**Table S1.** Mean values (±1SE) of the total, bacteria, actinomycetes (ACT), fungi, gram-positive bacteria (GP), gram-negative bacteria (GN), arbuscular mycorrhizal fungi (AMF) PLFAs, and the ratio of fungi to bacteria (F:B ratio) across the two soil depths under the four cropping years. Y0: continuous maize-wheat rotation in recent decades; Y3: mugwort cropping for 3 years (since 2017); Y6: mugwort cropping for 6 years (since 2014); Y20: mugwort cropping for 20 years. Different letters indicate significant differences among the four cropping years at *P* < 0.05.

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Cropping years | Total | Bacteria | ACT | Fungi | GP | GN | AMF | F:B ratio |
| Y0 | 26.68±1.45ab | 15.29±0.79a | 3.45±0.13b | 0.55±0.06ab | 7.24±0.37a | 8.05±0.44a | 0.73±0.04a | 0.04±0.00a |
| Y3 | 31.02±1.88a | 17.69±1.19a | 4.12±0.22a | 0.83±0.09a | 7.94±0.47a | 9.76±0.75a | 0.82±0.07a | 0.05±0.00a |
| Y6 | 26.41±1.81ab | 15.23±1.06a | 3.70±0.27ab | 0.61±0.11ab | 7.27±0.42a | 7.96±0.75a | 0.66±0.05ab | 0.04±0.00a |
| Y20 | 21.18±1.99b | 11.77±1.03b | 2.07±0.18c | 0.52±0.10b | 5.89±0.47b | 5.87±0.62b | 0.54±0.07b | 0.04±0.01a |

**Figure S1.** The variations of microbial composition of gram-positive bacteria (GP), gram-negative bacteria (GN), fungi, actinomycetes (ACT), and general FAME under the four different cropping years of mugwort. Y0: continuous maize-wheat rotation; Y3: mugwort cropping for 3 years; Y6: mugwort cropping for 6 years; Y20: mugwort cropping for 20 years.

**Figure S2.** Relationships of PLFAs with root biomass. See abbreviations in Table S1.

**Figure S3.** Changes in PLFAs along the chronosequences. See abbreviations in Table S1.

