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SHORT COMMUNICATION

Assessing spider diversity in grasslands – does pitfall trap color matter?

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Abstract. We analyzed effects of pitfall trap color on spider catches using four different pitfall trap colors (white, yellow, green, brown). For each color, we installed 17 pitfall traps at two grassland sites, respectively, and sampled 77 species from 6,202 individuals. Number of species showed no significant differences but Shannon- and Simpson-diversity were significantly higher in green and brown traps while number of individuals increased in white ones. Species inventories were not complete in the different pitfall trap colors but species accumulation increased significantly slower in white and brown traps. Trap color significantly affected hunting type with ground hunters being associated with bright and web-builders associated with dark colors. Attractiveness of different trap colors may arise due to differences in biological preconditions, albedo and microclimate which in turn can affect diversity and community structure of spiders. Trap color has a significant impact on spider catches and should be considered when planning surveys. We recommend the use of a combination of white and brown (or transparent) pitfall traps to gain complete and diverse species inventories in spiders.

Keywords: Araneae, by-catch, capture efficiency, functional traits, sampling design

Pitfall trapping is one of the most commonly used methods to sample ground-dwelling arthropods. The method is easy to use, time-efficient, associated with low costs and suitable for studying the occurrence and relative abundance of ground-dwelling arthropods (New 1999). Moreover, pitfall trap catches are rich in both species and individuals (Spence & Niemelä 1994) and therefore yield reliable data for a broad range of ecological and biological studies (Sonoda et al. 2013; Corcuera et al. 2016; Yekwayo et al. 2016).

However, capture efficiency of pitfall traps depends strongly on properties such as trap size and the fluid employed (see Brown & Matthews 2016 for a comprehensive review). Interestingly, effects of pitfall trap color have rarely been investigated (but see Buchholz et al. 2010). This could be a drawback, since colored traps could either attract or deter specific taxa as it is known for other types of traps such as pan traps (Heneberg & Bogusch 2014; Moreira et al. 2016). Buchholz et al. (2010) observed that brightly colored pitfall traps (white, yellow) caught significantly more spiders and beetles than more inconspicuous colors such as brown and green. This is of concern, since most studies on epigeal invertebrates use white-colored plastic cups (e.g., Schirmel et al. 2010; Kataja-aho et al. 2016; Meriste et al. 2016) or transparent glass jars (then perceived as brown or soil color) (e.g., Negro et al. 2009; Sadler et al. 2006; Buchholz & Hartmann 2008), thus yielding significantly different catch sums. However, since these results were merely based on individual sums rather than on diversity, it should be valuable to provide a more detailed analysis on species and functional level. Based on the data of Buchholz et al. (2010), we therefore evaluated possible effects of pitfall trap colors (four colors; two dark-shaded, two bright-shaded) on (1) alpha-diversity, (2) species composition of spider communities, and (3) life-history traits.

The study was conducted close to the city of Münster (51°57′46.6″N, 7°37′43.3″E) in North Rhine-Westphalia, Germany. The sub-oceanic climate in this region has a mean annual temperature of 7.9°C and an annual precipitation of 758 mm (Landesanstalt für Ökologie, Bodenordnung und Forsten NRW, 2005). Two sites with a homogeneous vegetation structure were selected that consisted of (1) a sparsely vegetated, dry grassland (Corynephoretum: nutrient-poor grassland on inland dunes; coverage of herbaceous plants [CH] = 20%, height of herbaceous plants [HH] = 15 cm), and (2) a densely vegetated grassland site (Lolio-Cynosuretum: mesotrophic grasslands; CH = 100%, HH =50–60 cm). In total, 68 colored pitfall traps made of plastic jars and filled with a 3% formalin solution and detergent were set. The traps with a diameter of 9 cm and a height of 12 cm were brown, green, white, or yellow and arranged in a grid in rotational order (white–yellow–green–brown, yellow–green–brown–white, green–brown–white–yellow, brown–white–yellow–green). The distance between the traps was 5 m. At the dry grassland site, 24 pitfall traps (6 traps per color) were set and 44 at the densely vegetated grassland site (11 traps per color) (see Buchholz et al. 2010 for more details). These traps were used to catch spiders from 24 April to 6 June 2009 and emptied fortnightly.

Spiders were preserved in ethyl alcohol and determined using standard references (Roberts 1998). For all subsequent analyses, we took into account only adult specimens. To express alpha-diversity, we calculated the observed species numbers and Shannon and Simpson diversity indices.

To assess whether there were significant differences in alpha-diversity among the four pitfall colors (explaining variable: pitfall trap color), we used generalized linear models (GLM) and Holm-Sidak post-hoc tests for pairwise comparisons. Furthermore, species accumulation curves have been calculated with 10,000 permutations to evaluate the completeness of species inventories for each pitfall trap color. We tested differences in curve growth applying a repeated measures ANOVA and Holm-Sidak pairwise-tests. For this, we took data series for each curve and calculated growth between each data point resulting in sixteen growth values per curve.

To determine whether pitfall trap color affected species composition, we applied permutational multivariate analysis of variance (R function: adonis in VEGAN package) (10,000 permutations). Finally, we calculated lightness association for each species. First, we referred to the HSL categorization of colors (Wyszecki & Stiles 2000) and took the lightness values for each color (white = 100%, yellow = 50%, green = 34%, brown = 31%). Second, we calculated Pearson correlation coefficients to express lightness association of spider species. Significance levels of correlations were computed using permutations tests (number of permutations = 99999). Based on Pearson correlation

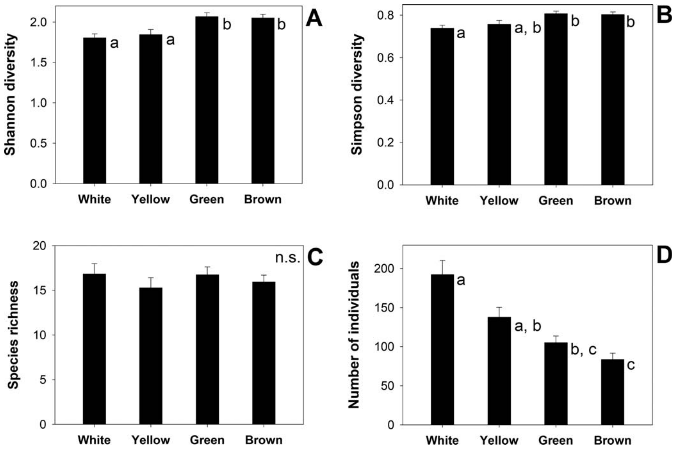

Figure 1.—Alpha-diversity of grassland assemblages differed among pitfall trap color in terms of Shannon- (**A:** GLM, F = 6.7, df = 3, P = 0.0009) and Simpson-diversity (**B:** GLM, F = 5.5, df = 3, P = 0.003), species richness (**C:** GLM, n.s.) and number of individuals (**D:** GLM, F = 15.5, df = 3, P < 0.0001). Pairwise differences (Holm-Sidak, P < 0.05) are presented with lower case letters; values with the same letter are not significantly different.

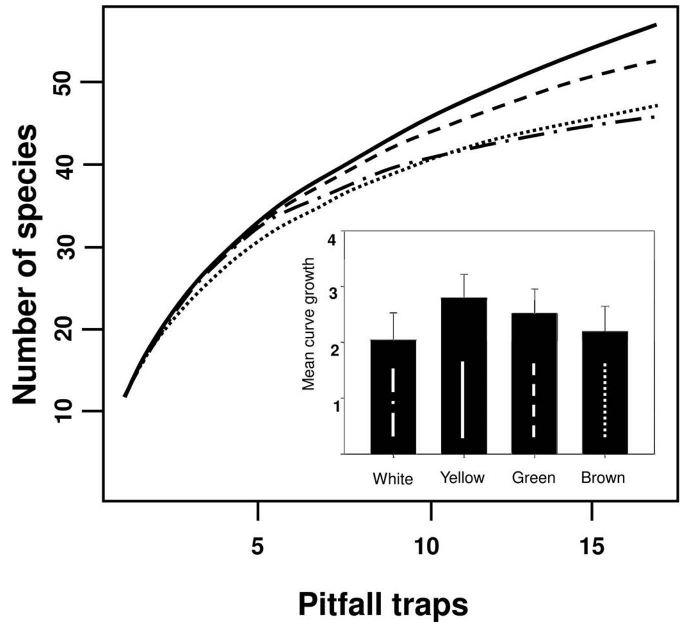

Figure 2.—Sample-based species accumulation curves of the different pitfall trap colors (white = dot-dashed line, yellow = line, green = dashed line, brown = dotted line). X-axis = number of traps. Growth of accumulation curves differed significantly among pitfall trap colors (repeated measures ANOVA, F = 124.9, df = 3, P < 0.001; all pairwise comparisons are significant with P < 0.01).

coefficients, we ran further GLMs to assess whether life-history traits (hunting mode and daily activity) had different light associations. All statistical analyses were performed using the R software environment (version 3.3.1, R Core Team 2016).

A total of 6,202 spiders belonging to 77 species were determined (grassland = 54, dry grassland = 37) (See appendix S1 online at http://dx.doi.org/10.1636/JoA-S-16-062.s1). Most abundant species in grassland sites were *Pardosa prativaga* (L. Koch, 1870) (*n* = 1,962), *Pardosa amentata* (Clerck, 1757) (*n* = 1,417), *Pachygnatha degeeri* Sundevall, 1830 (*n* = 489), and *Pardosa pullata* (Clerck, 1757) (*n* = 468). In dry grasslands, most abundant species were *Xerolycosa miniata* (C. L. Koch, 1834) (*n* = 269), *Pardosa lugubris* (Walckenaer, 1802) (*n* = 108), and *Drassyllus pusillus* (C. L Koch, 1833) (*n* = 16).

Although the number of species showed no significant differences among pitfall trap colors, Shannon-diversity (GLM, F = 6.7, df = 3, *P* = 0.0009) and Simpson-diversity (GLM, F = 5.5, df = 3, *P* = 0.003) were significantly higher in dark-colored (green and brown) than in bright pitfall traps (Fig. 1). In contrast, the number of individuals (GLM, F = 15.5, df = 3, *P* < 0.0001) decreased by 42% from bright to dark traps (Fig. 1).

Species correlations to pitfall trap color were generally missing except for *Pardosa prativaga* and *Trochosa terricola* Thorell, 1856, which were associated with bright colors and *Palliduphantes pallidus* (O. Pickard-Cambridge, 1871), which was associated with dark colors (Appendix S1).

Species inventories were not complete, that is, the species accumulation curves indicated that the complete diversity of (rarer) species had not been sampled in any of the different pitfall trap colors. However species accumulation curves increased significantly more slowly in white (2.04 ± 0.49, mean ± SEM, repeated measures ANOVA: F = 124.9, df = 3, *P* < 0.001) and brown pitfall traps (2.20 ± 0.45) than in yellow (2.79 ± 0.43) and green ones (2.52 ± 0.44) (Fig. 2).

Pitfall trap color association differed among hunting types (GLM, F = 3.7, df = 4, *P* = 0.009) but pairwise comparisons indicated significant differences only between ground hunters and web builders (Fig. 3). The former had a higher association with bright colors while the latter were associated with dark traps. Regarding activity patterns (diurnal, nocturnal or diurnal-nocturnal), no significant differences were found in pitfall trap color association (GLM, F = 0.9, df = 2, *P* = 0.41) (Fig. 4).

Previous work revealed that catches among different pitfall trap colors significantly differ in terms of total numbers of individuals captured (Buchholz et al. 2010). However, this study shows that analyzing alpha diversity, community structure and trait composition yielded a more nuanced result which should be considered when using pitfall traps. One main insight from this study is that hunting guilds

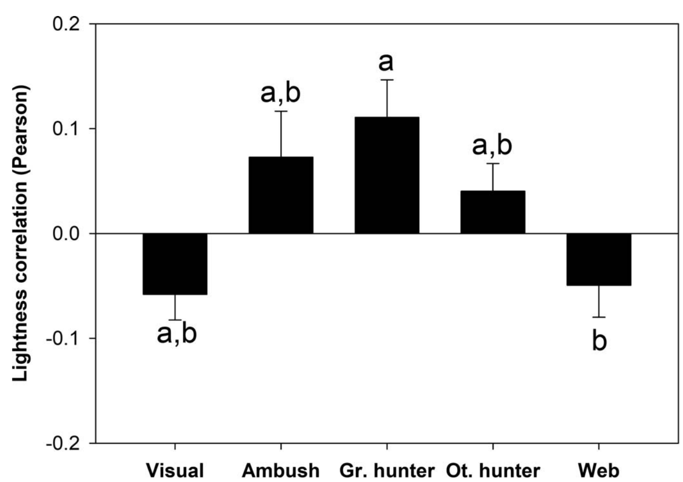

Figure 3.—Differences in lightness correlation among hunting types (GLM, F = 3.7, df = 4, P = 0.009). Pairwise comparisons (Holm-Sidak, P < 0.05) are presented with lower case letters, indicating significant differences only between ground hunters and web builders. Gr. Hunter = ground hunter, Ot. Hunter = other hunter. See appendix S1 online at http://dx.doi.org/10.1636/JoA-S-16-062.s1 for species assigned to each hunting style.

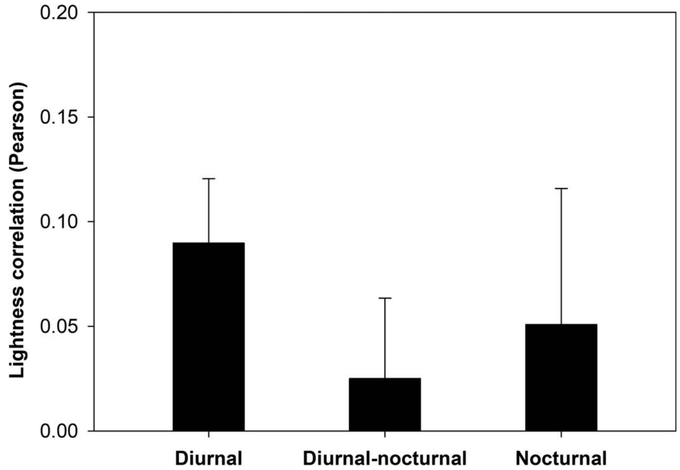

Figure 4.—Lightness correlation among activity categories (GLM, F = 0.9, dF = 2, P = 0.41). See appendix S1 online at http://dx.doi.org/10.1636/JoA-S-16-062.s1 for species assigned to each activity category.

significantly responded to different pitfall trap colors since ground hunters showed higher association to bright colors while web-builders were associated with dark ones. One explanation could be that ground hunters are able to distinguish colors to some extent and to notice contrasts (Foelix 2011; Zopf et al. 2013; Zurek et al. 2015). Attraction to bright trap colors could therefore be due to an improved visibility of prey on a bright background. Another reason could be a higher albedo of bright surfaces making white and yellow pitfall traps clearly visible points of orientation for ground-hunting spiders. In turn, web-builders might prefer green and brown due to lower albedo and color pattern similar to herbal layer where they commonly install their webs. Finally, differences in color are associated with different temperatures in the surroundings of pitfall traps. In this context, microclimate is known to significantly affect pitfall trap catches, their diversity, species composition and even trait distribution (Saska et al. 2013). Differences in trait signals can result in species-turnover among pitfall trap colors as for example two ground hunting species (*Pardosa prativaga* and *Trochosa terricola*) were related to bright colors, contrary to web-building *Palliduphantes pallidus* occurring in dark colors.

In terms of diversity, Shannon and Simpson diversity were significantly higher in dark compared to bright colors. Both indices are known to be sensitive to evenness of individuals per species, with higher values in communities having even individual sums (Magurran & McGill 2011). Hence, Shannon and Simpson diversity were lower in bright pitfall traps due to high individual sums of few wolf spiders (e.g., *Pardosa amentata*, *P. prativaga*, Appendix S1). Species inventories were not complete in any pitfall trap color but significantly lower growth of species accumulation curves in white and brown pitfall traps indicates a higher completeness in these traps.

Buchholz et al. (2010) concluded that the use of bright pitfall traps should increase spider capture efficiency and therefore enhance the level of precision of species inventories. Diversity analyzes showed that although overall individual sums were lower in green and brown pitfall traps, alpha diversity was nevertheless highest at these colors. However, given that diversity indices can be biased by individual sums, and that observed species richness was similar, these results should not be overrated. One practical implication is that species-rich spider communities can be also sampled in inconspicuous traps while avoiding unnecessary high individual catches at the same time. These findings are important, since avoiding unintentional by-catches should be intended in science due to animal welfare, ethics, and species protection (New 1999). Correspondingly, in their review, Brown & Matthews (2016) recommended transparent pitfall traps for all ground-active arthropods. This is in line with our findings, as transparent plastic cups or jars should be perceived as brown, environment, or soil color by spiders. In parallel, another practical implication is that white pitfall traps yield a higher biomass and – more important – attract more ground hunting spiders. Furthermore, white pitfall traps yielded a more complete species inventory, followed by brown ones. Considering all this, the main recommendation is to use a combination of white and brown (or transparent) pitfall traps for complete and diverse species inventories.

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