**Results**

*Isotopic niches of earthworm species*

The isotopic niches of six earthworm species were analyzed in the study site BDTR1: *A. caliginosa*, *A. trapezoides*, *A. chlorotica*, *D. caroliniana*, *L. friend*i, and *L. rubellus*. Overall, the three endogeic species, *A. caliginosa*, *A. trapezoides*, and *A. chlorotica*, had higher δ13C and δ15N values and occupied the upper-right part of the isotopic space; the anecic species, *L. friendi*, had lower isotope values and thus isotopic niche slightly below those of the endogeic species; the epigeic species, *L. rubellus*, had the lowest values and occupied the lower-left part of the isotopic space; the native species, *D. canoliniana*, showed distinct isotopic niche position from the other species, despite the small sample size (Fig. 2a). Pairwise comparisons of the total isotopic niches further suggested niche differentiation between species of different ecological groups: *A. caliginosa* and *L. friendi* (PERMANOVA *P* = 0.01; PERMDISP *P* = 0.81), *A. caliginosa* and *L. rubellus* (PERMANOVA *P* = 0.002; PERMDISP *P* = 0.16), *A. trapezoides and L. rubellus* (PERMANOVA *P* = 0.03; PERMDISP *P* = 0.17), and *L. friendi and L. rubellus* (PERMANOVA *P* = 0.06; PERMDISP *P* = 0.12) (Appendix S1: Table S2). Among the three endogeic species, *A. chlorotica* had a larger core niche area (SEAb = 14.82) compared with *A. caliginosa* (SEAb = 5.29) and *A. trapezoides* (SEAb = 6.53). *L. friend*i and *L. rubellus* had core niche areas in between those of the endogeic species (SEAb = 8.88 and 10.91, respectively) (Fig. 2a, Appendix S1: Table S1, Appendix S1: Fig. S1a). The percentage overlaps in the SEAB between species pairs were low to moderate on average (< 50.0%), except for three endogeic and anecic species: 71.4% of the SEABof *L. friendi* overlapped with that of *A. chlorotica*; 79.1% and 59.7% of the SEABof *A. trapezoides* overlapped with that of *A. chlorotica* and *L. friendi*, respectively (Fig. 4a, Appendix S1: Table S3).

The isotopic niches of six earthworm species were analyzed in the study site BDTR2: *A. caliginosa*, *A. trapezoides*, *A. chlorotica*, *D. caroliniana*, *L. friend*i, and *L. rubellus*. Similar to BDTR1, the three endogeic species, *A. caliginosa*, *A. trapezoides*, and *A. chlorotica*, had higher δ15N values and occupied the upper part of the isotopic space, whereas the anecic species, *L. friendi*, and the epigeic species, *L. rubellus*, had lower δ15N values and occupied the lower part of the isotopic space (Fig. 2b). The native species, *D. canoliniana*, also showed distinct isotopic niche position from the other species, indicating its unique feeding habits (Fig. 2b). Nonetheless, pairwise comparisons of the total isotopic niches did not reveal any significant niche differences between species (Appendix S1: Table S2). Among the three endogeic species, *A. caliginosa* had a larger core niche area (SEAb = 11.10) compared with *A. chlorotica* (SEAb = 5.82) and *A. trapezoides* (SEAb = 3.25). *L. friend*i and *L. rubellus* had core niche areas in between those of the endogeic species (SEAb = 7.35 and 7.26, respectively) (Fig. 2b, Appendix S1: Table S1, Appendix S1: Fig. S1b). The percentage overlaps in the SEAB between species pairs were low to moderate on average (< 50.0%), except for two endogeic species: 67.5% of the SEABof *A. trapezoides* overlapped with that of *A. caliginosa* (Fig. 4b, Appendix S1: Table S3).

The isotopic niches of two endogeic earthworm species were analyzed in the study site BARC: *A. caliginosa,* which had higher δ15N values and occupied the upper part of the isotopic space,and *A. trapezoides*, which had lower δ15N values and occupied the lower part of the isotopic space (Fig. 2c). Comparison of the total isotopic niches suggested niche differentiation between the two species (PERMANOVA *P* = 0.004; PERMDISP *P* = 0.38) (Appendix S1: Table S2). The core niche areas of *A. caliginosa* and *A. trapezoides* were similar in size (SEAB = 3.04 and 4.18, respectively) (Fig. 2c, Appendix S1: Table S1, Appendix S1: Fig. S1c) and showed low degrees of overlap (*ca.* 10.0%) (Fig. 4c, Appendix S1: Table S3).

*Variations in niche patterns of A. caliginosa and A. trapezoides*

Two epigiec species, *A. caliginosa* and *A. trapezoides,* exhibited variations in their isotopic niche patterns across the three study sites. In BDTR1 and BARC, the sizes of the core isotopic niche areas (SEAB) of *A. caliginosa* and *A. trapezoides* were comparatively similar, although the niche of *A. trapezoides* tended to be larger than that of *A. caliginosa* (*A. caliginosa* vs. *A. trapezoides*; BDTR1: SEAB = 5.29 vs. 6.53; BARC: SEAB = 3.04 vs. 4.18) (Appendix S1: Fig. S1a and c, Appendix S1: Table S1). The core niche areas of *A. caliginosa* and *A. trapezoides* were relatively separated, with the SEAB of *A. caliginosa* lying above that of *A. trapezoides* (i.e., higher δ15N values for *A. caliginosa* than for *A. trapezoides*) (Fig. 2a and c). The percentages of mutual SEAB overlaps between the two species were only low to moderate (*A. caliginosa* vs. *A. trapezoides*; BDTR1: 28.9% vs. 23.0%; BARC: 11.7% vs. 8.8%) (Fig. 4a and c, Appendix S1: Table S3).

In contrast to the patterns in BDTR1 and BARC, the size of the core niche area of *A. caliginosa* was more than three timeslarger than that of *A. trapezoides* in BDTR2 (*A. caliginosa* vs. *A. trapezoides*; SEAB = 11.10 vs. 3.25) (Appendix S1: Fig. S1b, Appendix S1: Table S1). Moreover, the core niches of the two species overlapped substantially (Fig. 2b), yet the percentages of mutual SEAB overlaps were highly asymmetrical (*A. caliginosa* vs. *A. trapezoides*; 21.2 vs.67.5%) due to the large differences in their niche sizes (Fig. 4b, Appendix S1: Table S3).

Finally, the total isotopic niches of the two species differed significantly in the study site BARC (PERMANOVA *P* = 0.004, PERMDISP *P* = 0.38) but not in BDTR1 (PERMANOVA *P* = 0.14, PERMDISP *P* = 0.91) and BDTR2 (PERMANOVA *P* = 0.56, PERMDISP *P* = 0.09) (Appendix S1: Table S2).

**Figures**

SEAb_biplot1.tiff

SEAb_biplot2.tiff

**Figure 2.** Stable isotope ratios (δ13C and δ15N) of earthworm species in the study sites BDTR1, BDTR2, and BARC. Ovals are Bayesian standard ellipse area (SEAB) representing the core isotopic niche of each species. Black points and error bars represent the means and standard errors of the stable isotope ratios of soil samples at 0-5 and 5-10 cm depth. The isotope ratios of earthworm samples were standardized using background soil isotope signatures (see Methods for more details). Note that there is no oval for *Diplocardia caroliniana* due to insufficient sample sizes for computation of SEAB.

**Figure 3.** Stable isotope ratios (δ13C and δ15N) of earthworm species in the study sites SERC1 and SERC2. Ovals are Bayesian standard ellipse area (SEAB) representing the core isotopic niche of each species. Black points and error bars represent the means and standard errors of the stable isotope ratios of soil samples at 0-5 and 5-10 cm depth. The isotope ratios of earthworm samples were standardized using background soil isotope signatures (see Methods for more details). Note that there is no oval for *Lumbricus terrestris* due to insufficient sample sizes for computation of SEAB.

Overlap_grey.tiff

**Figure 4.** The percentage of the SEAB of species A that overlaps with the percentage of the SEAB of species B in the study sites BDTR1, BDTR2, and BARC (see also Appendix S1: Table S3 for the numerical results).