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Mycorrhizal fungi-mediated uptake of tree-derived nitrogen by maize in smallholder farms

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Trees within farmers' fields can enhance systems' longer-term productivity, for example, via nutrient amelioration, which is indispensable to attain sustainable agroecosystems. While arbuscular mycorrhizal fungi (AMF) are known to improve plant access to soil nutrients, the potential of AMF to mediate nutrient uptake of tree-derived nitrogen (N) by crops from beyond the crops' rooting zones is unclear. We hypothesized that AMF quantitatively contribute to the crop uptake of tree-derived N. We set up root- and AMF-exclusion and control plots around faidherbia trees (*Faidherbia albida*) and used the ^{15}N natural abundance technique to determine the magnitude of AMF-mediated uptake of tree-derived N by maize from beyond its rooting zone in smallholder fields. We further tested whether AMF-mediated N uptake decreases with distance from tree. We show that within one cropping season, maize obtained approximately 35 kg ha^{-1} biologically fixed N from faidherbia. One-third of tree-derived N in maize leaves was attributed to AMF-mediated N uptake from beyond the maize rooting zone and two-thirds to N from tree leaf litter, regardless of distance from tree. As hypothesized, maize grown close (1 m) to faidherbia obtained significantly more tree-derived N than that at farther distances (4 and 5 m). Thus, the faidherbia–AMF association can enhance agroecosystem functioning.

Sound management practices are essential to attain food security, which is still at continuous risk in sub-Saharan Africa[1]. Agroforestry can provide a framework for sustainable farming: trees distributed throughout farmers' fields can enhance soil fertility via above- and belowground organic matter inputs[2–6]. Nutrients of these inputs become available to surrounding crops through various mechanisms such as mineralization[2–4] and mechanisms mediated by arbuscular mycorrhizal fungi (AMF)[6–8].

AMF, ubiquitous and abundant mutualistic root symbionts that associate with 72% of terrestrial plants[9], form extensive mycelia that increase the exploited soil volume[10] and may interconnect plants' rooting zones. As such, AMF can enable the uptake and transfer of nutrients from root exudates and mediate indirect nutrient transfer between plants[6,10]. Moreover, AMF may link different plants via mycelia[11] and enable direct interplant nutrient transfer[8,12,13]. In return, AMF require plant carbohydrates[10]. Nitrogen (N)-fixing trees can provide microdose fertilization, potentially increasing crop yield[14–21], but the role of AMF in N acquisition by crops remains poorly explored. Indigenous AMF could increase the uptake of tree-derived biologically fixed N_2 by crops from outside the crops' rooting zones, particularly in subsistence farming where soil nutrient inputs are low and farmers need to leverage ecosystem processes to improve food security.

We estimated the total amount of biologically fixed N_2 derived from faidherbia trees (*Faidherbia albida*; Fabaceae; hereafter referred to as 'tree-derived N') in surrounding maize plants and quantified the potential significance of indigenous AMF in making tree-derived N accessible to maize plants within a season and subsequent effects on maize yield in Malawian farmers' fields. Faidherbia trees are known for their potential to increase soil fertility and crop yield[14–21] and for hosting AMF in topsoil and deep soil layers[22]. We used the ^{15}N natural abundance technique to distinguish between tree-derived N and N derived from soil, in combination with root- and AMF-exclusion and control plots (Fig. 1). Three types of plots were installed around eight faidherbia trees that were distributed in farmers' fields under maize cultivation to distinguish between three types of interactions between faidherbia and maize: (1) fully restricted belowground interactions, limiting the access of maize to tree-derived N from leaf litter ('Litter only' plot); (2) belowground interactions restricted to those enabled via mycelia of indigenous AMF ('Litter&AMF' plot); and (3) unrestricted interaction between tree and maize ('Litter&AMF&Roots' plot). Note that we added neither tree leaf litter nor AMF inoculum and that the plot designations refer to the sources of or pathways potentially mediating tree-derived N uptake by maize: (1) leaf litter only, (2) leaf litter and indigenous AMF (hereafter referred to as "AMF") potentially exploiting the tree rooting zone, or (3) leaf litter, AMF potentially exploiting the tree rooting zone and direct contact with tree roots. This set-up allowed disentangling the effects of litter, AMF and tree roots on the amount of tree-derived N in maize across distance. We focused our study on maize-based agroforestry systems of Malawian smallholders because they exemplify a common agroecosystem that must be managed more sustainably to attain food security[1].

Results

Faidherbia trees were actively fixing atmospheric N_2. Foliar δ^{15}N of faidherbia ranged from −0.24 to 1.59‰ with an average of 0.92 ± 0.24‰ (mean ± standard error (SE), $n = 8$). The fact that the δ^{15}N values of faidherbia ranged around 0 indicates that the trees acquired almost all of their N from the atmosphere via symbiotic N_2 fixation. Foliar N concentration of faidherbia ranged from 3.62 to

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Fig. 1 | Low-input agroforestry system with maize and *F. albida* in central Malawi and visualization of the experimental manipulation used to study the effect of AMF-mediated uptake of tree-derived biologically fixed N_2 by maize. **a**, *F. albida* trees distributed throughout smallholder farmers' maize fields. **b**, Experimental plots to manipulate the type of belowground interaction between tree and maize plants. Root- and AMF-exclusion and control plots together with the ^{15}N natural abundance technique were used to estimate the contribution of AMF-mediated uptake of tree-derived biologically fixed N_2 by maize from beyond its rooting zone.

5.38% with an average of 4.44 ± 0.21% (mean ± SE, $n = 8$), suggesting that the trees were well supplied with N.

Plot type and distance effects on maize and soil $\delta^{15}N$ and N. The foliar $\delta^{15}N$ of maize differed among the plot types (type of interaction between tree and maize) ($F_{2,94} = 10.71$ (F statistic including, in subscript, the degrees of freedom in the numerator and denominator), $P < 0.001$; Table 1). The leaves of maize plants grown in the plot type Litter only were enriched in ^{15}N by 1.3-fold and 1.2-fold; that is, they acquired significantly less tree-derived N and more soil N relative to maize grown in the Litter&AMF plots and Litter&AMF&Roots plots, respectively ($P < 0.001$). However, there was no significant difference in foliar $\delta^{15}N$ for maize grown in the Litter&AMF versus the Litter&AMF&Roots plots. Distance from faidherbia also affected maize foliar $\delta^{15}N$ ($F_{4,94} = 5.93$, $P < 0.001$), with maize grown at 4 and 5 m from the tree being more enriched in ^{15}N by 1.4-fold and 1.5-fold, respectively, than maize grown at 1 m from the tree ($P = 0.001$ and $P < 0.001$, respectively; Table 1). There was no interaction between the two main factors, plot type and distance from faidherbia, on foliar $\delta^{15}N$. Similar results were obtained for the

Table 1 | $\delta^{15}N$ (‰) signatures and N concentrations of maize leaf and paired soil samples at increasing distances of the maize plants from *F. albida*

		Distance from tree (m)				
Plot type		1	2	3	4	5
Litter only	$\delta^{15}N_{maize}$	$3.5 \pm 0.4^{A,b}$	$4.1 \pm 0.6^{A,a,b}$	$4.0 \pm 0.4^{A,a,b}$	$4.5 \pm 0.5^{A,a}$	$4.7 \pm 0.5^{A,a}$
	$\delta^{15}N_{soil}$	$5.6 \pm 0.2^{A,a}$	$5.8 \pm 0.2^{A,a}$	$5.8 \pm 0.2^{A,a}$	$5.8 \pm 0.1^{A,a}$	$5.7 \pm 0.1^{A,a}$
	% N_{maize}	$1.7 \pm 0.1^{A,a}$	$1.7 \pm 0.2^{A,a,b}$	$1.6 \pm 0.1^{A,a,b,c}$	$1.6 \pm 0.1^{A,b,c}$	$1.6 \pm 0.1^{A,c}$
Litter&AMF	$\delta^{15}N_{maize}$	$2.5 \pm 0.4^{B,b}$	$3.0 \pm 0.2^{B,a,b}$	$3.4 \pm 0.4^{B,a,b}$	$3.6 \pm 0.4^{B,a}$	$3.7 \pm 0.5^{B,a}$
	$\delta^{15}N_{soil}$	$5.7 \pm 0.2^{A,a}$	$5.7 \pm 0.2^{A,a}$	$5.8 \pm 0.1^{A,a}$	$5.8 \pm 0.2^{A,a}$	$5.8 \pm 0.1^{A,a}$
	% N_{maize}	$2.1 \pm 0.1^{A,a}$	$1.8 \pm 0.1^{A,a,b}$	$1.8 \pm 0.1^{A,a,b,c}$	$1.5 \pm 0.1^{A,b,c}$	$1.4 \pm 0.1^{A,c}$
Litter&AMF&Roots	$\delta^{15}N_{maize}$	$2.4 \pm 0.3^{B,b}$	$3.0 \pm 0.3^{B,a,b}$	$2.8 \pm 0.3^{B,a,b}$	$3.9 \pm 0.4^{B,a}$	$3.7 \pm 0.4^{B,a}$
	$\delta^{15}N_{soil}$	$5.5 \pm 0.2^{A,a}$	$5.6 \pm 0.2^{A,a}$	$5.6 \pm 0.2^{A,a}$	$5.8 \pm 0.2^{A,a}$	$5.9 \pm 0.2^{A,a}$
	% N_{maize}	$1.8 \pm 0.1^{A,a}$	$1.9 \pm 0.1^{A,a,b}$	$1.8 \pm 0.1^{A,a,b,c}$	$1.8 \pm 0.1^{A,b,c}$	$1.5 \pm 0.1^{A,c}$

Plot type refers to the type of interaction between tree and maize, as follows. Litter only: pond liner to eliminate all belowground interactions between tree and maize, limiting tree-derived N inputs to leaf litter only; Litter&AMF: 40 µm mesh to restrict interactions between tree and maize roots to those via extraradical mycelia of AMF only and eliminate the possibility that tree and maize roots could intermingle; Litter&AMF&Roots: no lining for unrestricted interactions between tree and maize and to control for the potential effect of soil excavation during the set-up of the experimental plots. Values represent mean ± SE ($n = 8$). Different letters indicate significant differences ($P < 0.05$) across plot type (capital letters) and across distance from tree (lowercase letters) for each of the three response variables ($\delta^{15}N_{maize}$, $\delta^{15}N_{soil}$ and % N_{maize}). There was no significant interaction between plot type and distance from tree for any of the response variables. The average $\delta^{15}N_{soil}$ across plot type and distance from tree was 5.74 ± 0.04‰. The average % N_{maize} at 1, 2, 3, 4 and 5 m was 1.86 ± 0.07, 1.82 ± 0.06, 1.72 ± 0.05, 1.63 ± 0.07 and 1.51 ± 0.07, respectively. The foliar $\delta^{15}N$ signature of faidherbia ranged from −0.24 to 1.59‰ (average: 0.92 ± 0.24‰) and the foliar N concentration of faidherbia ranged from 3.62 to 5.38% (average: 4.44 ± 0.21%; mean ± SE, $n = 8$).

effect of plot type and distance from faidherbia on the proportion of tree-derived N in maize (as determined using equation (1); data not shown). Foliar N concentration in maize was significantly affected by distance from faidherbia ($F_{4,94} = 5.80$, $P < 0.001$; Table 1) with 1.2-fold greater foliar N concentration in maize grown at 1 and 2 m compared with 5 m ($P < 0.001$ and $P = 0.004$, respectively) across all three plot types. Foliar N concentration in maize at 1 m was 1.1-fold greater than at 4 m ($P = 0.047$). Plot type did not significantly affect foliar N concentration ($F_{2,94} = 1.50$, $P = 0.23$). On average, foliar N concentration was 1.86 ± 0.07, 1.82 ± 0.06, 1.72 ± 0.05, 1.63 ± 0.07 and 1.54 ± 0.07 at 1, 2, 3, 4 and 5 m, respectively (mean ± SE, $n = 24$). Nitrogen concentration and $\delta^{15}N$ in paired soil samples were not affected by plot type ($F_{2,94} = 1.53$, $P = 0.22$ and $F_{2,94} = 1.34$, $P = 0.27$, respectively) or distance from faidherbia ($F_{4,94} = 0.91$, $P = 0.46$ and $F_{4,94} = 1.47$, $P = 0.22$, respectively) and were on average 0.16 ± 0.002% and 5.74 ± 0.04‰, respectively (Table 1).

Litter-, AMF- and root-mediated N uptake across distance. The proportion of tree-derived N in maize as a result of litter-, AMF- and root-mediated processes did not differ with distance from faidherbia trees ($F_{4,27} = 1.24$, $P = 0.32$; $F_{4,27} = 0.30$, $P = 0.87$; and $F_{4,25} = 0.90$, $P = 0.48$, respectively; as determined using equations (2) and (3); Fig. 2). The effect of roots on tree-derived N in maize was negligible (Fig. 2).

Yield and total tree-derived N in maize. Shoot biomass, grain yield, N content and total tree-derived N in maize leaves on a per plot basis did not significantly differ between plot types ($F_{2,14} = 0.35$, $P = 0.71$; $F_{2,14} = 0.86$, $P = 0.44$; $F_{2,14} = 0.32$, $P = 0.73$; and $F_{2,14} = 2.09$, $P = 0.16$, respectively; Table 2). The total amount of tree-derived N in maize plants grown within 5 m from faidherbia across all three plot types (estimated using average values for maize leaves across distances and plots and using per tree values obtained from stalk, grain and cob subsamples; Methods) summed up to 35 ± 7 kg ha^{-1} N of the total amount of 120 ± 7 kg ha^{-1} N, hence making up about 30% of total N in maize.

Discussion

Incorporating faidherbia trees in agroecosystems can benefit crop yields through microdose fertilization. N_2-fixing trees provide high-quality above- and belowground organic matter inputs to the soil, for example, in the form of tree leaf litter input, root exudates and root turnover[2–4]. Faidherbia leaf litter input alone can provide 50 to 80 kg ha^{-1} N to the soil under faidherbia trees within a given season[18,19]. However, how much of this tree leaf litter-derived N is mineralized and subsequently incorporated into crop biomass is yet unknown. The distinct isotopic N signature, ^{15}N:^{14}N ratio, of N_2-fixing faidherbia allows distinguishing between tree-derived biologically fixed N_2 and residual soil N[23,24]. Because maize does not biologically fix N_2, its isotopic N signature is determined largely by the residual soil N; thus, the ^{15}N natural abundance technique allows tracing the isotopically distinct tree-derived, biologically fixed N_2 into maize[23,24]. Specifically, determining the $\delta^{15}N$ of maize leaf, paired soil samples and faidherbia reference samples and using an isotope mixing model allowed calculating the proportion of tree-derived N in maize. Note that the ^{15}N natural abundance technique does not detect any non-biologically fixed N_2 derived from the trees, resulting in an underestimation of the tree-derived N in maize. We found that in total, over the course of one season, tree-derived N accounts for 35 kg ha^{-1} N in maize, which corresponds to about 30% of the total N in maize. Therefore, our results confirm the importance of faidherbia trees in improving the N budget of crops in farmers' fields. The broad-scale recommended rate of N fertilization in Malawi is 96 kg ha^{-1} N, but on average only 18 kg ha^{-1} N is applied by farmers[25,26], and many farmers, as those in our study region, lack access to fertilizer and thus do not apply any fertilizer to their field (according to conversations with the farmers we worked with). We demonstrate that faidherbia provides more than one-third of the recommended dose of fertilizer and almost twice the amount that is on average applied by subsistence farmers.

Microdose fertilization has been shown to result in substantial yield increases. For example, microdose fertilization of 24 kg ha^{-1} N resulted in a 64% increase in maize grain yield relative to an unfertilized control[27]. We found maize yields within 5 m of faidherbia were approximately 50% greater compared with yields of maize plants away from faidherbia (determined in a previous study[32]). Specifically, maize grain yield was 3.7 ± 0.4 t ha^{-1} under faidherbia compared with 2.5 ± 0.6 t ha^{-1} at about 35 m away from faidherbia. We note that the yield determined away from faidherbia was based on green cob dry weight[32], while yield under faidherbia was determined from mature cobs, after the plants had fully matured, which might have resulted in a slight underestimation of yield

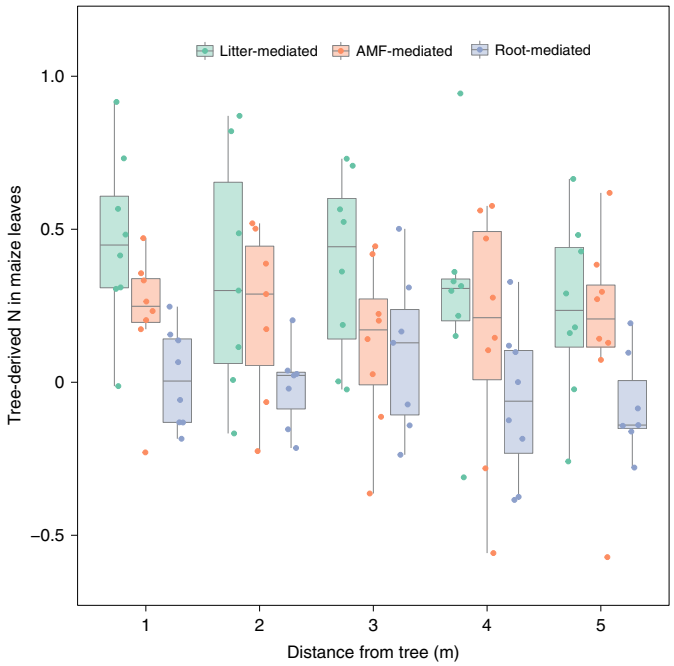

Fig. 2 | Proportion of tree-derived N in leaves of maize surrounding *F. albida*. The proportion of tree-derived N in maize leaves from tree leaf litter, AMF-mediated uptake by maize or direct root-to-root contact between tree and maize (litter-, AMF- and root-mediated, respectively; see Methods for further details, particularly equations (2) and (3)) with distance from *F. albida* ($n = 8$). Boxplot elements are defined as follows: the centre line represents the median, box limits represent the upper and lower quartiles, whiskers represent 1.5 times interquartile range and the points represent each individual data point.

away from faidherbia and therefore a slight overestimation of the approximate 50% increase in yield under relative to away from faidherbia. Nevertheless, our estimates are in line with previous findings[27]. Furthermore, our results show that foliar N concentration was greater within the immediate vicinity (1–2 m) of faidherbia compared with farther distances (4–5 m). The same holds true for tree-derived N in maize leaves as indicated by the increase in $\delta^{15}N$ in maize leaves with distance from tree. We conclude that faidherbia trees are effective in providing N microdose fertilization to crops in subsistence farmers' fields and, therefore, may contribute to increased yields.

Mycorrhizal fungi increase uptake of tree-derived N by maize. Greater crop yields and soil nutrient contents previously observed around N_2-fixing trees within agricultural fields have been ascribed mostly to high-quality organic matter inputs to the soil[14–21]. The contribution of AMF in making these inputs available to crops has gained much less attention. There has been some evidence that AMF facilitate N transfer from trees to surrounding plants[8], but verification of this mechanism under field conditions on smallholder farms has been lacking. Faidherbia has been shown to associate with AMF, in both topsoil and great soil depth[22]. Our experimental design and methods did not allow to provide evidence for the existence of direct linkages between tree and maize via a mycorrhizal mycelia. Therefore, we cannot differentiate between direct root-to-root- and indirect soil-to-root-mediated N transfer enabled by AMF. However, combining the ^{15}N natural abundance technique with root- and AMF-exclusion plots allowed us to quantify the effect of AMF-mediated uptake of tree-derived N by maize from beyond its rooting zone, whether via direct or indirect transfer, in farmers' fields. While we cannot exclude the possibility that other filamentous fungi may have contributed to the uptake of tree-derived N by maize from beyond its rooting zone[28,29], we assume that most of the uptake was AMF-mediated. We estimated that the AMF-mediated uptake of tree-derived N by maize plants accounted for 28% of the total tree-derived N in maize leaves within 5 m around faidherbia trees (Table 2). Tree litter was responsible for most, about two-thirds, of the tree-derived N in maize, and the presence of tree roots within the rooting zone of maize (if at all present) had a negligible effect on the uptake of tree-derived N by maize (Fig. 2 and Tables 1 and 2).

Table 2 | Maize shoot biomass, grain yield, leaf N content and total tree-derived biologically fixed N_2 in maize leaves of all maize plants grown within the different experimental plots within a 5 m radius around *F. albida*

Plot type	Biomass ($t\,ha^{-1}$)	Yield ($t\,ha^{-1}$)	N content in leaves ($kg\,ha^{-1}$)	Tree-derived N in leaves ($kg\,ha^{-1}$)
Litter only	4.0 ± 0.3	4.1 ± 0.3	18.2 ± 1.3	6.3 ± 1.8
Litter&AMF	3.7 ± 0.6	4.3 ± 0.4	17.3 ± 2.4	8.8 ± 1.5
Litter&AMF&Roots	3.7 ± 0.5	3.7 ± 0.4	18.5 ± 2.5	8.9 ± 1.8

None of the variables were significantly affected by plot type (that is, the type of interaction between tree and maize). Values represent mean ± SE ($n = 8$).

The experimental plots were located under the tree crown (average crown radius of 5 ± 0.4 m), and therefore, it was expected that the proportion of tree-derived N obtained by maize from tree litter input was the same within the 5 m radius around faidherbia (Fig. 2). Similarly, the $\delta^{15}N$ and total N concentration of the surface soil must have been affected mostly by tree leaf litter input which, given homogeneous tree leaf litter input across the plots, may explain why we observed no increase of soil $\delta^{15}N$ and total N concentration with distance from tree. The contribution of AMF-mediated N transfer from tree to crops (direct or indirect) versus uptake via root-to-root contact and direct uptake of N from tree root exudates by crop roots to the crops' N budget probably depends on the architecture of the tree root system and the distance from tree. Specifically, we expected a decrease in the contribution of AMF- and root-mediated uptake of tree-derived N by maize with distance from tree due to an increasing distance between the maize and tree rooting zones. Indeed, given the maize foliar $\delta^{15}N$ but not the soil $\delta^{15}N$ increased with distance from tree, our data provide some evidence for this hypothesis. However, the proportion of tree-derived N obtained by maize via litter-, AMF- and root-mediated processes was not significantly affected by distance from tree (Fig. 2). We did not examine the tree root system, but observed no fine tree roots within a radius of 5 m from faidherbia (at a depth of 0 to 50 cm), which is in line with previous findings about faidherbia's deep taproot development[30]. Even if maize roots usually grow deeper than 50 cm in the absence of our experimental plots, the lack of fine tree roots within the top 50 cm and across the 5 m from the base of the tree suggests that root-to-root contact between faidherbia and maize is typically minimal and explains why we found no additional increase in tree-derived N obtained by maize plants grown in the Litter&AMF&Roots plots relative to those grown in the Litter&AMF plots (Table 2). Therefore, given the apparent separation of faidherbia and maize roots, our results highlight the potential importance of AMF in connecting the soil volume between the rooting zone of faidherbia and maize for maize to gain access to a larger pool of tree-derived N.

Despite the contribution of AMF to the proportion of tree-derived N in maize, maize shoot biomass and grain yield were not significantly increased (Table 2). This is probably because total foliar N content was not affected by plot type (type of interaction between tree and maize; Table 2). Total tree-derived N in maize leaves was also not significantly different between plot types, but

the data follow the same trend as the proportion of tree-derived N (Table 2). While AMF-mediated uptake of tree-derived N by maize from beyond its rooting zone may not increase the N content in maize, an AMF-mediated uptake of tree-derived N from beyond the rooting zone of maize may improve internal N cycling within the agroecosystem.

In conclusion, this study provides insight into the underlying ecological process through which N input from trees may be made accessible to crops. It appears that AMF connect the space between the rooting zones of trees and crops via mycelia and, as such, increase the amount of tree-derived N accessible to crops. Especially in low-input cropping systems such as those in Malawi, N microdose fertilization by faidherbia trees and AMF-mediated uptake of tree-derived N by crops could enhance sustainability of agroecosystems in the longer term.

Methods

Study site. The study site was located in central Malawi, in the lowlands of Dedza district. The fields are distributed within an area of approximately 5 km^2 around the village of Ndindi in the Golomoti Extension Planning Area (14.3° S, 34.6° E). Soil types are fluvisols[31] with mostly sandy clay loam textures. At 1 and 4 m from faidherbia, respectively, mean soil total C was 28.7 ± 0.44 g kg^{-1} and 26.8 ± 0.55 g kg^{-1}, mean soil total N was 2.2 ± 0.05 g kg^{-1} and 2.0 ± 0.04 g kg^{-1}, mean soil total P was 2.1 ± 0.11 g kg^{-1} and 2.1 ± 0.06 g kg^{-1}, mean soil resin P was 33.2 ± 6.01 mg kg^{-1} and 46.5 ± 6.73 mg kg^{-1} and mean weight diameter (a measure of soil aggregate stability) was 3.15 ± 0.07 mm and 3.17 ± 0.08 mm, as determined in a previous study[32] (mean ± SE; n = 10). At 35 m from faidherbia, mean soil total C was 17.8 ± 2.14 g kg^{-1}, mean soil total N was 1.5 ± 0.28 g kg^{-1}, mean soil total P was 1.9 ± 0.09 g kg^{-1} and mean soil resin P was 24.8 ± 6.66 mg kg^{-1}, and mean weight diameter of the soil aggregates was 2.01 ± 0.22 mm (mean ± SE; n = 10). The climate is subtropical, humid with a unimodal precipitation pattern. Most rain falls from November to March, and the average annual precipitation is 884 mm (ref. [33]). Farmers prepare their fields by hand hoeing (~15 cm deep). Maize is planted at the onset of the rainy season around December and typically harvested in April/May. Farmers' fields within the study site are not amended with chemical fertilizer, and weeding is done with a hand hoe.

Tree selection. We focused our study on faidherbia (*Faidherbia albida*) because this leguminous tree has been highly promoted as an agroforestry species due to its 'reverse phenology'. The trees' foliage is shed with the onset of the rainy season[18], resulting in minimized light competition between trees and crops at crop establishment and in high-quality litter inputs at a time when soil moisture conditions are favourable for rapid mineralization[15,17]. A total of eight single-standing faidherbia trees distributed throughout farmers' maize fields were selected for this study. All were well-established trees of a similar size (average diameter at breast height 53 ± 3.5 cm, height 16 ± 1.0 m, crown radius 5 ± 0.4 m), single-standing (at least 40 m away from the base of any neighbouring tree) and with a recent cropping history of maize. Within our study system, farmers do not commonly prune faidherbia trees, and none of the selected study trees was pruned. Hence, the N derived from the trees originated from leaf litter and/or roots.

Experimental set-up (year 1). At the beginning of the growing season of 2016/2017, we excavated three rectangular plots (1 m × 5 m, 0.5 m deep) around each tree within farmers' maize fields (Fig. 1). Plots started at a distance of 0.7 m from the base of the tree and were oriented towards 0° (North), 120° (Southeast) and 240° (Southwest). Each plot was fitted with a different bottom and sidewall lining, resulting in three types of experimental plots (three types of interactions between tree and maize). The types of interactions were randomly assigned to the experimental plots to account for possible differences in microclimate. The three types of lining were as follows:

- Pond liner (AlfaFol PVC pond liner 0.5 mm thick; Oase Living Water) to eliminate all belowground interactions between tree and maize, limiting tree-derived N inputs to leaf litter only (Litter only plot)
- A 40 µm mesh (SEFAR Petex 07-40/25, Sefar AG) to restrict interactions between tree and maize to those via the extraradical mycelia of AMF only and eliminate the possibility that tree and maize roots could intermingle (Litter&AMF plot)
- No lining for unrestricted interactions between tree and maize and to control for the potential effect of soil excavation during the set-up of the experimental plots (Litter&AMF&Roots plot)

Soils host by default AMF[34], which when no mineral fertilizer is applied are usually particularly abundant and beneficial to the mineral nutrition of plants[35]. Hence, the '&AMF' in the plot type designation does not refer to the addition of AMF but indicates the presence of extraradical mycelia of indigenous AMF connecting the soil volume exploited by the roots of the maize plants and the soil volume used by the tree. Note that our experimental design does not exclude the possibility that filamentous fungi other than AMF may have also contributed to the uptake of tree-derived N from beyond its rooting zone[28,29]. Further note that we did not add any tree leaf litter to the plots but that the plot names refer to the source of tree-derived N that maize could access. Leaf litter that had accumulated on the soil surface was removed before excavating the plots to prevent leaf litter N input to deeper soil. The excavated soil was piled up next to the plot, and after the lining had been put in place, the soil was placed back into the plot, beginning with the soil from the top of the pile and continuing to that on the bottom to keep the original soil depth position. After completion of the experimental plots, maize was sown along two rows within each plot and across farmers' fields (at least within a 10-m radius circle around each study tree). Farmers weeded, harvested and eventually prepared the fields (including our experimental plots) for the next growing season following their common practices (using a hand hoe, planting around December and harvesting around April/May, as previously described). No measurements were taken in the year of the experimental plot set-up to let the soil and AMF mycelia recover from the experimental plot installation. Leaving the system to recover for one year minimized the risk of potentially altered mineralization of soil organic matter resulting from the experimental plot installation influencing our results.

Sample collection (year 2). One year after the experimental plot set-up, at the beginning of the growing season of 2017/2018, maize was sown into each experimental plot and across farmers' fields (at least within a 10 m radius around each study tree). In each plot, maize was sown along two rows (0.6 m apart) at every metre, from 1 to 5 m from the base of the trees. The experimental plots were continuously hand weeded by the farmers. Seventeen weeks after sowing, at the time of harvest, maize leaf samples and paired soil samples (0–15 cm) were collected at every metre from the tree trunk in each plot. The maize leaf samples and soil samples from the same distance of the same plot were pooled into a composite sample per distance from the tree, resulting in five leaf and five soil samples per plot per tree and a total of 120 leaf and soil samples each, for physiochemical analyses. Faidherbia leaf samples were collected to obtain reference values of the $\delta^{15}N$ signature and N concentration to estimate the amount of tree-derived N in maize. Dry, homogenized plant and soil samples were analysed for $\delta^{15}N$ and total N with an elemental analyser (Vario PyroCube, Elementar) connected to an isotope ratio mass spectrometer (Isoprime 100, Elementar) in continuous flow mode.

The fraction (proportion) of tree-derived N ($frac_{N(tree)}$) in the leaves of the maize plants was determined for all five distances (1–5 m) from the trees, according to the following equation:

$$frac_{N(tree)} = \frac{\delta^{15}N_{maize} - \delta^{15}N_{soil}}{\delta^{15}N_{tree} - \delta^{15}N_{soil}} \quad (1)$$

where $\delta^{15}N_{maize}$ and $\delta^{15}N_{soil}$ are the $\delta^{15}N$ (‰) of maize leaf and soil samples at each distance, respectively, and $\delta^{15}N_{tree}$ are the $\delta^{15}N$ (‰) of the corresponding faidherbia tree leaf samples.

We determined AMF-mediated and root-mediated uptake of tree-derived N by maize as follows:

$$frac_{AMF\_med} = frac_{Litter\&AMF} - frac_{Litter} \quad (2)$$

$$frac_{Root\_med} = frac_{Litter\&AMF\&Roots} - frac_{Litter\&AMF} \quad (3)$$

where $frac_{AMF\_med}$ and $frac_{Root\_med}$ are the proportion of tree-derived N in maize leaves obtained via AMF- and root-mediated processes, respectively, $frac_{Litter}$ is the proportion of tree-derived N in maize leaves in the Litter only plots, $frac_{Litter\&AMF}$ is the proportion of tree-derived N in maize leaves in the Litter&AMF plots and $frac_{Litter\&AMF\&Roots}$ is the proportion of tree-derived N in maize leaves in the Litter&AMF&Roots plots. We inferred the effect of tree roots in the absence of AMF by the 'difference calculation method' because it is impossible to establish an 'AMF-free control treatment' in farmers' fields. While it would have been ideal to have an AMF-free soil, there is a plethora of literature discussing the challenge of establishing an AMF-free control soil[36–39]. AMF are ubiquitous in field soil, and the hitherto best AMF-free control soil is using reduced mycorrhizal colonization (rmc) mutants[37]. However, even the rmc varieties are being colonized by AMF[37,40] and hence, for the purpose of our study, would not be suitable for a +legume root/AMF-free treatment. Furthermore, there is no rmc mutant for faidherbia, and even if there were, establishing a new tree in the field would result in a generation-time experiment. The use of fungicides, for example, benomyl, to create an AMF-free soil is highly debated because of side effects on the saprotrophic ascomycetes[36]. Not only would the use of fungicides have caused such major experimental artifacts[41], but moreover, the use of fungicides in farmers' fields would have been a very invasive approach. In addition, given that we would have had to treat the soil down to a very deep depth[42] to treat the soil around the rooting zone of maize, this method was unfeasible.

Total maize biomass and mature cob fresh weight per experimental plot were determined in the field, and composited, homogenized subsamples per tree were oven dried to determine dry weights. Dry weight of each fraction (leaves, stalks, grain and cob) per tree were determined from the total shoot and the cob weights, using known proportions for maize[43]. The oven-dried, composited biomass and cob samples

obtained per tree were subsequently separated into stalk, grain and cob subsamples, which were analysed for total N concentration and $\delta^{15}N$ signature, as described in the preceding. For maize leaves, we used the average total N concentration and $\delta^{15}N$ signature across distance and plots to obtain a per tree estimate. We used the total N concentration and $\delta^{15}N$ signature of each fraction (stalks, grain, cob and leaves) to determine the N content of each fraction per tree and to estimate the total N content in maize and the total amount of tree-derived N in maize grown within 5 m from faidherbia (see the following). The total amount of tree-derived N ($total_{N(tree)}$; kg ha^{-1} N) in maize leaves was calculated on a per plot basis as follows:

$$total_{N(tree)} = (N_{maize})\left(frac_{N(tree)}\right) \quad (4)$$

where N_{maize} is the total amount of N in maize leaves per hectare (kg ha^{-1} N), and $frac_{N(tree)}$ is the proportion of tree-derived N in maize leaves as calculated by equation (1). To obtain total tree-derived N in maize on a per tree basis, we estimated total tree-derived N per fraction and subsequently determined the sum of total tree-derived N per fraction for all fractions (leaves, stalks, grain and cobs) combined. Hence, we adjusted equation (4) to have N_{maize} and $frac_{N(tree)}$ represent the total N content and the proportion of tree-derived N in maize stalk, grain or cob to determine the total amount of tree-derived N ($total_{N(tree)}$) in maize stalk, grain or cob, respectively, for each tree. For maize leaves, we used the average obtained across plots. The proportions of tree-derived N ($frac_{N(tree)}$) in maize stalk, grain and cob on a per tree basis were calculated using equation (1) with $\delta^{15}N_{maize}$ as the $\delta^{15}N$ (‰) of maize stalks, grain or cob per tree, respectively, $\delta^{15}N_{soil}$ as the average $\delta^{15}N$ (‰) of soil determined per tree and $\delta^{15}N_{tree}$ as the $\delta^{15}N$ (‰) of the corresponding faidherbia tree leaf samples. For maize leaves, we used the average obtained across distance and plots. Similarly, we calculated total N in maize (kg ha^{-1}) by determining N content per plant fraction on the basis of N concentration obtained per fraction per tree and dry weight per fraction per tree (for leaves, an average across distance and plots was used) and taking the sum of the N content of all fractions.

Maize root colonization by AMF. Given the relatively low number of plants per plot and the fact that making the plots larger to allow for more plants per plot was financially unfeasible, we decided against assessing root colonization by AMF to increase the chances of obtaining reliable maize yield and $\delta^{15}N$ data at the time of harvest. We refer to a previous study[32] in which we assessed root colonization by AMF within 15 m around the same study trees and found no difference in root colonization of maize plants grown close to versus at farther distances (at 1 versus 15 m) from the trees. Percentage colonization by hyphae and arbuscules was on average 33 ± 1.2% and 31 ± 0.8% within the 15 m radius around faidherbia trees, respectively, and the average number of vesicles was 92 ± 25 (mean ± SE, $n = 10$). Furthermore, in a greenhouse experiment, we determined a high AMF infection potential of the Malawian field soil leading to highly colonized maize roots regardless of whether the soil could have been colonized by AMF originating from the trees (J.D., unpublished data). Hence, we feel confident in assuming that maize root colonization was approximately the same across all plots and did not affect the outcome of our results.

Maize yield under versus away from faidherbia trees. To assess the effect of trees on maize grain yield, we compared the average grain yield within 5 m from faidherbia with grain yield estimates derived from green cob dry weights of maize plants that grew about 35 m from faidherbia trees in the 2015/2016 cropping season.

Statistical analyses. All statistical analyses were performed in the software environment R (R 4.0.3 GUI 1.73)[44]. We used linear mixed-effects models to assess the effect of plot type and/or distance of the maize plants from the tree on the various response variables (see the following) while accounting for variation caused by inherent differences across individual trees (fields) by including 'tree' as a random-effect variable in the models in addition to the study factors that were tested as fixed effects[45]. Specifically, we used linear mixed-effects models with individual tree included in the models as a random-effect variable and plot type and distance as fixed-effect variables to analyse the effect of plot type and distance on the $\delta^{15}N$ signature of maize leaves and soil, foliar and soil N concentrations, and the proportion of tree-derived N in maize leaves ($frac_{N(tree)}$). Further, we used linear mixed-effects models with individual tree included in the model as a random-effect variable and plot type only as a fixed-effect variable to analyse the effect of plot type on maize biomass and yield, N content and tree-derived N in maize leaves ($total_{N(tree)}$). Similarly, the impact of distance from faidherbia on the proportion of tree-derived N in maize leaves from litter-, AMF- and root-mediated processes was assessed with linear mixed-effects models with distance included in the model as a fixed-effect variable and individual tree as a random-effect variable. The Cook's distance measure was used to detect outliers. There was no significance found for the interaction between plot type and distance from faidherbia trees. Post hoc pairwise means comparisons were made using Tukey's test to calculate least-squares means (function 'lsmeans' in R package 'lsmeans') in those cases in which the main effects were significant.

Reporting Summary. Further information on research design is available in the Nature Research Reporting Summary linked to this article.

Data availability

The analysed datasets and R scripts used to analyse the data are available in the Zenodo repository: https://doi.org/10.5281/zenodo.5275322. Source data are provided with this paper.

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Author contributions

J.D., J.S., W.J.B.-H. and H.A.G. designed the experiment. J.D. and J.S. collected the data. J.D. performed data analyses with input from J.S. The manuscript was written by J.D. with input from J.S., W.J.B.-H. and H.A.G.

Competing interests

The authors declare no competing interests.

Additional information

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Study description	This study assessed the role of arbuscular mycorrhizal fungi (AMF) in making tree-derived nitrogen (N) available to crop uptake. Treatments consisted of experimental root and AMF exclusion plots located around single-standing trees that implemented three types of interaction possible between tree and crop (no belowground interaction, interaction via AMF only, full interaction between tree and crop). Within each plot, samples were collected from 5 distances. Main factors hence were type of interaction and distance-from-tree. Replication was 8 for a total of 120 experimental units.
Research sample	Faidherbia albida and maize were selected because they represent a common agroecosystem in central Malawi that has to be managed more sustainably to help attain food security.
Sampling strategy	Samples were collected at 5 distances within treatment plots. A replication of 8 was chosen because it was within the feasible range and large enough to determine treatment effects despite inherent unexplained variability between replicates.
Data collection	Soil and plant samples were collected at the time of harvest by Dr. Janina Dierks and Prof. Johan Six with assistance from local farmers. Samples were air-dried and eventually processed by Dr. Dierks.
Timing and spatial scale	Sample collection occurred at the time of maize harvest (April 2018) in order to assess the amount of tree-derived N obtained by maize within one cropping season. Trees i.e. sample locations were within a 5 km2 area.
Data exclusions	No data were excluded from the analyses.
Reproducibility	Each sample used for analyses was a composite of two samples collected from the same distance within each plot to account for potential variability across individual samples. We did not attempt to repeat the experiment.
Randomization	Eight single-standing trees were selected based on similar size criteria and sufficient distance to neighboring trees. Covariates were controlled for in the statistical model by including tree (i.e. field) as a random effect variable.
Blinding	Blinding was not used at any point in this study. This was not necessary because experimental units were sample locations.

Did the study involve field work? ☒ Yes ☐ No

Field work, collection and transport

Field conditions	The study was conducted in central Malawi, where samples were collected from agricultural fields of smallholder farmers. Sample collection occurred at the time of harvest when rainfall events were scarce and soil and plant tissue were relatively dry.
Location	The study was conducted in the village of Ndindi in the Golomoti Extension Planning Area, in the lowlands of Dedza district in central Malawi (Latitude: -14.3; Longitude: 34.6; elevation: 521 m)
Access & import/export	Before conducting the research we obtained permission from the local chiefs and extension workers. We obtained phytosanitary certificates in Malawi from the Malawi Department of Agriculture and import permits from the Swiss Bundesamt für Landwirtschaft (BLW).
Disturbance	Treatment installation (root and AMF exclusion plots) required excavation to a depth of 50 cm. This caused soil disturbance within the refined area of the plots (1 m x 5 m each). Soil excavated was placed back into the plots after installation of lining roughly to the same depth from which originally excavated. Subsequently, crops were planted and the area was managed as the remainder of the field to allow recovery.

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Validation — *Describe the validation of each primary antibody for the species and application, noting any validation statements on the manufacturer's website, relevant citations, antibody profiles in online databases, or data provided in the manuscript.*

Eukaryotic cell lines

Policy information about cell lines

Cell line source(s) — *State the source of each cell line used.*

Authentication — *Describe the authentication procedures for each cell line used OR declare that none of the cell lines used were authenticated.*

Mycoplasma contamination — *Confirm that all cell lines tested negative for mycoplasma contamination OR describe the results of the testing for mycoplasma contamination OR declare that the cell lines were not tested for mycoplasma contamination.*

Commonly misidentified lines (See ICLAC register) — *Name any commonly misidentified cell lines used in the study and provide a rationale for their use.*

Palaeontology and Archaeology

Specimen provenance — *Provide provenance information for specimens and describe permits that were obtained for the work (including the name of the issuing authority, the date of issue, and any identifying information). Permits should encompass collection and, where applicable, export.*

Specimen deposition — *Indicate where the specimens have been deposited to permit free access by other researchers.*

Dating methods — *If new dates are provided, describe how they were obtained (e.g. collection, storage, sample pretreatment and measurement), where they were obtained (i.e. lab name), the calibration program and the protocol for quality assurance OR state that no new dates are provided.*

☐ Tick this box to confirm that the raw and calibrated dates are available in the paper or in Supplementary Information.

Ethics oversight — *Identify the organization(s) that approved or provided guidance on the study protocol, OR state that no ethical approval or guidance was required and explain why not.*

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Animals and other organisms

Policy information about studies involving animals; ARRIVE guidelines recommended for reporting animal research

Laboratory animals — *For laboratory animals, report species, strain, sex and age OR state that the study did not involve laboratory animals.*

Wild animals — *Provide details on animals observed in or captured in the field; report species, sex and age where possible. Describe how animals were caught and transported and what happened to captive animals after the study (if killed, explain why and describe method; if released, say where and when) OR state that the study did not involve wild animals.*

Field-collected samples — *For laboratory work with field-collected samples, describe all relevant parameters such as housing, maintenance, temperature, photoperiod and end-of-experiment protocol OR state that the study did not involve samples collected from the field.*

Ethics oversight — *Identify the organization(s) that approved or provided guidance on the study protocol, OR state that no ethical approval or guidance was required and explain why not.*

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Human research participants

Policy information about studies involving human research participants

Population characteristics	*Describe the covariate-relevant population characteristics of the human research participants (e.g. age, gender, genotypic information, past and current diagnosis and treatment categories). If you filled out the behavioural & social sciences study design questions and have nothing to add here, write "See above."*
Recruitment	*Describe how participants were recruited. Outline any potential self-selection bias or other biases that may be present and how these are likely to impact results.*
Ethics oversight	*Identify the organization(s) that approved the study protocol.*

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Clinical data

Policy information about clinical studies

All manuscripts should comply with the ICMJE guidelines for publication of clinical research and a completed CONSORT checklist must be included with all submissions.

Clinical trial registration	*Provide the trial registration number from ClinicalTrials.gov or an equivalent agency.*
Study protocol	*Note where the full trial protocol can be accessed OR if not available, explain why.*
Data collection	*Describe the settings and locales of data collection, noting the time periods of recruitment and data collection.*
Outcomes	*Describe how you pre-defined primary and secondary outcome measures and how you assessed these measures.*

Dual use research of concern

Policy information about dual use research of concern

Hazards

Could the accidental, deliberate or reckless misuse of agents or technologies generated in the work, or the application of information presented in the manuscript, pose a threat to:

No	Yes	
☐	☐	Public health
☐	☐	National security
☐	☐	Crops and/or livestock
☐	☐	Ecosystems
☐	☐	Any other significant area

Experiments of concern

Does the work involve any of these experiments of concern:

No	Yes	
☐	☐	Demonstrate how to render a vaccine ineffective
☐	☐	Confer resistance to therapeutically useful antibiotics or antiviral agents
☐	☐	Enhance the virulence of a pathogen or render a nonpathogen virulent
☐	☐	Increase transmissibility of a pathogen
☐	☐	Alter the host range of a pathogen
☐	☐	Enable evasion of diagnostic/detection modalities
☐	☐	Enable the weaponization of a biological agent or toxin
☐	☐	Any other potentially harmful combination of experiments and agents

ChIP-seq

Data deposition

☐ Confirm that both raw and final processed data have been deposited in a public database such as GEO.

☐ Confirm that you have deposited or provided access to graph files (e.g. BED files) for the called peaks.

Data access links *May remain private before publication.*	*For "Initial submission" or "Revised version" documents, provide reviewer access links. For your "Final submission" document, provide a link to the deposited data.*
Files in database submission	*Provide a list of all files available in the database submission.*
Genome browser session (e.g. UCSC)	*Provide a link to an anonymized genome browser session for "Initial submission" and "Revised version" documents only, to enable peer review. Write "no longer applicable" for "Final submission" documents.*

Methodology

Replicates	*Describe the experimental replicates, specifying number, type and replicate agreement.*
Sequencing depth	*Describe the sequencing depth for each experiment, providing the total number of reads, uniquely mapped reads, length of reads and whether they were paired- or single-end.*
Antibodies	*Describe the antibodies used for the ChIP-seq experiments; as applicable, provide supplier name, catalog number, clone name, and lot number.*
Peak calling parameters	*Specify the command line program and parameters used for read mapping and peak calling, including the ChIP, control and index files used.*
Data quality	*Describe the methods used to ensure data quality in full detail, including how many peaks are at FDR 5% and above 5-fold enrichment.*
Software	*Describe the software used to collect and analyze the ChIP-seq data. For custom code that has been deposited into a community repository, provide accession details.*

Flow Cytometry

Plots

Confirm that:

☐ The axis labels state the marker and fluorochrome used (e.g. CD4-FITC).

☐ The axis scales are clearly visible. Include numbers along axes only for bottom left plot of group (a 'group' is an analysis of identical markers).

☐ All plots are contour plots with outliers or pseudocolor plots.

☐ A numerical value for number of cells or percentage (with statistics) is provided.

Methodology

Sample preparation	*Describe the sample preparation, detailing the biological source of the cells and any tissue processing steps used.*
Instrument	*Identify the instrument used for data collection, specifying make and model number.*
Software	*Describe the software used to collect and analyze the flow cytometry data. For custom code that has been deposited into a community repository, provide accession details.*
Cell population abundance	*Describe the abundance of the relevant cell populations within post-sort fractions, providing details on the purity of the samples and how it was determined.*
Gating strategy	*Describe the gating strategy used for all relevant experiments, specifying the preliminary FSC/SSC gates of the starting cell population, indicating where boundaries between "positive" and "negative" staining cell populations are defined.*

☐ Tick this box to confirm that a figure exemplifying the gating strategy is provided in the Supplementary Information.

Magnetic resonance imaging

Experimental design

Design type	*Indicate task or resting state; event-related or block design.*
Design specifications	*Specify the number of blocks, trials or experimental units per session and/or subject, and specify the length of each trial or block (if trials are blocked) and interval between trials.*
Behavioral performance measures	*State number and/or type of variables recorded (e.g. correct button press, response time) and what statistics were used to establish that the subjects were performing the task as expected (e.g. mean, range, and/or standard deviation across subjects).*

Acquisition

Imaging type(s) *Specify: functional, structural, diffusion, perfusion.*

Field strength *Specify in Tesla*

Sequence & imaging parameters *Specify the pulse sequence type (gradient echo, spin echo, etc.), imaging type (EPI, spiral, etc.), field of view, matrix size, slice thickness, orientation and TE/TR/flip angle.*

Area of acquisition *State whether a whole brain scan was used OR define the area of acquisition, describing how the region was determined.*

Diffusion MRI ☐ Used ☐ Not used

Preprocessing

Preprocessing software *Provide detail on software version and revision number and on specific parameters (model/functions, brain extraction, segmentation, smoothing kernel size, etc.).*

Normalization *If data were normalized/standardized, describe the approach(es): specify linear or non-linear and define image types used for transformation OR indicate that data were not normalized and explain rationale for lack of normalization.*

Normalization template *Describe the template used for normalization/transformation, specifying subject space or group standardized space (e.g. original Talairach, MNI305, ICBM152) OR indicate that the data were not normalized.*

Noise and artifact removal *Describe your procedure(s) for artifact and structured noise removal, specifying motion parameters, tissue signals and physiological signals (heart rate, respiration).*

Volume censoring *Define your software and/or method and criteria for volume censoring, and state the extent of such censoring.*

Statistical modeling & inference

Model type and settings *Specify type (mass univariate, multivariate, RSA, predictive, etc.) and describe essential details of the model at the first and second levels (e.g. fixed, random or mixed effects; drift or auto-correlation).*

Effect(s) tested *Define precise effect in terms of the task or stimulus conditions instead of psychological concepts and indicate whether ANOVA or factorial designs were used.*

Specify type of analysis: ☐ Whole brain ☐ ROI-based ☐ Both

Statistic type for inference (See Eklund et al. 2016) *Specify voxel-wise or cluster-wise and report all relevant parameters for cluster-wise methods.*

Correction *Describe the type of correction and how it is obtained for multiple comparisons (e.g. FWE, FDR, permutation or Monte Carlo).*

Models & analysis

n/a	Involved in the study
☐	☐ Functional and/or effective connectivity
☐	☐ Graph analysis
☐	☐ Multivariate modeling or predictive analysis

Functional and/or effective connectivity *Report the measures of dependence used and the model details (e.g. Pearson correlation, partial correlation, mutual information).*

Graph analysis *Report the dependent variable and connectivity measure, specifying weighted graph or binarized graph, subject- or group-level, and the global and/or node summaries used (e.g. clustering coefficient, efficiency, etc.).*

Multivariate modeling and predictive analysis *Specify independent variables, features extraction and dimension reduction, model, training and evaluation metrics.*