How can miscanthus fields be reintegrated into a crop rotation?

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Funding information
Bundesministerium für Bildung und Forschung, Grant/Award Number: 031B0163

Abstract
The bioeconomy, with its aim of replacing fossil by biobased resources, is increasingly focusing on biomass production from perennial crops, such as miscanthus. To date, research on miscanthus has explored a number of cultivation aspects; however, one major issue has not yet been addressed: How can former miscanthus fields be reintegrated into a crop rotation? This encompasses the questions of which following crop most efficiently suppresses resprouting miscanthus and what happens to the soil nitrogen content after a miscanthus removal. This study aimed to answer both questions. For this purpose, four spring crops (ryegrass, rapeseed, barley, maize) and fallow as control were cultivated after a Miscanthus sinensis removal. To test the effect of the removal on soil nitrogen content, each spring crop (excluding fallow) was divided into fertilized and unfertilized plots. After the spring crop harvest, winter wheat was cultivated to clarify which spring crop had most efficiently suppressed the resprouting miscanthus. The results indicate that fertilized crops had 35% less miscanthus biomass per hectare than unfertilized crops, probably due to the higher plant density and/or better development of the fertilized crops during the growing season. The soil mineral nitrogen (N$_{\text{min}}$) content was found to increase during the vegetation period following the miscanthus removal (average +14.85 kg/ha), but was generally on a low level. We conclude that nitrogen from miscanthus residues is partly fixed in organic matter and is thus not plant-available in the first cropping season. As some nitrogen is supplied by the decomposition of miscanthus residues, our results suggest that the crop cultivated after a miscanthus removal requires less fertilization. Of all the follow-on spring crops tested, maize coped with the prevailing soil conditions and resprouting miscanthus most efficiently, resulting in satisfactory yields, and thus seems to be a suitable crop for cultivation after miscanthus.

KEYWORDS
follow-on crop, maize, miscanthus clearance, nitrogen content, resprouting, ryegrass, spring barley, winter wheat

1 | INTRODUCTION

In the bioeconomy, which aims to replace fossil by biobased resources, there is increasing focus on biomass production by perennial crops. Miscanthus, for example, fulfils several ecological functions, such as soil carbon sequestration and erosion control through its year-round soil coverage and cultivation period of up to 20 years (Lewandowski, Kicherer, & Vonier,
These result in positive effects including improved soil structure and reduced nutrient run-off (McCalmont et al., 2017). Various studies have reported a carbon sequestration under miscanthus in the range of 0.5–2.2 t C ha$^{-1}$a$^{-1}$ (Blanco-Canqui, 2010; McCalmont et al., 2017). Additionally, miscanthus is high yielding. Schmidt, Lemaigre, Ruf, Delfosse, and Emmerling (2018) have shown that, in temperate climates, yields of between 22 t ha$^{-1}$ a$^{-1}$ (brown harvest after winter) and 27 t ha$^{-1}$ a$^{-1}$ (green harvest in autumn) can be achieved.

It is these high yields together with environmentally benign characteristics that render miscanthus a promising bioeconomy crop with a number of utilization options. Currently, its most common utilization pathway is combustion (Iqbal & Lewandowski, 2016). However, other potential pathways include ethanol production (van der Weijde et al., 2017), anaerobic digestion (Mangold et al., 2018; Mayer et al., 2014) and building materials.

Most recent studies on miscanthus have focused on its improvement for those utilization pathways or dealt with agronomic aspects such as establishment options (Boersma & Heaton, 2014; Clifton-Brown, Hastings, & Mos, 2017; Xue, Kalinina, & Lewandowski, 2015), general cultivation practices including row distance and fertilization (Larsen, Jørgensen, Kjeldsen, & Lærke, 2014), or examined the performance of various genotypes on different site conditions (Lewandowski et al., 2016).

Although miscanthus has been explored from these different perspectives, one main research question has to date been almost neglected: How can former miscanthus fields be reintegrated into rotations with annual crops?

This question of the reintegration or removal of former miscanthus fields has only been dealt with in a few studies. McCalmont et al. (2018), for example, investigated the nitrous oxide emissions after a miscanthus removal. Dufossé, Drewer, Gabrielle, and Drouet (2014) analysed the effect of a miscanthus removal on soil nutrient stock, greenhouse-gas emissions and the yield of the following crop (wheat). Drewer, Dufossé, Skiba, and Gabrielle (2016) examined the effect of a removal on the isotopic signature of soil carbon.

In the three studies mentioned above, miscanthus was removed by the application of glyphosate. However, glyphosate application is currently the subject of controversial debate as negative environmental impacts are expected (Myers et al., 2016). Therefore, the question arises as to how miscanthus can be cleared from the field at the end of its lifetime without using glyphosate. This question is crucial for farmers, as the resprouting of miscanthus through its rhizomes may impair follow-on crops. The problem can be seen in the example of *Elymus repens* L., a rhizomatous perennial weed, which can cause high yield losses in crops, but can be successfully removed by glyphosate application (Ringselle, Bergkvist, Arosson, & Andersson, 2015). Ringselle et al. (2015) showed that soil tillage or covering by crops can be a nonchemical alternative to reduce *E. repens*. Therefore, the question arises whether tillage and the cultivation of follow-on crops can also suppress resprouting miscanthus.

An additional important question is the effect of a previous crop on soil nitrogen (N) content. In Germany, this has become particularly important since the recent release of the amendment to the Fertiliser Application Ordinance (Bundesministerium für Ernährung und Landwirtschaft (BMEL), 2019). Today, there is still little information available on the effect of miscanthus cultivation and its subsequent removal on soil N content. It is well known that the removal of permanent grassland leads to an increase in soil N content (Seidel, Kayser, Müller, & Isselstein, 2009). An increase is to be expected when both living and dead plant material as well as soil organic matter are mineralized (Seidel et al., 2009). This, in turn, can lead to nitrate leaching (Seidel et al., 2009). A removal of permanent crops, such as grassland or miscanthus, may also lead to high nitrous oxide emissions (Dufossé et al., 2014; McCalmont et al., 2018; Pinto et al., 2004; Vellinga, Pol-van, & Kuikman, 2004).

On the other hand, it is known from cereal cultivation that if straw (which has high carbon content) is left on the field and the soil N content is low, the following crops are negatively affected as soil bacteria use up the N for the decomposition of the straw (Reinertsen, Elliott, Cochran, & Campbell, 1984). This may also be the case after a miscanthus removal, as rhizomes and litter remain on the field. If N content is low and the follow-on crop is not adequately fertilized, it may suffer from N shortage.

It is currently unclear which effect is to be expected in the first year after a miscanthus removal: high N availability as observed after a grassland removal or a possible N shortage as in some cases of cereal cultivation.

The aim of this study was to answer both questions mentioned above. (a) Can resprouting miscanthus be suppressed by soil tillage and the cultivation of follow-on crops? (b) What is the soil N availability for a follow-on crop after a miscanthus removal?

## 2 | MATERIALS AND METHODS

### 2.1 | Field trial

The field trial was conducted at the University of Hohenheim’s research station ‘Ihinger Hof’ (48.7° latitude, 8.9° longitude, approximately 475 m a.s.l.). The location is characterized by a long-term average annual air temperature of 9.5°C and an annual precipitation of 720 mm. The soil is classified as luvisol (soil type: loam; pH: 6.9) and soil nutrient content of both P and K was classified as ‘C’ according to VDLUFA (Association of German Agricultural Analytic and Research Institutes) soil classification. The weather conditions during the field trial, shown
on a monthly basis from February 2017 to July 2018, can be found in Table S1.

The miscanthus genotype OPM-111 (polycross of parents selected in Indiana, US from five M. sinensis accessions collected in central Japan) was planted in May 2013 and, after 4 years of cultivation, removed by ploughing. As the miscanthus stock was inhomogeneous, the density was determined by counting plants/m² in each plot on 15 December 2016 to test whether there were significant differences between the plots before the removal was carried out.

The miscanthus field was harvested on 15 February 2017 using a field chopper. Two weeks later, the field was ploughed to a depth of 20 cm and harrowed with a rotary power harrow. Afterwards, a split-plot design was used to allocate spring crops and fertilization treatments to the field. All variants of the follow-on spring crops (annual ryegrass, spring barley, summer rapeseed, maize; each both fertilized and unfertilized) were then sown in strip plots in a randomized complete block design with four replicates (Figure 1). The strip plots of the spring crops (main factor) were randomly divided into unfertilized and commonly fertilized subplots. The plot size was 18 m² (6 m × 3 m) for ryegrass (Lolium perenne L.), barley (Hordeum vulgare L.), rapeseed (Brassica napus L.) (fertilized/unfertilized), and the control fallow (unfertilized) and 27 m² (6 m × 4.5 m) for maize (Zea mays L.) (fertilized/unfertilized), where it was adopted to the working width of the sowing machine. Details of the spring crop cultivation are given in Table 1.

The rapeseed stock was inhomogeneous and weeds dominated each plot, probably due to the low temperatures in April (down to −5°C on 20 April 2017). For this reason, we decided to omit rapeseed from the N analysis.

For the harvest of all spring crops and fallow, 2 m² was cut from the middle of each plot and the harvested biomass was then divided into the fractions given in Table 1. The entire biomass in the 2 m² was harvested with an electric cutter at a cutting height of 5 cm. The biomass was then transported to the laboratory for separation. It was weighed, chopped and dried at 60°C to constant weight to estimate the dry matter content (DMC). From the DMC, the dry matter yield (DMY) was calculated. The barley was dried as whole crop and then threshed to determine grain and straw yield.

After the harvest of the 2 m², the rest of the biomass from each plot was cut on the same day and discarded.

In autumn 2017, all plots were ploughed and, 3 days later, sown with winter wheat. Details of winter wheat cultivation are given in Table 2.

At the winter wheat harvest, the same 2 m² as in the previously grown spring crops were cut, supported by GPS tracking and separated into wheat and miscanthus. The biomass fractions were weighed and dried at 60°C to constant weight for DMC estimation. The wheat was then threshed to determine straw and grain yields. The DMY of grain, straw and, where present, miscanthus was calculated from the DMC.

Figure 1 gives a schematic overview of the field trial for the 2 years 2017 and 2018.

2.2 | Nitrogen content of soil and crop

The soil N_min (NO₃-N) content was analysed (following guideline VDLUFA A 6.1.4.1) in each plot twice: after the miscanthus harvest/before sowing of spring crops (20 February 2017) and after the spring crop harvest (16 October 2017). Soil samples were taken at three depths (0–30, 30–60 and 60–90 cm). For N_min content determination, a CaCl₂ extraction was performed with the fresh soil and the N_min measured using a flow injection analyser. Afterwards, N_min content per hectare was calculated using a bulk density of 1.4 g/cm³.

Nitrogen contents of the follow-on spring crops ryegrass (first and second cut), barley (grain and straw) and maize (whole plant) were analysed using a Vario Max CNS (Elementar Analysensysteme GmbH, Langenselbold, Germany), as described in the VDLUFA Method Book III, method 4.1.2 and DIN ISO 5725.
| Spring crop | Sowing | Fertilization | Crop protection | Harvest |
|-------------|--------|---------------|----------------|---------|
| Ryegrass +N | 16 March 2017 45 kg/ha variety ‘Pollanum’ | 24 April 2017 first fertilization | None | 4 July 2017 first harvest<sup>a</sup> |
|             | Sown with a seed drill combination Rolled 1 day later using a front roller | 6 July 2017 second fertilization Each time: 50 kg N/ha via calcium ammonium nitrate (27% N) | | 20 September second harvest<sup>a</sup> |
|             | 23 May 2017 cut to stimulate growth (grass was removed) | | | Each time biomass divided into fractions ‘ryegrass’ and ‘miscanthus’ |
|             | | None | | Weeds assigned to ryegrass, as only present in small amounts |
| Barley +N   | 16 March 2017 168 kg/ha variety ‘RGT Planet’ (malting barley) | 24 April 2017 70 kg N/ha via calcium ammonium nitrate (27% N) | 4 May 2017 herbicides ‘Axial’ 50 (0.9 L/ha; Syngenta; components: pinoxaden, cloquintocet) ‘Alliance Suprim’ (0.1 kg/ha; Nufarm; components: diflufenican, metsulfuron) 8 June 2017 Fungicides ‘Aviator Xpro’ (0.6 L/ha; Bayer; components: bixafen, prothioconazole) ‘Fandango’ (0.6 L/ha; Bayer; components: prothioconazole, fluoxastrobin) Insecticide ‘Lambda WG’ (150 g/ha; Syngenta; component: lambda-cyhalothrin) | 24 July 2017 Biomass divided into fractions ‘barley’, ‘miscanthus’ and ‘weeds’ |
| Barley −N   | 16 March 2017 | | | |
| Rapeseed +N | 27 March 2017 4.78 kg/ha variety ‘Makro’ | 24 April 2017 first fertilization (70 kg N/ha) 14 June 2017 second fertilization (50 kg N/ha) Each time via calcium ammonium nitrate (27% N) | 4 May 2017 herbicide ‘Butisan’ (1.5 L/ha; BASF; component: metazachlor) 14 June 2017 insecticide ‘Biscaya’ (0.3 L/ha; Bayer; component: thiacloprid) | 20 September 2017 Biomass divided into fractions ‘miscanthus’ and ‘remaining biomass’, which included rapeseed (whole crop due to inhomogeneous rapeseed stocks) and weeds |
| Rapeseed −N | | | | |
| Maize +N    | 17 May 2017 Plots harrowed with rotary power harrow 18 May 2017 93,200 grains/ha variety ‘Ridley’ Sown with a pneumatic single grain seeder | 22 May 2017 first fertilization (120 kg N/ha) 14 June 2017 second fertilization (120 kg N/ha) Each time via calcium ammonium nitrate (27% N) | 13 June 2017 herbicides MaisTer Power (1.4 L/ha; Bayer; components: foramsulfuron, thiencarbazone, iodosulfuron, cyprosulfamid) Bromotril 225 EC (0.3 L/ha; Adama; component: bromoxynil) | 20 September 2017 Biomass divided into fractions ‘miscanthus’ and ‘maize’ |
| Maize −N    | | | | Weeds were neglected due to their small amount |
| Fallow      | None | None | None | 20 September 2017 Biomass divided into the fractions ‘miscanthus’ and ‘remaining biomass’ |

<sup>a</sup>The same 2 m²—tracked by GPS—were harvested on both harvest dates.
### TABLE 2  Overview of winter wheat cultivation

| Sowing                                                                 | 16 October 2017 |
|-----------------------------------------------------------------------|------------------|
| 157 kg/ha variety ‘Rebell’                                            | Sown with a seed d’reyegrass; rill combination |
| Fertilization                                                         | 3 April 2018     |
| 150 kg/ha urea (69 kg N/ha)                                           | 1st fertilization |
| 70 kg N/ha                                                            | 2nd fertilization |
| 40 kg N/ha                                                            | 3rd fertilization |
| 2nd and 3rd fertilization via calcium ammonium nitrate (27% N)        |                  |

### 2.3  Statistical analysis

Data analysis was performed using the PROC MIXED procedure of Statistical Analysis Software SAS, version 9.4 (SAS Institute Inc.). Normal distribution and homogeneity of residuals were checked graphically.

For the statistical analysis of miscanthus regrowth, DMY of spring crops and winter wheat, the model given in Equation (1) was used. Crop, fertilization and their interactions were taken as fixed effects. The effect of miscanthus density determined before crop removal was taken as covariable. It was found to be nonsignificant in each test.

\[
y_{ihek} = \mu + g_i + d_h + (gd)_{ih} + fx_{ihk} + s_k + (sg)_{ik} + e_{ihk}, \tag{1}
\]

where \(y_{ihek}\) is the measurement for the \(i\)-th crop on the \(h\)-th fertilization level in the \(k\)-th field replicate. \(\mu\) is the general effect, \(g_i\) is the \(i\)-th crop effect (ryegrass; barley, maize, fallow land; rapeseed), \(d_h\) is the main effect of the \(h\)-th fertilization level (fertilized; unfertilized), \((gd)_{ih}\) is the interaction effect of the \(i\)-th crop with the \(h\)-th fertilization level, \(f\) is the slope of miscanthus density \(x_{ihk}\) before removal and \(s_k\) is the random effect of the \(k\)-th replicate in the field. \(e_{ihk}\) and \((sg)_{ik}\) are the residual and main plot error terms corresponding to \(y_{ihek}\) and the \(ik\)-th main plot.

For the N content, the model is slightly more complicated, as (a) ryegrass was cut twice and each cut analysed separately; and (b) barley was subdivided into grain and straw and each part analysed separately. Therefore, \(g_i\) is split into \(g_i\) and \((gm)_{il}\) or \(g_i\) and \((gn)_{io}\) with \(m_i\) being the effect of the \(i\)-th harvest date and \(n_o\) being the effect of the \(o\)-th effect of plant part.

\[
y_{ihlok} = \mu + g_i + d_h + (gd)_{ih} + fx_{ihk} + (gm)_{il} + (gd)_{ihl} + (gn)_{io} + (gdn)_{ilo} + s_k + (sg)_{ik} + e_{ihlok}. \tag{2}
\]

For the soil N\(_\text{min}\) content, soil sampling date \(p\) was added to give Equation (3).

\[
y_{ihqk} = \mu + g_i + d_h + p_q + (gd)_{ih} + (gp)_{iq} + (dp)_{hq} + (gd)_{ihq} + fx_{ihk} + s_k + (sg)_{ik} + e_{ihqk}. \tag{3}
\]

where \(p_q\) is the \(q\)-th soil sampling date (23 February 2017 or 16 October 2017).

Effects of field replicates were assumed to be random. Multiple \(t\) tests with a significance level of \(\alpha = .05\) were conducted only where differences were found via a global \(F\) test. In the figures, a letter display was used where identical letters indicate that means are not significantly different from each other.

### 3  RESULTS

#### 3.1  Resprouting miscanthus in follow-on spring crops

The average miscanthus density before clearance was 0.8 plants/m\(^2\) (Figure 2a). The highest density was found before ryegrass +N (0.96 plants/m\(^2\)) and the lowest before ryegrass –N (0.66 plants/m\(^2\)). As mentioned above, miscanthus density was taken as a covariable in each statistical analysis and, in each analysis, was found to have no significant effect (see Table 3).

Figure 2b shows the average number of miscanthus stems, counted in July 2017, in the follow-on spring crops. As shown in Table 3, crop (0.0032) and fertilization (0.0005) had a significant effect on resprouting, but their interaction did not. The lowest number of miscanthus stems was found in barley +N (0.61 stems/m\(^2\)), although the former miscanthus plant density was quite high (0.85 plants/m\(^2\)). The highest number of miscanthus stems was found in rapeseed –N (12.24 stems/m\(^2\)) (Figure 2b), where the former miscanthus plant density...
Comparing fertilized with unfertilized crops, (significantly) less stems were found in fertilized than in unfertilized plots for each crop except for maize (see upper-case letters in Figure 2b). The miscanthus resprouting in the follow-on crops can be seen in Figure 3.
FIGURE 3  Resprouting miscanthus in follow-on spring crops after miscanthus removal in annual ryegrass (a), spring barley (b), summer rapeseed (c), maize (d) and fallow (e) in 2017. (f) shows the resprouting miscanthus in winter wheat, sown in autumn 2017 after harvest of follow-on spring crops (a–e)

TABLE 4  Dry matter yield (DMY) and biomass amount (dry matter = DM) of spring crops, miscanthus and winter wheat. Yield/biomass amount is shown in kg/ha for miscanthus and in t/ha for all other crops. Different upper-case letters indicate significant differences between fertilized and unfertilized variant of same (previous) spring crop (NS = not significantly different, $\alpha = .05$). Different lower-case letters indicate significant differences between spring crops (fertilized crops are compared with each other; unfertilized crops including fallow are compared with each other)

| (Previous) spring crop | DMY (t/ha) | Biomass amount (DM), kg/ha | DMY, t/ha | Biomass amount (DM), kg/ha |
|------------------------|------------|-----------------------------|------------|-----------------------------|
|                        | Spring crop | Miscanthus (in spring crop) | Winter wheat (grain) | Miscanthus (in winter wheat) |
| Ryegrass +N            | 3.95 ± 1.20$^{NS,a}$ | 130 ± 320$^{b,a}$ | 5.00 ± 0.48 | 4.92 ± 4.09$^{ns}$ |
| Ryegrass −N            | 2.47 ± 1.20$^{NS,a}$ | 870 ± 320$^{bc,e}$ | 5.68 ± 0.48 | 4.20 ± 4.79$^{b}$ |
| Barley +N              | 2.54 ± 1.17$^{b,\text{(grain)}}$ | 70 ± 310$^{b}$ | 5.43 ± 0.45 | 0.00 ± 3.90$^{ns}$ |
| Barley −N              | 0.69 ± 1.16$^{b}$ (grain) | 380 ± 310$^{c}$ | 5.63 ± 0.45 | 0.78 ± 3.90$^{b}$ |
| Maize +N               | 26.92 ± 1.17$^{A}$ (whole crop) | 360 ± 310$^{b}$ | 6.57 ± 0.45 | 2.10 ± 3.91$^{ns}$ |
| Maize −N               | 17.54 ± 1.17$^{b}$ (whole crop) | 390 ± 310$^{c}$ | 5.72 ± 0.45 | 7.60 ± 3.89$^{ab}$ |
| Fallow                 | 1.31 ± 1.10 (total biomass) | 1,480 ± 250$^{ab}$ | 5.22 ± 0.31 | 17.34 ± 2.75$^{a}$ |
| Rapeseed +N            | 2.50 ± 1.19$^{NS}$ (whole crop ± weeds) | 1,760 ± 320$^{a}$ | 5.41 ± 0.46 | 1.34 ± 4.00$^{ns}$ |
| Rapeseed −N            | 0.97 ± 1.18$^{NS}$ (whole crop ± weeds) | 1,930 ± 310$^{a}$ | 4.89 ± 0.46 | 3.42 ± 3.95$^{b}$ |

*Average of first and second cut; the same 2 m²—tracked by GPS—were harvested on both harvest dates.
3.2 Yield of spring crops and winter wheat, including miscanthus biomass

At the harvest of each follow-on spring crop, the biomass amounts of both the crop and miscanthus were determined. As the rapeseed developed poorly, all biomass growing on the plot was harvested with the rapeseed being harvested as whole crop (not, as usual, only the seeds).

Table 4 gives an overview of the DMY of each follow-on spring crop. Fertilization and the interactions of Fertilization × Crop had a significant effect on the DMY (see Table 3) and resulted in higher DMY for fertilized than unfertilized spring crops. For maize and barley (grain), this effect resulted in a significantly higher DMY for the fertilized than the unfertilized variant (see upper-case letters in Table 4).

The amount of miscanthus in the follow-on spring crops was significantly affected by crop (Table 3). With an average of 1,840 kg DM/ha, rapeseed had the significantly highest amount of miscanthus in all fertilized and unfertilized crops (see lower-case letters in Table 4). The lowest amount of miscanthus biomass was found in barley +N (70 kg/ha) (see Table 4). Fertilization resulted in lower amounts of miscanthus in all crops, but this effect was not significant. The average amount of miscanthus was 580 kg/ha in fertilized crops and 890 kg/ha in unfertilized crops (excluding fallow).

The DMY of the winter wheat (grain) cultivated after the spring crops was not significantly affected by the (previous) crop, fertilization or the interaction of both (see Table 3). The DMY of wheat (grain) for each previous crop is shown in Table 4.

In general, the amount of resprouting miscanthus in winter wheat was low (average: 4.63 kg/ha). It was significantly affected by the previous crop (Table 3). The highest amount of miscanthus biomass was found on previously fallow land (17.34 kg DM/ha, Table 4). The amount of miscanthus biomass was lower in all previously fertilized plots (2.09 kg/ha) than unfertilized plots (4.0 kg/ha, excluding fallow), except for ryegrass. The previous barley +N plots were entirely free of miscanthus biomass (Table 4).

3.3 Soil $N_{\text{min}}$ content and N content of spring crops

Table 5 gives the $p$-values for $F$ tests of fixed effects for each soil depth and also for the total (0–90 cm). Table 6 shows the $N_{\text{min}}$ contents (NO$_3$-N) of the three depths (0–30, 30–60, 60–90 cm) and the total $N_{\text{min}}$ (0–90 cm) determined after the miscanthus harvest (d1; 23 February 2017) and after the spring crop harvest (d2; 16 October 2017). The average $N_{\text{min}}$ contents measured on both d1 (2.4 kg/ha) and d2 (17.27 kg/ha) were generally low. $N_{\text{min}}$ content increased significantly from d1 to d2 in all unfertilized plots at each soil depth (see Table 5). This is particularly visible in fallow land, which was not fertilized: the $N_{\text{min}}$ amount increased from 2.0 kg $N_{\text{min}}$/ha in the first assessment to 38.71 kg $N_{\text{min}}$/ha in the second (Table 6). This was the highest $N_{\text{min}}$ amount found in all plots, both fertilized and unfertilized. The higher $N_{\text{min}}$ contents of unfertilized than fertilized crops at d2 (for example in ryegrass, barley, rapeseed 0–30 cm) were not significant and probably reflect a natural fluctuation.

In addition, Table 5 shows that fertilization and miscanthus density before removal did not significantly affect the soil $N_{\text{min}}$ content. In Table 6, there is no letter display, as significant differences were only found for soil sampling date and between different (fertilized) crops (which was expected and is thus not shown by letters) but not for fertilization itself (Table 5), which was the focus of our study. (Differences between unfertilized crops only occurred for fallow, which had a significant higher $N_{\text{min}}$ content than other crops at 0–30 and 0–90 cm.)

Figure 4 shows the N content of the spring crops ryegrass (first and second cut), barley (grain and straw) and maize (whole crop). As shown in Table 3, crop and the interactions of crop with fertilization had a significant effect on the N content, shown by the letter display in Figure 4. Fertilization itself, however, did not have a significant effect on the N content.

In ryegrass, the N content was higher in unfertilized (1.4% of DM; average of harvest date (HD)1 and HD2) than fertilized (1.2% of DM; average of HD1 and HD2) crops, but this was only significant at HD2. A comparison of the two

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**TABLE 5** $p$ Values for $F$ tests of fixed effects ($\alpha = .05$) for soil $N_{\text{min}}$ content of samples taken on 23 February 2017 and 16 October 2017, at three soil depths (0–30; 30–60; 60–90 cm) and in total (0–90 cm)

| Depth in cm | Crop | Fertilization | Date | Crop × Fertilization | Crop × Date | Fertilization × Date | Crop × Fertilization × Date | Density |
|-------------|------|--------------|------|----------------------|-------------|----------------------|----------------------------|--------|
| 0–30        | 0.0069 | 0.3273      | <0.0001 | 0.0091                | 0.0087      | 0.0954                | 0.0040                      | 0.0735 |
| 30–60       | 0.3255 | 0.7520      | 0.0004  | 0.9155                | 0.1153      | 0.9656                | 0.9482                      | 0.8503 |
| 60–90       | 0.1287 | 0.7861      | <0.0001 | 0.6355                | 0.0093      | 0.7277                | 0.5582                      | 0.9332 |
| 0–90        | 0.0332 | 0.7455      | <0.0001 | 0.3554                | 0.0011      | 0.4341                | 0.3309                      | 0.6071 |
cuts (average of fertilized/unfertilized crops) showed the N content of the first cut (1.36% of DM) to be higher than the second (1.26% of DM). In barley, the N content of fertilized crops (grain: 1.64% of DM; straw: 0.61% of DM) was higher than for unfertilized crops (grain: 1.37% of DM; straw: 0.49% of DM) (see Figure 4). In maize, the N content was similar in unfertilized (0.94% of DM) and fertilized crops (0.91% of DM) (Figure 4).

Using DMY to calculate the N removed by the harvested biomass gives a higher N removal for fertilized than unfertilized plots for ryegrass, barley and maize, due to the overall higher yield. Maize had the highest removal at, on average, 206 kg N/ha. This was followed by ryegrass which removed on average 25 kg N/ha (first cut) and 17 kg N/ha (second cut). Barley had an average N removal of 26 kg/ha (grain) and 5 kg/ha (straw).

### DISCUSSION

This study analysed how former miscanthus fields can be reintegrated into annual crop rotations. The aim was to determine whether respreading miscanthus can be suppressed by soil tillage and the cultivation of follow-on annual crops and what happens to soil N content after miscanthus removal. The results showed that the miscanthus stand was successfully cleared by the follow-on cultivation of ryegrass, barley or maize. In addition, it was shown that, after the miscanthus removal, mineral nitrogen (N$_{\text{min}}$) content increased significantly from the first to the second soil sampling, not only in fertilized but also in unfertilized plots. However, the N$_{\text{min}}$ contents were generally on a low level after the miscanthus removal.

#### TABLE 6

|                  | kg N$_{\text{min}}$/ha | 0–30 cm | 30–60 cm | 60–90 cm | Total 0–90 cm |
|------------------|-------------------------|---------|----------|----------|--------------|
|                  |                         | d1      | d2       | d1       | d2           | d1       | d2       |
| Ryegrass +N      | 1.01 ± 1.7              | 4.25 ± 1.7 | 0.81 ± 3.8 | 2.72 ± 2.6 | 0.65 ± 3.6 | 8.05 ± 2.6 | 1.91 ± 4.9 | 15.02 ± 4.9 |
| Ryegrass −N      | 1.01 ± 1.7              | 4.78 ± 1.7 | 0.84 ± 2.7 | 2.72 ± 2.7 | 0.81 ± 2.6 | 2.70 ± 2.6 | 2.67 ± 4.9 | 10.26 ± 4.9 |
| Barley +N        | 0.96 ± 1.7              | 4.43 ± 1.7 | 0.50 ± 2.6 | 3.95 ± 2.6 | 1.21 ± 2.6 | 1.87 ± 2.6 | 2.66 ± 4.9 | 10.23 ± 4.9 |
| Barley −N        | 1.23 ± 1.8              | 6.14 ± 1.8 | 1.30 ± 2.6 | 7.11 ± 2.6 | 1.05 ± 2.6 | 1.21 ± 2.6 | 3.56 ± 5.0 | 18.97 ± 5.0 |
| Maize +N         | 0.85 ± 1.7              | 17.55 ± 1.7 | 0.33 ± 2.6 | 4.98 ± 2.6 | 0.71 ± 2.6 | 2.13 ± 2.6 | 1.88 ± 4.9 | 24.66 ± 4.9 |
| Maize −N         | 1.88 ± 1.7              | 4.04 ± 1.7 | 0.86 ± 2.6 | 3.67 ± 2.6 | 0.39 ± 2.9 | 1.96 ± 2.6 | 3.21 ± 4.9 | 9.67 ± 4.9 |
| Fallow           | 1.01 ± 1.3              | 10.06 ± 1.2 | 0.57 ± 2.1 | 12.69 ± 1.9 | 0.31 ± 2.7 | 15.97 ± 2.0 | 2.0 ± 3.7 | 38.71 ± 3.7 |
| Rapeseed +N      | 0.64 ± 1.8              | 3.02 ± 1.8 | 0.50 ± 2.7 | 3.57 ± 2.7 | 0.52 ± 3.8 | 7.18 ± 2.7 | 1.40 ± 5.1 | 13.74 ± 5.1 |
| Rapeseed −N      | 1.45 ± 1.8              | 4.87 ± 1.8 | 0.64 ± 3.8 | 3.65 ± 2.7 | 1.14 ± 3.0 | 5.70 ± 2.7 | 2.61 ± 5.1 | 14.25 ± 5.1 |

**FIGURE 4** Nitrogen content (% of DM) of biomass from the three spring crops ryegrass (first harvest date (HD1) and second HD2 cut), spring barley (grain and straw), and maize (whole crop), for both fertilized (+N) and nonfertilized (−N) variants. Error bars represent standard error. Letter display indicates significant differences between fertilized and unfertilized variant of same previous spring crop for same harvest date or plant part (NS = not significantly different, $\alpha = .05$)
The following sections discuss (a) the effect of crop management and crop competition on the suppression of resprouting miscanthus; and (b) N management after a miscanthus removal.

4.1 Effect of crop management and crop competition on resprouting of miscanthus

The results of this study show that less miscanthus regrew in the fertilized than unfertilized follow-on spring crops, although this effect was not significant. This trend also carried through to the winter wheat cultivated in the next season, except for the previous ryegrass +N plot. This was most likely due to the higher vigour of fertilized than unfertilized crops, resulting in a higher plant density and/or better development of plant stocks over the growing season, as it is well known that a higher vigour of plants leads to more efficient weed suppression (e.g. Bertholdsson, 2005).

Another finding of the study was that barley +N had the lowest miscanthus regrowth on all three assessment dates (during growing season and at harvest of spring crops; at harvest of winter wheat). The highest miscanthus biomass was found in rapeseed (+/− N) and in fallow land. Due to the poor establishment and low crop density of rapeseed, its effect in suppressing resprouting miscanthus was not much different to that of fallow land.

A comparison of the three crops maize, barley and ryegrass reveals how two different ‘mechanisms’ of miscanthus suppression become relevant: the effect of ‘crop management’ on the one hand and of ‘crop competition’ on the other.

In maize, both mechanisms applied. Initially, the mechanism ‘crop management’ took effect as maize was sown latest of all crops. The miscanthus that had sprouted up until the sowing of maize was removed mechanically by a second harrowing of the field before sowing. Thus, the miscanthus rhizomes were first reduced to smaller pieces, resulting in a lower resprouting rate (Isensee, Ohls, & Quas, 1994). Secondly, the harrowing brought the rhizomes up to the soil surface and the consequent drying out also hindered resprouting (Isensee et al., 1994). However, once the maize was sown, the miscanthus was able to develop until the maize had sprouted and covered the whole soil surface. Later, when the maize canopy had closed, the mechanism of ‘crop competition’ came into effect: the miscanthus was efficiently suppressed, as the greater plant height of maize enabled it to intercept the photosynthetically active radiation more successfully (Bertholdsson, 2005). This resulted in a comparatively low number of miscanthus stems (counted on 4 July) and ultimately a low proportion of miscanthus in the total biomass at harvest, mainly due to the high biomass yield of maize (up to 26.92 t DM/ha for maize +N).

In ricegrass, the mechanism of ‘crop management’ contributed to the efficient removal of miscanthus. Its resprouting was impaired by mowing the ricegrass three times: at the growth-stimulating cut in mid-May and the two harvests in July and September. After each cut, the miscanthus started to regrow, finally leading to its exhaustion (Isensee et al., 1994). As shown by Ringselle et al. (2015), E. repens can be reduced by 50% through the cultivation of ryegrass if this is cut twice. Our study showed that miscanthus regrowth can also be sufficiently suppressed by mowing. The average proportion of miscanthus in total biomass was 6.8% for ryegrass +N and 23.7% for ryegrass −N. However, the amount of miscanthus biomass increased from the first to the second harvest (+230 kg DM/ha in ryegrass +N; +540 kg DM in ryegrass −N). This is most likely due to the lower ryegrass yield, which decreased from the first to the second harvest in both variants (−0.5 t DM/ha for ryegrass +N; −0.45 t DM/ha for ryegrass −N), leading to a less efficient suppression of miscanthus. As the ryegrass plots were cut three times (growth-stimulating cut, first and second harvest), we would have expected the amount of miscanthus biomass to decrease. Therefore, further research should clarify whether the timing of the mowing is also important and affects miscanthus regrowth, as mentioned by Ringselle et al. (2015) for E. repens.

In barley, the mechanism of ‘crop competition’ was predominantly effective. Barley was able to prevent the resprouting of miscanthus by its stronger competition for light, water and nutrients, probably due to its fast tillering, initial shoot and root growth rates and its high stock density (Bertholdsson, 2005; Isensee et al., 1994; Seavers & Wright, 1999). The miscanthus proportion in fertilized barley was 3.4% and, as mentioned above, it had the lowest amount of miscanthus biomass of all crops. Therefore, we conclude that barley was the most effective of all tested crops at suppressing resprouting miscanthus after a removal.

However, at 2.65 t DM/ha (+N) and 0.72 t DM/ha (−N), the yield of barley grain in our study was low compared, for example, to an average yield of 7.34 t DM/ha for the barley variety ‘RGT Planet’ at the research station ‘Ihinger Hof’ in 2017. One reason for the low yield in our field trial was that the developed green ears of some barley plants had fallen onto the soil surface. The ryegrass yield in our study (3.21 t DM/ha; first plus second cut) was also quite low. By contrast, Dufossé et al. (2014) found no difference in wheat grain yield between that grown after miscanthus and a control. As we have no explanation for the ‘ear loss’ in barley and the low ryegrass yield in our study, further research is required to clarify whether both were caused by the previous miscanthus crop or by other environmental conditions.

It should also be mentioned that maize, barley, rapeseed and winter wheat were sprayed with herbicides (MaisTer Power, Axial, Butisan and Atlantis respectively) effective
against grassy weed species. These herbicides had a potential effect on miscanthus resprouting and formed part of crop management.

The miscanthus stand in our study was 4 years old, which means that it was comparatively young. We assume that young miscanthus rhizomes have a higher resprouting rate than in older stands. However, further research is necessary to analyse how crop age influences resprouting of miscanthus rhizomes after removal. Additionally, it should be clarified whether other genotypes, for example, *Miscanthus x giganteus*, have higher resprouting rates than the *M. sinensis* genotype used in our study, as *M. sinensis* grows in tufts.

In summary, we found that different mechanisms are effective in suppressing resprouting miscanthus. Intensive soil tillage in a late-sown crop such as maize can destroy miscanthus that has already sprouted. Frequent mowing, as conducted in ryegrass, can probably exhaust the regrowth, as suggested by Isensee et al. (1994). A dense plant population, as in barley, can suppress miscanthus through competition for nutrients, light and water. Which of the spring crops is cultivated after miscanthus is each farmer’s individual decision and will be linked to the farm’s fodder requirements. However, as the yield of the following crop is likely to be a crucial criterion for the farmer, we recommend cultivating maize, due to its high yields and sufficient miscanthus suppression.

### 4.2 | How to optimize nitrogen management after a miscanthus removal

As mentioned in the introduction, the new German fertiliser ordinance requires the possible N provision from previous crops to be considered for the cultivation of follow-on ones. However, it is not clear whether soil N increases after a miscanthus removal, as with grassland removal, or whether a high C:N ratio leads to a N deficiency, as is sometimes the case after cereal cultivation, impairing a follow-on crop.

The results of our study show that soil mineralized nitrogen (N$_{\text{min}}$) content increased significantly from the first to the second soil sampling in unfertilized as well as fertilized plots at all tested soil depths (Table 5). For example, the total soil N$_{\text{min}}$ content of (unfertilized) fallow land increased from 2 kg/ha (after miscanthus harvest; 20 February 2017) to 38.71 kg/ha (before wheat sowing; 16 October 2017) (Table 6). According to Dufossé et al. (2014), the increase in N$_{\text{min}}$ content can be attributed to the miscanthus residues, such as litter, roots and rhizomes, remaining on the field. In our study, the amounts were approximately 1.03 t DM/ha (N content about 12 kg/ha) for litter and 4.77 t DM/ha (N content about 27 kg/ha) for roots and rhizomes (results not shown). Dufossé et al. (2014) found a steady increase in N$_{\text{min}}$ content from January to March 2012 (miscanthus removal was conducted in three steps between June and September 2011), probably due to increasing temperatures, and reached a peak of 200 kg N$_{\text{min}}$/ha in April/May 2012. According to these findings, the increasing N content of unfertilized plots in our study can be attributed to the miscanthus residues, which were most likely mineralized over the vegetation period of the follow-on spring crops, increasing the soil N$_{\text{min}}$ content.

However, the increase in soil N$_{\text{min}}$ from d1 to d2 remained at a low level, for example, +13.11 kg/ha in ryegrass +N and +7.57 kg/ha in barley +N, although these were fertilized with 100 kg N and 70 kg N respectively. The N removal through the biomass of these two crops averaged 49 kg N/ha. Thus, the question arises as to what happened to the remaining N applied to ryegrass +N (about 35 kg N/ha) and barley +N (10 kg N/ha). We assume that the additional fertilizer led to a lower soil N$_{\text{min}}$ content and attribute this to the so-called ‘negative priming effect’: the application of mineral fertilizer led to an N immobilization, making less N (temporarily) available to the crop (Kuzyakov, Friedel, & Stahr, 2000). As this ‘negative priming effect’ was found mainly in ryegrass and barley, the temporary lack of available N could be an explanation for their low yields. This assumption is strengthened by the fact that the N content of ryegrass +N was lower than ryegrass −N, but needs to be confirmed by further studies. If these results are confirmed, ryegrass and barley cannot be recommended to be grown after a miscanthus removal, due to their low yields.

Maize +N, which received the highest amount of fertilizer of all crops (240 kg N/ha), had a removal of 248 kg N/ha through its biomass. On the second sampling date, 24 kg N$_{\text{min}}$/ha were found in the soil, which means that the maize biomass had taken up almost the complete amount of fertilized N applied.

As stated above, a total of 39 kg N/ha were present on the field from litter and rhizomes. However, the average (of all crops and fertilization levels) increase in soil N$_{\text{min}}$ was only 14.85 kg N/ha, leading to the conclusion that some N from miscanthus residues was not plant-available and probably fixed in the organic matter, as suggested by Dufossé et al. (2014). It is possible that a large proportion of the residues may have started to decompose but had not completely decomposed in the first year after the miscanthus removal. After the winter wheat was sown, the N$_{\text{min}}$ contents stayed comparatively stable over winter in all plots (in soil depth 0–30 cm; results not shown). It then increased again in spring after the winter wheat was fertilized (results not shown). This indicates that even in the second year after a miscanthus removal, a flush of N is not to be expected. For final clarification, further research needs to analyse the decomposition rate of miscanthus residues after its removal under different soil and climatic conditions. In addition, cultivating crops that take up a high amount of N is recommended after a miscanthus removal to avoid losses through N release from decomposing miscanthus residues.
To summarize, our results show that soil N$_{\text{min}}$ content increases after a miscanthus removal, but on a low level. It seems that the N from plant residues is partly fixed in organic matter and is thus not plant-available. However, as some N is supplied by the decomposition of miscanthus residues, our findings suggest that the crop cultivated after a miscanthus removal requires less fertilization. In addition, the results revealed that the fertilized crops were more efficient at suppressing resprouting miscanthus and had a higher yield, indicating that fertilization should not be omitted after a miscanthus removal.

Our results point to a ‘negative priming effect’ in ryegrass +N and barley +N, which probably led to a (temporary) N immobilization. Thus, it is questionable whether these two crops are suitable for cultivation after a miscanthus removal. Maize, by contrast, tolerated the existing soil conditions quite well and seems to be a suitable crop for cultivation after miscanthus.

ACKNOWLEDGEMENTS

The research for this study was performed in the project MISCOMAR (funding code: 031B0163) supported by the German Federal Ministry of Education and Research within the framework of the ERA-NET cofund FACCE SURPLUS (no. 652615). The authors are grateful to the research station ‘Ihinger Hof’ for managing the field trial. Additionally, we thank the laboratory staff, especially Dagmar Mezger and Martin Zahner, for analysis of the biomass and soil samples. We also give thanks to Jens Hartung for his support in statistical analysis. Particular thanks go to Nicole Gaudet for proofreading the manuscript.

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**How to cite this article:** Mangold A, Lewandowski I, Kiesel A. How can miscanthus fields be reintegrated into a crop rotation? *GCB Bioenergy*. 2019;11:1348–1360. https://doi.org/10.1111/gcbb.12636