndVal analysis approach provides information only on some aspects of bioindicator species' niche (e.g. habitat specialty and fidelity) (Dufrene and Legendre, 1997; McGeoch and Chown, 1998), but there are other additional properties - i.e., traits/characteristics which a given potential bioindicator taxa should possess if it is to be considered as reliable that such species should also fulfill to be regarded as a reliable bioindicators (Kremen, 1992; Hiltyand Merelender, 2000; Manne and Williams, 2003). Among such species-specific properties which we used as additional *a priori* suitability selection criteria were whether the potential bioindicator species: i) has a clear taxonomic status, i) is a non-migrant, with wide distribution (national, regional or global distribution), iii) is easy to find and measure (i.e. high abundance) (McGeoch, 1998; Hiltyand Merelender, 2000). Species identified as potential bioindicators based on the IndVal analysis were therefore refined using these *a priori* suitability criteria based on species-specific information obtained from Redman et al. (2009) and Bird Life International (2015).

2.3.2. Relationships between Grazing Level, Habitat Structure and Bioindicators

We tested the responses of both habitat variables and bioindicators (identified for the light grazing site from the first sampling period) to grazing pressure. We used the summed abundances of all species identified as potential bioindicators, rather than individual species' abundance, as an input for these analyses, following De Cáceres et al. (2010). Using the summed abundance is advantageous to minimize dependence on individual species and to improve confidence by basing conclusions on a wider array of responses than on response of individual species (Hilty and Merenlander, 2000; McGeoch et al., 2002). These analyses were undertaken using Generalized Linear Mixed Models (GLMMs) with normal distribution and identity link function in SPSS version 20 (IBM Corporation, 2001). In the models, vegetation attributes (height and cover), and abundance of the bioindicators were entered as dependent variables, while grazing level (light vs heavy grazing) as fixed factor and site (light vs heavy grazing sites) identity as a random factor to account for potential independence of transects within a site (Quinn and Keough, 2002). We also examined the relationship of habitat variables with abundance of the bioindicators within the light grazing site using a linear regression model. As most habitat variables had showed co-linearity between each other, we undertook PCA and used the first two component axes that explained 82% of the variation in the dataset for the regression modelling (for detail on the correlation between each pair of the variables and between them and the PCA components, see Appendix A and B).

2.3.3. Testing Consistency of the Bioindicators

Using independent data collected during the second sampling period (in November 2014) from same transects along which the first dataset was collected, we tested the consistency of species initially selected as potential bioindicators from the first sampling period. The IndVal analysis method and the other additional suitability criteria used for the initial selection (see Bioindicator selection above) were followed to identify bioindicator species from this independent dataset collected during the second sampling period. Those species that were initially selected from the first sampling period data as potential bioindicators were considered to be reliable bioindicators if they attained again IndVals of >60% based on the dataset of the second sampling period. Thus, the selection process was refined whereby only a subset of species that showed consistency across the two sampling periods were finally retained as robust bioindicators.

2.3.4. Application of Bioindicators for Ecological Monitoring

To assess the potential application of species- those species that showed consistency across sampling periods and thus were finally selected as reliable bioindicators- in ecological monitoring, we tested the predictive power of the bioindicators for each of the six habitat variables in the light grazing site. We then developed predictive models relating each habitat variable that showed strong positive correlation with abundance of the bioindicators. These analyses were undertaken using simple linear regression models, where each habitat variable was treated separately as response variables, while average (from the two sampling periods) of the summed abundance of those four bird species as predictor. We assumed that strong and significant correlation between a given habitat variable and abundance of the bioindicators in the light grazing site (protected area) implies that the bioindicators will be used confidently for long-term monitoring of that habitat variable in the site.

3. RESULTS

Overall, 33 species (24 and 25 species form the light and heavy gazing sites, respectively) were recorded in the study sites across the two sampling periods. Number of species recorded during the two sampling periods was almost similar between sites, but was 27% fewer during the second sampling period than during the first sampling period in the light grazing site (Table 1). Nonetheless, 32% fewer individuals were recorded across sites during the second sampling period compared to the first sampling period. Significant difference (P<0.05) within sites in number of individuals between the two sampling periods was revealed only for the heavy grazing site; bird individuals were 3% more in the light grazing site but 47% fewer in the heavy grazing in the second sampling period (Table 1).

Table 1. Number of bird species and individuals recorded during the first and second sampling periods (S.P.) in the light and heavy grazing sites (Addisu Asefa et al., 2015b).

	No. species			***No. individuals***		
	First S.P.	***Second S.P.***	***Total***	***First S.P.***	***Second S.P.***	***Total***
Light	22	16	24	617	639	1256
Heavy	21	19	25	1511	799	2310
Total	26	23	33	2128	1438	3566

3.1. Initial Identification of Potential Bioindicators

Based on the IndVal criteria (i.e. IndVal >60%), six species were identified as potential bioindicators from the dataset collected during the first sampling period for the light grazing site (Table 2a). These included: Moorland Chat (*Cercomela sordid*), Common Stone Chat (*Saxicola torquatus*), Streaky Seedeater (*Serinus striolatus*), Common Waxbill (*Estrilda astrild*), Rouget's Rail (*Rougetius rougetii*) and Winding Cisticola (*Cisticola galactotes*). Similarly, five species, including Ethiopian Siskin (*Serinus nigriceps*), Ground Scraper Thrush (*Turdus litisitsirup*) and Wattled Ibis (*Bostrychia carunculata*), Thekla Lark (*Galeridatheklae*) and Cape canary (*Serinus canicollis*), were identified from the heavy grazing site (Table 2b). All these 11 species had also fulfilled the additional *a priori* suitability selection criteria, thus were considered as potential bioindicators for their respective site.

Table 2. Indicator values of bird species selected as potential bioindicators selected during the two sampling periods for the light and heavy grazing sites in the Bale Mountains Afromontane grassland habitat. (Species with significant IndVal >60% were then regarded as bioindicator species, and those species that attained these IndVal criteria during both sampling periods were considered as reliable bioindicators.)

Species	***First sampling period***	***Second sampling period***
(a) light grazing site		
Morland Chat (*Cercomela sordid*)	62.73	65.19
Common Stone Chat (*Saxicola torquatus*)	85.71	89.76
Common Waxbill (*Estrilda astrilda*)*	58.81	21.43
Rouget's Rail (*Rougetius rougetii*)*	78.57	14.29
Streaky Seedeater (*Serinus striolatus*)	84.57	60.52
Winding Cisticola (*Cisticola galactotes*)	72.33	60.86
(b) heavy grazing site		
Ethiopian Siskin (*Serinus nigriceps*)*	67.21	17.89
Ground Scraper Thrush (*Turdus litisitsirup*)	93.48	73.93
Thekla Lark (*Galerida theklae*)*	80.33	48.52
Wattled Ibis (*Bostrychia carunculata*)	78.57	85.71
Cape Canary (*Serinus canicollis*)*	84.55	2.04

Note: * = species discarded from the final selection.

3.2. Relationships of Grazing Level with Bioindicators and Vegetation Parameters

Of the six vegetation variables considered in this study, only three (shrub height and cover and grass height) showed significant differences between the light and heavy grazing sites (ANOVA, in all cases, $F_{1,26}$ = 6.516-32.415, P <0.01). All these variables were in greater values in the light grazing site compared to the heavy grazing site (Fig 2). Similarly, abundance of the bioindicators -i.e. summed abundance of the set of species identified from the first sampling period- was significantly higher in the light grazing site compared to the heavy grazing site (mean (SE) abundance, light grazing site = 24.43 (1.65); heavy grazing site = 6.71 (1.04); $F_{1,26}$ = 82.496, P <0.001). Regressing the summed abundance of these bioindicators against the first two PCA axes scores showed significant relationship (ANOVA, $F_{2,25}$= 43.957, P <0.0001); they explained 77.9% of the variation in abundance of the bioindicators. However, only the first PCA component (which was positively correlated with shrub height and cover and grass height, but negatively with grass cover) showed significant regression coefficient estimate(PCA1: B = 9.643, P <0.0001; PCA2: B = 0.895, P >0.05) with the bioindicators' abundance.

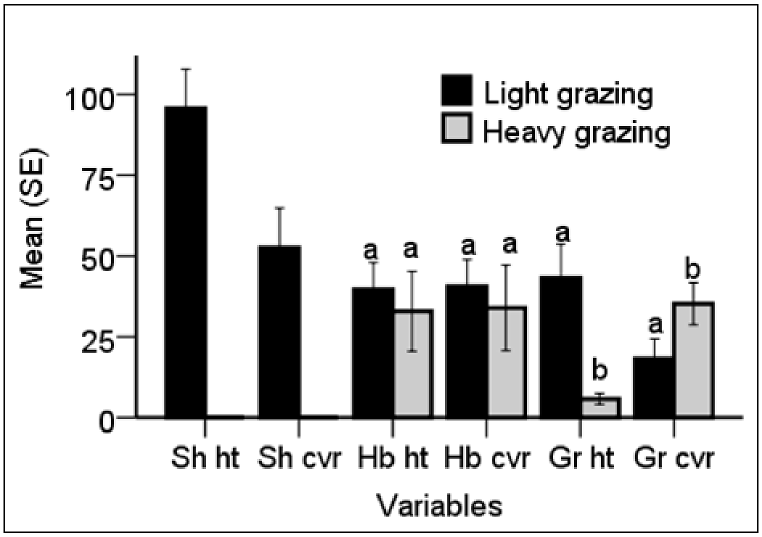

Figure 2. Mean (SE) values of the six habitat variables in the light and heavy grazing sites. Means with different letters are significantly different at P <0.05). (Variable abbreviations: Shht = shrub height, Shcvr = shrub cover, Hbht = herb height, Hbcvr = herb cover, Gr ht = grass height, Gr cvr = grass cover).

3.3. Consistency of the Bioindicators

From the independent dataset collected during the second sampling period (i.e. dry season data), four species were identified as potential bioindicators in each site, based on the IndVal analysis approach (Table 2a and b). However, two [i.e., Red-throated Pipit (*Anthuscer vinus*) and Yellow Wagtail (*Motacilla flava*)] of the four species identified for the heavy grazing site were migrants. Both species were discarded from potential bioindicator species list. Thus, the four species identified for the light grazing site (Alpine Chat, Common Stone Chat, Streaky Seedeater and Winding Cisticola) and the two non-migrant species (Ground Scraper Thrush and Wattled Ibis) identified for the heavy grazing site were retained as potential bioindicators of each respective site and compared with species identified during the first sampling period. Despite the lower number of potential bioindicator species identified during the second sampling period, all the four species identified for the light grazing site and the two species for the heavy grazing site were among those species that were initially identified from the first sampling period (Table 2a and b). These results indicate that three-fourth of species initially identified for the light grazing site, but only two-fifth of the species identified for the heavy grazing site, were found to be consistent across the two sampling periods. These consistently selected species were thus regarded as robust bioindicators of habitat condition in the site for which they were supposed to be bioindicators.

Table 3. Results of regression analyses between each of the six vegetation variables and abundance of the finally selected reliable bioindicator species for the light grazing site (In all cases, df = 1 and 12).

Dependent variable	***R***	***ANOVA***			
		Source	***MS***	***F***	***p***
Shrub height	0.832	Regression	4576.448	4576.448	P <0.001
		Residual	2027.909	168.992	
Shrub cover	0.632	Regression	2729.52	7.962	P <0.05
		Residual	342.826		
Herb height	0.847	Regression	2288.057	30.468	P <0.001
		Residual	75.096		
Herb cover	0.039	Regression	3.859	0.018	P =0.895
		Residual	212.678		
Grass height	0.921	Regression	2879.156	67.407	P <0.001
		Residual	42.713		
Grass cover	0.501	Regression	430.409	4.016	P =0.068
		Residual	107.162		

3.4. Application of the Bioindicators for Ecological Monitoring

Although we identified bioindicators and tested their consistency for both the light and heavy grazing sites, we focused analysis of the application of the bioindicators for ecological monitoring only on those bioindicators selected for the light grazing site. We decided this because our goal was to propose the use of birds as bioindicators for long-term monitoring of habitat change in light grazing site (protected site). Our results showed that mean abundance of the bioindicators i.e. average (from the two sampling periods) of summed abundance of the four species finally retained as reliable bioindicators was significantly higher in the light grazing site compared to the heavy grazing site [mean (SE) abundance, light grazing site = 15.00 (1.08); heavy grazing site = 4.29 (2.16); $F_{1,26} = 76.994$, $P < 0.001$]. Results of regression analyses showed that four (heights of shrub, herb and grass, and cover of shrub) of the six habitat variables had significant positive relationships with the average of summed abundance of the four consistently selected bioindicator species in the light grazing site (in all cases, ANOVA: $F_{1,12}$ = 7.962-4576.448, P <0.05; Table 3). The coefficients of determination (R) of regression models relating each of these four vegetation parameters against the abundance of the bioindicators in this site were significantly high and ranged between 0.632-0.921 (Table 3). These suggest that 40-85% of the variations in these vegetation variables in the site were explained by variations in abundances of the bioindicators. Estimated parameters (regression slopes) for these four vegetation variables were significant and indicted positive relationships between habitat variables and abundance of the bioindicators (Table 4). These results, therefore, suggest that the four species finally retained as reliable bioindicators for the light grazing site can readily and successfully be used for long-term monitoring of trends in vegetation structural composition in the site.

Table 4. Predictive regression models relating the values of four vegetation variables with abundance of the bioindicators (BI_{abun}) for the light grazing site in the northern BMNP. Abundance was modelled as average number of individuals of the four species finally selected as reliable bioindicators from the two sampling periods, and vegetation height was expressed in cm and cover expressed in percentage.

Dependent variable	***Equation***
Shrub height (SHH)	$SHH = 25.845\ (13.882) + 4.663\ (0.896) * BI_{abun}$
Shrub cover (SHC)	$SHC = -1.443(19.772) + 3.601(1.276) * BI_{abun}$
Herb height (HH)	$HH = -9.811\ (9.254) + 3.297(0.597)* BI_{abun}$
Grass height (GH)	$GH = -10.332\ (6.979) +3.698(0.450) * BI_{abun}$

4. DISCUSSION

In this study potential bioindicator bird species were initially identified from a first sampling period based on *a priori* established selection suitability criteria, and the robustness (consistency) of these initially identified species were tested on independent dataset collected from same sites during the second sampling period. Sets of species were found to be robust bioindicators, i.e. had consistently fulfilled all the bioindicator selection criteria during the two sampling periods in a site. Whereas, certain initially identified potential bioindicator species showed a wide variation in their indicator values (IndVals); these species were therefore considered to be unreliable and thus were discarded from the final suite of species. This testing process has led to refine the selection process by retaining only subset of species that were found to be robust bioindicators and improved the confidence with which the final suite of species may be regarded as reliable bioindicators (McGeoch et al., 2002). Overall, similar to reports of several authors around the globe (e.g. Kitching et al., 2000; Andersen et al., 2002; McGeoch et al., 2002; Vilches et al., 2013), our results provide additional insights into the potential application of the bioindication concept in biodiversity conservation programmes. Our findings, in particular, support previous works of many authors (e.g. Morrison, 1986; Temple and Wiens, 1989) on the application of ecological bioindicator birds as a simple and cost-effective means of monitoring ecosystem changes.

Several statistical methods (e.g. TWINSPA, IndVal, Ordination methods such as Correspondence Analysis etc; for detail see Dufrêne and Legendre, 1997) have been developed to identify bioindicator species that characterizes group of samples/sites that share similar ecological characteristics. Among such methods, the IndVal approach has been the most popular and recommended method for indicator species selection (for detail on the performance of the different approaches see, Dufrêne and Legendre, 1997; McGeoch and Chown, 1998; McGeoch et al., 2002; De Cáceres et al., 2010). However, in views of some authors (e.g. Hilty and Merenlander, 2000) the IndVal approach itself, despite several variants have been developed to overcome such issues, still suffers from some shortcomings, especially if the selection criteria relies solely on the degree of species' IndVals and the objective of the selection is to consider species for bioindicator-based ecological monitoring. Although assessment of species' local abundance and frequency of occurrence (using the IndVal method) is the foremost priority step in ecological bioindicator selection, other species-specific traits that influence their utility as reliable bioindicator should also be accounted for. For example, relying on a migratory species selected, by virtue of its being attaining high degree of IndVal, as bioindicator-based ecological monitoring may lead to erroneous conclusions and wrong management decisions to be made (Hilty and Merelender, 2000; Manne and Williams, 2003). Further, the choice of the minimum threshold IndVals should be achieved by a species to be considered as bioindicator is usually arbitrarily defined based on authors' judgement (De Cáceres et al., 2010). Therefore, in addition to the IndVal criteria, species-specific life-history traits, ecological specializations (diet/habitat) and other relevant traits should be used as secondary selection criteria to refine the selection process. Following such two-step process is essential to retain only suite of reliable species (those species that show consistency in indication power across sampling periods) for effective application of the bioindication concept, thus is advocated for all studies concerned with bioindicator identification and application.

In bioindication studies aimed for ecological monitoring, most methods used for bioindicators selection relies on quantitative (e.g. abundance) and/or qualitative (frequency of occurrence) data collected on species of assemblages, but data collected on such attributes (especially for mobile animals like birds) in a given site could be a result of chance event or sampling errors. Thus, before developing recommendations for their application in ecological monitoring programs, whether attributes of the bioindicators are strongly and significantly related to environmental stressors (e.g. grazing disturbances) and/or with the ecological variables which the bioindicators are supposed to be indicatives should be confirmed (McGeoch, 1998). As was true in the present study, in addition to its importance to make predictions of future states in some vegetation structures (see discussion below), this testing process improves more the confidence with which the selected potential bioindicator species would be further considered to be appropriate for ecological monitoring (Majer and Nichols, 1998; McGeoch, 1998). However, some authors (e.g. McGeoch, 1998; McGeoch et al., 2002) still suggest that—because of species-specific differential responses to variations in spatio-temporal environmental conditions—achieving significant relationships between attributes of potential bioindicators and ecosystem in a given site alone may not be sufficient enough to reliably apply such species for monitoring purposes. Therefore, set of initially identified potential species should only be considered as reliable bioindicator if they show consistency—i.e. selected again as bioindicator species—when tested based on data independent from those used for initial identification, for example by re-sampling under different temporal or spatial conditions (Weaver, 1995; Majer and Nichols; 1998, McGeoch, 1998; McGeoch et al., 2002). In this study, such consistency testing has enabled to finally propose subset of species, among initially identified potential ecological bioindicators, that would be effectively applied for monitoring the effects of livestock grazing on Afromontane grassland habitat in the BMNP of Ethiopia. Thus, as suggested by McGeoch et al. (2002), our sampling in different seasons was appropriate as it helped us discard inappropriate species and retain only the 4 species that attained high IndVal during both seasons.

Overall, there are, at least, five main reasons why the finally selected suite of bioindicator species would be reliably used for ecological monitoring in the light grazing site of our study area. First, all the four species occur abundantly across their ranges (Bird Life International, 2015), suggesting that the high likelihood of sampling them during subsequent survey periods. Second, all these species can easily be distinguished from their co-occurring similar species (for such species, see Redman et al., 2009), making them easily identified both by non-bird specialists (with little training inputs) and specialists during monitoring work. Third; their abundance was significantly different between the light and heavy grazing sites, indicating their sensitivity or responsiveness to disturbance. Fourth, population abundance of the bioindicators showed strong positive correlations with the habitat variables which they are proposed to indicate. Finally, they were found to consistently fulfill the selection criteria across the two sampling periods, which indicates their high roboustness to effectively indicate future habitat changes.

Heavy livestock grazing in the Afromontane grassland habitat of the BMNP has resulted in reduced cover and height of shrub and height of grasses (Yosef Mamo et al., 2014; Addisu Asefa et al., 2015b). These plant functional groups have been considered as valuable resources for the maintenance of viable population sizes of several wild ungulate species (Yosef Mamo et al., 2015). For example, the two dominant shrub species (*A. afra and H. splendidium*) in the light grazing site are key hiding and foraging resources for the local and global conservation significant wild mammal, mountain nyala (Befekadu Refera and Afework Bekele, 2002; Yosef Mamo et al., 2012). While, tall grasses provide similar functions for the grassland specialist grazer species, such as bohor reedbuck (Bezawork Afework et al., 2009). Consequently, one of the main management objectives of the BMNP has been to reduce the impact of livestock grazing on the extent and structure of shrub and grass vegetation in the grassland ecosystem, and birds were proposed as bioindicators to monitor of habitat change in the area (OARDB, 2007). Our study therefore directly fits to the conservation management objective of the BMNP, and as such the information obtained and the conclusions drawn from this study will contribute to informed management of the light grazing site of the montane grassland in the BMNP. Applying bioindicator-based long-term ecological monitoring in the site will also compliment information obtained from the on-going ecological and threats monitoring programme in the BMNP (Kinahan, 2010).

In order to practically use the proposed final suite of bioindicators for ecological monitoring in future, we recommend that these species should be counted following same procedures and time of the year—either in June or November, or in both months—used in the present study. Counting should be done, depending on availability of resources needed to undertake the survey, once every year or two years. Then, by inserting the new abundance values of the bioindicators obtained from successive survey periods into the equations provided on table 4, one can estimate the status or trends in the state of each habitat variable. In addition to understanding trends in the state of the habitat variables, information derived from such monitoring activities would help managers know trends in the environmental stressor (i.e. grazing) and decide on what management actions should be taken to mitigate the stressor and its impact on the ecosystem.

5. CONCLUSION

From the study, it is demonstrated that how to: (i) select reliable ecological bioindicator bird species; (ii) use birds for scientifically rigorous bioindicator-based ecological monitoring programme in a given area; and (iii) transform information derived from such bioindicator-based ecological monitoring programs into direct practical application (i.e., for making informed conservation management decision). In addition, the results provide valuable information for effective management of the MBNP. The methods followed in this study can serve as a showcase which can be adopted by researches interested in the study and application of bioindicator-based ecological monitoring systems in protected areas.

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Appendix A. Pearson correlation coefficients between each pair of the six habitat variables.

	Shrub cover	***Grass height***	***Grass cover***	***Herb height***	***Herb cover***
Shrub height	0.808	0.908	-0.939	0.28	0.262
Shrub cover		0.774	-0.728	0.191	0.163
Grass height			-0.423	0.36	0.302
Grass cover				-0.369	-0.481
Herb height					0.732

Appendix B. Correlations of the six habitat variables with the first two Principal Component Axes (PCA).

Variable	***PCA1***	***PCA2***
Shrub height	0.876	-0.341
Shrub cover	0.858	-0.410
Grass height	0.866	-0.271
Grass cover	-0.772	-0.074
Herb height	0.569	0.710
Herb cover	0.568	0.746