



1 **Nonlinear turnover rates of soil carbon following cultivation of native grasslands and**
2 **subsequent afforestation of croplands**

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28 Abstract

29 Land use conversions can strongly impact soil organic matter (SOM) storage, which
30 creates paramount opportunities for sequestering atmospheric carbon into the soil. It is known
31 that land uses such as annual cropping and afforestation can decrease and increase SOM,
32 respectively; however, the rates of these changes over time remain elusive. This study focused on
33 extracting the kinetics (k) of turnover rates that describe these long-term changes in soil C
34 storage and also quantifying the sources of soil C. We used topsoil organic carbon density and
35 $\delta^{13}\text{C}$ isotopic composition data from multiple chronosequences and paired sites in Russia and
36 United States. Reconstruction of soil C storage trajectory over 250 years following conversion
37 from native grassland to continual annual cropland revealed a C depletion rate of 0.010 years^{-1}
38 (first-order k rate constant), which translates into a mean residence time (MRT) of 100 years
39 ($R^2 \geq 0.90$). Conversely, soil C accretion was observed over 70 years following afforestation of
40 annual croplands at a much faster k rate of 0.055 years^{-1} . The corresponding MRT was only 18
41 years ($R^2 = 0.997$) after a lag phase of 5 years. Over these 23 years of afforestation, trees
42 contributed 14 Mg C Ha^{-1} to soil C accrual in the 0 to 15 cm depth increment. This tree-C
43 contribution reached 22 Mg C Ha^{-1} at 70 years after tree planting. Over these 70 years of
44 afforestation, the proportion of tree-C to whole soil C increased to reach a sizeable 79%.
45 Furthermore, assuming steady state of soil C in the adjacent croplands, we also estimated that
46 45% of the prairie-C existent at time of tree planting was still present in the afforested soils 70
47 years later. As intrinsic of k modelling, the derived turnover rates that represent soil C changes
48 over time are nonlinear. Soil C changes were much more dynamic during the first decades
49 following a land use conversion than afterwards when the new land use system approached
50 equilibrium. Collectively, results substantiated that C sequestration in afforested lands is a



51 suitable means to proactively mitigate escalating climate change within a typical person's
52 lifetime, as indicated by MRTs of few decades.

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56 **1 Introduction**

57 The global effects of escalating climate change are in part driven by land use choices.
58 Indeed, implementing certain land use changes can worsen or in other cases mitigate emissions
59 of greenhouse gases from terrestrial ecosystems to the atmosphere (Paustian et al., 1992; Post
60 and Kwon, 2000; Thilakarathna and Hernandez-Ramirez in press). Essentially, land use options
61 that unintentionally accelerate biological oxidation of soil organic matter (SOM) contribute over
62 time to atmospheric carbon dioxide concentrations (Sauer et al., 2007; Laganieri et al., 2010; Li
63 et al., 2018), providing a portion of the radiative forcing that has been causing part of the global
64 warming effect over the last decades (Guo and Gifford, 2002; Parry et al. 2007). Compared with
65 contrasting types of land use systems, annual croplands commonly showed SOM depletion and
66 marked reductions in soil C storage, in particular relative to their natural ecosystem counterparts
67 (Chendev et al., 2015b; Hebb et al., 2017; Kiani et al., 2017).

68 Contrary to the potentially detrimental effects of annual cropping on SOM accumulation
69 and overall soil quality (Guenette and Hernandez-Ramirez, 2018; Laganieri et al., 2010; Kiani et
70 al., in press), tree planting offer multiple environmental services and societal benefits
71 (Hernandez-Ramirez et al., 2012; Sauer et al., 2012; Zhang et al., 2020). For instance, removing
72 C from the atmosphere is a paramount contribution by trees (Guo and Gifford, 2002; Li et al.,
73 2012; Li et al., 2018). In effect, soil C accrual (Paul et al., 2002; Dhillon and Van Rees, 2017;
74 Khaleel et al., 2020) and stabilization (Hernandez-Ramirez et al., 2011; Wang et al., 2016;
75 Quesada et al., 2020) beneath trees have been recognized as an effective means for sequestering
76 atmospheric C. In addition to soil accruals, soil microbiology is sustained and diversified beneath
77 mature trees (Kiani et al., 2017). Additional functions by tree vegetation include improving air
78 quality, enhanced microclimate, and erosion control (Sauer et al., 2007; Hernandez-Ramirez et



79 al., 2012; Chendev et al., 2015b). Establishing agroforestry practices such as shelterbelt systems
80 within annual croplands can provide a balance between continual food production and tree
81 benefits with only a fraction of the landscape occupied by trees (Amadi et al., 2016; Dhillon and
82 Van Rees, 2017).

83 Sequestering C in soils is governed by the balance of inputs of plant C with biological-
84 mediated decomposition and stabilization processes (Hernandez-Ramirez et al., 2009; Kiani et
85 al., 2017; Li et al., 2018). This overall functioning of C-related biology and cycling in soils can
86 be described as the turnover of soil C. Collectively, SOM decomposition, mineralization, gains
87 and losses and net accrual can be numerically integrated into C turnover rates (Richter et al.,
88 1999; Hernandez-Ramirez et al., 2011; Xiong et al., 2020). For instance, dynamic rates of net
89 depletion of SOM pools caused by continual cropping or tree contributions to soil C accretion
90 and cycling under afforestation can both be captured as C turnover rates (Guo and Gifford, 2002;
91 Hu et al. 2013). However, the outcomes of how slow or fast the accrual of SOM takes place as a
92 function of land use conversions need to be assessed in the long term, as this will inform how
93 lasting these effects are (Paustian et al., 1992; Hernandez-Ramirez et al., 2011). Indeed,
94 understanding long-term changes in SOM storage, instead of the results from a single point in
95 time, is highly valued knowledge (Paustian et al., 1992; Richter et al., 1999; Guo and Gifford,
96 2002). Extracting and making available in the literature the long-term turnover rates of SOM
97 depletion or accretion can enable useful projections of future soil C sequestration as a function of
98 implementing land use changes. Our study endeavors to address and fill this knowledge gap.

99 In this study, we compiled soil C storage data from several field sites comparing land use
100 systems in Russia and United States (Chendev et al., 2015a, 2015b) in conjunction with
101 published (Hernandez-Ramirez et al., 2011) and newly-available soil ^{13}C isotope data. Based on



these data assemblage, we now focused on evaluating the long-term turnover rates of soil C as a function of land use changes from native grasslands to annual croplands and subsequent converting annual croplands into afforestation. We aimed at extracting turnover rates of soil C depletion or accretion, which can enable future predictions of soil C storage depending on land use systems. Also, our study quantified and documented the contributions of tree biomass-C to soil C that was newly-accrued following afforestation. We further examined the stage and net losses of the C that existent in the soil under annual croplands prior to tree planting.

109

110 **2 Materials and Methods**

111 **2.1 Chronosequences from native grasslands to annual croplands in Russia**

Three long-term chronosequences of land use conversion (i.e., a range of different duration of cultivation) were used for extracting turnover parameters. These land use chronosequences were situated in Belgorod oblast, Russia within the districts of Prokhorovskiy (50°57' N, 36°44' E), Gubkinskiy (51°03' N, 37°22' E) and Ivnyanskiy (51°06' N, 36°24' E) as previously described by Chendev et al. (2015a) (Fig. 1). Following chronosequence methods as described by Laganier et al. (2010), each chronosequence had four or five age-sites encompassing a native grassland site that represented the time zero of conversion from grassland to annual cropland. These native grasslands were undisturbed steppe dominated by plant species with C3 photosynthetic pathway. The ages of the cropland sites were established through historical records and geographic approaches described by Chendev et al. (2012, 2015b). Composite soil samples (3 subsamples per sample) with at least 12 sampling locations per age-site were collected using the core method in 10 cm depth increments. Additional information about these sites is available at Chendev et al. (2015a), while the focus in our study remains on



125 developing models and extracting parameters of C turnover rates. Typical annual crops species
 126 included cereals, sunflower (*Helianthus annuus*) and beet (*Beta vulgaris*) managed under
 127 conventional tillage operations. Within the study region, soils were classified as loamy
 128 Chernozems (Russian Soil Classification System), annual precipitation ranged between 480 and
 129 580 mm and air temperature between 5.3 and 5.8 °C (Chendev et al., 2015a).

130 Soil organic C mass density for the 0 to 30 cm depth was calculated as the sum of
 131 products of organic C concentration (Hernandez-Ramirez et al., 2009), bulk density and soil
 132 layer thickness, with units of Mg C ha⁻¹.

133 First-order kinetic modelling follows:

$$134 \quad C_{(t)} = C_e + (C_o - C_e) e^{-kt} \quad [1]$$

135 where C_e is soil C storage at the oldest cropland site within each chronosequence which was
 136 assumed to be at new dynamic equilibrium (i.e., C inputs = C outputs), C_o is soil C storage at the
 137 native grassland site which was assumed to be the initial time of land use conversion from native
 138 grassland to annual croplands (time zero), k is the fitted first-order kinetic rate constant (yr⁻¹),
 139 which is equivalent to C turnover rate or net C mineralization (in the case of net C decreases),
 140 and t stands for time (yr). In the case of increases in soil C over time, turnover rates become
 141 equivalent to accretion rates. It is possible to model the soil C storage for each year, and hence,
 142 the difference between consecutive years provides an estimation of the annual net C change (Mg
 143 C ha⁻¹ yr⁻¹). In addition to balance between C inputs and C outputs, first-order kinetic modelling
 144 also assumed steady state conditions (i.e., $\delta C / \delta t = 0$). At the selected sampling sites, given the
 145 flat topography, we also assumed negligible C removals or additions due to erosion or
 146 deposition.



147 Mean residence time (MRT) of organic C in the soil was calculated as reciprocal of k.

148 Concomitantly, half-life of organic C in the soil was calculated as follows:

149 $\text{Half-life} = \ln(2)/k$ [2]

150 Note that under equilibrium, C output_e is also equivalent to C input_e, and they correspond to the
 151 annual C that enters and exits the soil C pool, respectively.

152 The performance of the derived first-order kinetic modelling was evaluated with the
 153 normalized root mean square error (RMSE_n) (Guenette and Hernandez-Ramirez, 2018; Kiani et
 154 al., in press), coefficient of determination (R^2) as well as a leave-one-out cross-validation of
 155 predicted versus measured C. Within the cross-validation, we tested the regression coefficient
 156 (β_1) of a linear regression established for predicted vs. measured C against the 1:1 line.

157 **2.2 Comparison of adjacent paired sites in Russia: native grasslands, annual croplands and** 158 **shelterbelts (trees)**

159 In addition to the three abovementioned chronosequences, three additional sites were
 160 studies in Russia: Streletskaya Steppe situated within Kursk oblast (51°32' N, 36°05' E),
 161 Yamskaya Steppe in Belgorod oblast (51°11' N, 37°37' E), and Kamennaya Steppe in Voronezh
 162 oblast (51°02' N, 40°44' E) (Fig. 1). Soils at all these paired sites were classified as loamy
 163 Chernozems. Following field methods as described by Laganier et al. (2010), each site
 164 encompassed adjacent locations representing three land uses: native grassland, annual croplands
 165 and broadleaf shelterbelts, as described by Chendev et al. (2015b). Soil sample collections were
 166 conducted similar as described above. Briefly, composite soil samples (3 subsamples per sample)
 167 were collected from the native grassland (n= 6 composite soil samples), annual croplands (24)
 168 and shelterbelts (18) in summer 2012. At time of soil sample collection, the ages of the annual
 169 croplands in Streletskaya and Yamskaya were at least 140 years, and at least 145 years of age in



170 Kamennaya. In all three sites, the shelterbelts had been planted 55 years prior to soil sample
 171 collection. Tree species in the shelterbelts include silver birch (*Betula verrucosa*) and black
 172 poplar (*Populus nigra*) at the Streletskaya site, American maple (*Acer negundo*) at the Yamskaya
 173 site, and balsam poplar (*Populus balsamifera*) and English oak (*Quercus robur*) at the
 174 Kamennaya site. Normal mean annual precipitations at Streletskaya, Yamskaya and Kamennaya
 175 correspond to 580, 530 and 480 mm yr⁻¹, respectively.

176 It is noted that although trees at the Streletskaya, Yamskaya and Kamennaya sites were
 177 planted 55 years prior to soil sample collection, for first-order kinetics modelling purposes, the
 178 tree-C contributions to soil C accrual were accounted beginning from 50 years prior to soil
 179 sample collection. This assumption is based on a literature review by Paul et al. (2002) who
 180 suggested a lag phase of 5 years for tree-C contributions to effective start contributing to net
 181 storage of soil C. Moreover, because of the uncertainty of how close the afforested soils were to
 182 steady state and equilibrium of soil C storage, we evaluated two scenarios of first-order kinetics
 183 modelling using Eq. [1]. We assessed trajectory ‘A’ under the premise that full steady state has
 184 been reached at time of soil sample collection, and also trajectory ‘B’ where we assumed that the
 185 C storage in these afforested soils had asymptotically reached 95% of the theoretical equilibrium
 186 or ceiling capacity. We reported both trajectories and their associated C accretion rates (k).

187 **2.3 Pairwise comparisons in United States: annual croplands and afforestation**

188 Three field sites were studied within the Northern Great Plains of United States near the
 189 cities of Huron (South Dakota, 44°15′ N, 98° 15′ W), Norfolk (Nebraska, 42° 03′ N, 97° 22′ W)
 190 and Mead (Nebraska, 41°9′ N, 96°29′ W) (Fig. 1). While the focus remained on investigating the
 191 turnover rates of soil C as a function of land use changes, it is noted that pedogenic ages of the
 192 soils sampled in United States were relatively shorter than the soils studied in Russian sites. This



193 is because of the differences in geological times of parent materials exposure on the ground
194 surface between geographic regions since the sites in United States experienced the last
195 glaciation (i.e., Wisconsin glaciation).

196 The three sites in United States encompassed afforested areas and adjacent annual
197 croplands, co-located in paired sites as Laganriere et al. (2010). The native vegetation at the sites
198 had been tall prairie (e.g., big bluestem *Andropogon gerardii* Vitman), which had been converted
199 into annual croplands, and trees were subsequently planted in areas of the croplands.

200 Afforestation took the forms of shelterbelts in Norfolk and Mead, and a forest plantation in
201 Huron. The Huron site also had an adjacent field with a representative undisturbed native prairie,
202 which was also sampled as a reference. In contrast to the long-term croplands in Norfolk and
203 Mead, the cropland at Huron had only 21 years since conversion from native grassland at time of
204 soil sample collection. Field sample collections were conducted in 2004 in Mead, and in 2012 in
205 both Norfolk and Huron. The trees had been planted 19, 35 and 70 years prior to soil sample
206 collections in Huron, Mead and Norfolk, respectively. Tree species included green ash (*Fraxinus*
207 *pennsylvanica* Marshall), red cedar (*Juniperus virginiana* L.), and oak (*Quercus macrocarpa*) in
208 Huron; red cedar, scotch pine (*Pinus sylvestris* L.), and cottonwood (*Populus deltoides* Bartram)
209 in Mead; Siberian elm (*Ulmus pumila*), red mulberry (*Morus rubra*), and cottonwood in Norfolk.
210 Annual croplands were managed under conventional farming practices. Annual crop species at
211 the study sites included wheat (*Triticum aestivum* L.), corn (*Zea mays* L.), soybean [*Glycine max*
212 (L.) Merr.], and sorghum (*Sorghum bicolor* L. Moench). An alfalfa (*Medicago sativa* L.) forage
213 field adjacent to the shelterbelt in Norfolk was also sampled. Normal annual precipitation in
214 Huron, Norfolk and Mead were 582, 696 and 747 mm yr⁻¹, respectively. Normal air temperature
215 in Huron, Norfolk and Mead were 7.7, 9.6, 9.9 °C, respectively. Soil textures were loamy sand in



216 Norfolk, sandy loam in Huron and silty clay loam in Mead, respectively. Soil pH were 6.1 in
217 Mead, 6.8 in Norfolk, and 7.0 in Huron.

218 Field methods of soil sample collections had previously been described in related reports
219 by Chendev et al. (2015a) for the Huron and Norfolk sites as well as by Sauer et al. (2007) for
220 Mead. Briefly, spatial grid patterns were established with composite samples (n= 4) collected
221 from each sampling location. Total grid sampling locations were 118 at Mead, 48 at Huron, and
222 42 at Norfolk. Plant tissue samples of the dominant species were also collected from each study
223 site.

224 Organic C concentration and $\delta^{13}\text{C}$ isotopic composition were determined in all soil and
225 plant samples via the dry combustion method using a Fison NA 15000 Elemental Analyzer
226 (ThermoQuest Corp., Austin, TX) interfaced to an isotope-ratio mass spectrometer Delta V
227 Advantage (Thermo Fisher Scientific, Waltham, MA). Pee Dee Belemnite was used as standard
228 and analytical precision of $\delta^{13}\text{C}$ measurements was 0.06‰. The $\delta^{13}\text{C}$ isotopic ratio was
229 expressed as:

230
$$\delta^{13}\text{C} (\text{‰}) = [({}^{13}\text{C}/{}^{12}\text{C} \text{ sample}) / ({}^{13}\text{C}/{}^{12}\text{C} \text{ standard}) - 1] \times 1000 \quad [3]$$

231 When integrating multiple soil layers of a profile, averages of $\delta^{13}\text{C}$ were weighted by the
232 soil C mass density at the corresponding soil layers.

233 Prior to land use conversion to croplands, the native grasslands in United States were
234 undisturbed and dominated by plant species with C4 photosynthetic pathway, with certain mixed
235 presence of C3 species. Based on this legacy contribution of prairie vegetation to soil C over the
236 Holocene, and assuming that net contributions to soil C storage were negligible during annual
237 cropping stages, approaches based on stable isotope signatures became feasible in these three
238 sites in United States. This method enabled us to examine the sources of soil C and also derive



the turnover rates of these soil C sources. This primarily applies because trees are C3 species. This approach assumed that the differences in ^{13}C isotopic signatures between C4-C3 mixed (native grassland) and C3 (trees) overrides any potential differential effect of C isotopic fractionations during SOM decomposition of C3 vs. C4 substrates, between aboveground and below ground plant materials (roots vs. litter), or because of SOM interactions with soil mineral surfaces (Martin et al., 1990; Hernandez-Ramirez et al., 2011). Assuming mass conservation, the measured soil C storages were allocated into two sources: new tree-C and remaining prairie-C (native soil) as follows:

$$\text{Tree-C} + \text{Prairie-C} = 1 \quad [4]$$

$$\text{Tree-C} = (\delta^{13}\text{C}_{\text{afforested soil}} - \delta^{13}\text{C}_{\text{native soil}}) / (\delta^{13}\text{C}_{\text{tree}} - \delta^{13}\text{C}_{\text{native soil}}) \quad [5]$$

It was inferred that all soil C different from the C identified as new ‘Tree-C’ was preexisting soil C attributable to remaining ‘Prairie-C’. Likewise, we assumed that the ‘ $\delta^{13}\text{C}$ native soil’ were represented reasonably well by the $\delta^{13}\text{C}$ measured in soil samples taken from the annually-cropped fields adjacent to the afforested soils. Although they were adjacent, the sampling locations providing the ‘ $\delta^{13}\text{C}$ native soil’ were sufficiently distant from afforested areas to preclude influence of trees on soil $\delta^{13}\text{C}$. The ‘ $\delta^{13}\text{C}$ native soil’ were -17.3‰ in Huron, -17.0‰ in Mead, and -17.5‰ in Norfolk, which are noted to be consistent with each other as these sites share a common natural history of tall prairie native vegetation. These ‘ $\delta^{13}\text{C}$ native soil’ are also consistent with earlier measurements in prairie soils by Follett et al. (1997) and Hernandez-Ramirez et al. (2011). Furthermore, the $\delta^{13}\text{C}$ measured in tree samples averaged -27.6‰ in Huron, -26.6‰ in Mead, and -27.9‰ in Norfolk as typical isotopic compositions of C3 plant species. At the Norfolk site, tissue samples of alfalfa canopy yielded -27.4‰. Also, in Norfolk, ‘ $\delta^{13}\text{C}$ afforested soil’ included all data from the soil samples taken within 10 m north and 10 m



262 south from the center of the shelterbelt. In Huron, ‘ $\delta^{13}\text{C}$ afforested soil’ sample included all data
 263 from soil samples taken at least 17 m away from the edge between the forest plantation and the
 264 adjacent annually-cropped field. In Mead, as earlier presented by Hernandez-Ramirez et al.
 265 (2011), ‘ $\delta^{13}\text{C}$ afforested soil’ sample included all data from soil samples collected between the
 266 existing two tree rows.

267 The mass densities of soil C derived from new tree-C and remaining prairie-C were
 268 calculated by multiplying whole soil C storage beneath the trees with the corresponding fractions
 269 expressed in Eq. [4].

270 When assessing first-order kinetics modelling (Eq. [1]) of soil C beneath trees, we
 271 assumed that the remaining prairie-C in afforested soils at time of soil sample collection was at
 272 steady state and had also reached new equilibria in the case of the long-term annual croplands at
 273 Norfolk and Mead. In the specific case of Huron, because the afforested soil had experienced
 274 only two years of annual cropping prior to tree planting, we assumed that the native grassland
 275 prior to land use conversion to annual cropping was at equilibrium and steady state.

276 Relationships between allocations of soil C sources (% tree-C, and % prairie-C) and time
 277 since tree planting (years) were examined through linear regression analyses. Likewise, a similar
 278 linear regression was developed for the proportions of remaining prairie-C in the afforested soils
 279 relative to whole C present in the adjacent cropland soils. We used SigmaStat Version 4.0
 280 software (Systat Software, San Jose, CA) and an α critical level of 0.05. Where error terms (\pm)
 281 are presented, they correspond to the standard errors of the means.

282

283 **3 Results**

284 **3.1 Soil C after conversion of grasslands to cropland and then shelterbelt: Russian cases**



285 Long-term cultivation of native grasslands decreased soil organic C storage in a nonlinear
 286 fashion (Fig. 2). Within each of the three available land use chronosequences (i.e., each
 287 encompassing a range of different duration of cropping since land use conversion), the declining
 288 trajectory of soil C was represented reasonably well by first-order kinetic modelling. (A). The
 289 RMSEn were all lower than 4% and the R^2 were greater than 90%, which supports the suitable
 290 performance of k models (Fig. 2A, Fig. 2B, and Fig. 2C). Likewise, cross-validation results of
 291 cropland soils within the age range from 10 to 200 years further indicated the high accuracy of k
 292 predictions when compared with the 1:1 agreement line. This was based on a non-significant t-
 293 test with $\beta_1=1$ as null hypothesis (Fig. 2D).

294 Within the soil layer of 0 to 30 cm depth in the chronosequences in Belgorod, Russia,
 295 turnover rates (k) of soil C ranged from 0.0091 to 0.0183 yr^{-1} in Gubkinskiy and Prokhorovskiy,
 296 respectively. Over the entire time spans of the three chronosequences, net soil C losses were in
 297 the relatively narrow range from 31.2 Mg C ha^{-1} in Prokhorovskiy (Fig. 2A) to 36.9 Mg C ha^{-1} in
 298 Gubkinskiy (Fig. 2B). Focusing on these losses of the preexisting soil C, the estimated lapses for
 299 half of these C losses to take place was between 38 and 76 years after the time of land use
 300 conversion from native grassland to annual cropland (Fig. 2A and Fig. 2B, respectively). It is
 301 noted that new dynamic equilibria were assumed to have taken place in the oldest cropland soil
 302 within each chronosequence (i.e., > 200 yrs) as part of first-order kinetics modelling.
 303 Furthermore, when examining the assumption of steady state (i.e., $\delta C/\delta t = 0$), soil C trajectories
 304 at both Prokhorovskiy and Ivnyanskiy sites showed reasonable approximations to this premise,
 305 with relatively low annual C losses occurring towards the end of these chronosequences. On the
 306 other hand, the Gubkinskiy site still exhibited vigorous C losses at the end of this
 307 chronosequence, which can question steady state assumption at this site. In further details, the



trajectory at the Prokhorovskiy site showed estimated C losses of only 9.4 Kg C ha⁻¹ yr⁻¹ during the last year of this chronosequence (Fig. 2A), which can be considered negligible and in clear agreement with a steady state condition. Conversely, the last year of the Gubkinskiy chronosequence still showed a C loss of 34.5 Kg C ha⁻¹ yr⁻¹. It is noteworthy that while these annual C outputs typically take the form of soil respiratory losses (CO₂-C) that are resultant from microbial mineralization of existing SOM. These estimations of net changes at near steady state do not account for the CO₂ derived from the decomposition of recently-added plant residues, but just the net change in SOM-C (Fig. 2A). For comparison purposes, the k-modelled trajectory of the Prokhorovskiy chronosequence had estimated C losses of 566 Kg C ha⁻¹ yr⁻¹ during the very first year after conversion from native grassland to annual cropland.

Afforestation implemented in the form of shelterbelts replenished soil C storage based on assessed pairwise comparisons (Table 1). Cropping led to decreasing soil C relative to adjacent native grasslands (Table 1), which was consistent with the results from chronosequences described above (Fig. 2). Nevertheless, afforestation following cropping increased soil C accrual. In further details, of the substantial soil C storage that had been depleted over time during annual cropping (i.e., -18.9 ± 5.3 Mg C ha⁻¹), afforestation replenished on average 81% of these cropping-induced C losses (Table 1). This replenishment of soil C by tree contributions was observed across the three sites, but with certain variability. The Yamskaya site showed a pronounced tree-induced replenishment of 138% of the lost soil C, which was even higher than the native grassland in terms of soil C storage. On the other hand, at the Kamennaya site, afforestation has only restored 5% of the soil C that had been lost during long-term cropping.

Using the insights gained from both chronosequences (Fig. 2) and pair site comparisons (Table 1), we undertook the reconstruction of soil C storage progression in the 0 to 30 cm soil



layer since the land use conversion from native grassland to annual cropland and subsequently into shelterbelt (Fig. 3). After normalizing all cropland-chronosequence data, turnover rates (k) and first-order kinetic models of soil C storage were estimated (Fig. 3). This long-term k model of soil C depletion in cropped soil had a reassuring coefficient of determination (R^2) of 90% and a very low RMSEn of only 3.34%, which collectively indicates the high precision of the k model. Over 250 years of cropland chronosequence, the C turnover rate was quantified as $0.010 \pm 0.004 \text{ years}^{-1}$, which is equivalent to a MRT of 100 years and a half-life of soil C of 69 years. This first-order trajectory of soil C depletion in croplands indicated that 28.9% of the initial soil C under native grassland was gradually lost – i.e., very likely to the atmosphere – over 250 years of annual cropping (i.e., from 1 to 0.711, Fig. 3). In further details, during the first year of cropping, we estimated that 0.310% of the pre-existing C was lost from the soil. Conversely, after 250 years, during the last year of the cropland-chronosequence trajectory, soil C losses were only 0.026% of the initial soil C – this is one order of magnitude lower than calculated for the first year of cropping. This deceleration in SOM mineralization while approaching a new equilibrium and at near steady state was captured reasonably well by first-order kinetics. Based on the soil C initially present under native grassland soils (3-chronosequence mean = $125.3 \text{ Mg C ha}^{-1}$, Fig. 2), these values of 0.31% and 0.026% were equivalent to C outputs of 392 and 33 $\text{Kg C ha}^{-1} \text{ yr}^{-1}$, respectively.

We projected two potential trajectories (A and B) of how afforested soils can restore soil C storage in cropland soils over five decades (Fig. 3). Upon accounting for the replenishment of 81% of the soil C storage caused by shelterbelts (Table 1), we estimated that the afforested soils reached a soil C storage of 94.5% (i.e., $0.81 \times 0.289 + 0.711 = 0.945$) relative to the soil C present in the initial native grassland soils. Trajectory A was estimated on the basis that



354 afforested soils fully reached a new steady state and equilibrium of soil C storage with first-order
 355 modelling. This trajectory showed a steep increase in soil C storage over the first decade of tree
 356 planting. In fact, the C accretion rate for trajectory A was 0.119 years^{-1} , which suggests a
 357 potential for high soil C accretion under fast C cycling with afforestation. Because the soils
 358 beneath the shelterbelt can still be actively accruing C even 55 years after tree planting, we also
 359 developed trajectory B, which targets a scenario where soil C storage reached 95% of a
 360 theoretical equilibrium (Fig. 3). For this trajectory, the resultant C accretion rate was 0.0334
 361 years^{-1} , which corresponds to a modelled MRT of 30 years. When focusing on these progressive
 362 gains of new soil C under trajectory B, the estimated time for half of this soil C portion to enter
 363 the soil was 21 years. Over the two last decades of this progression, the soil C accretion starts to
 364 gradually become asymptotic. During the first year of trajectory B, the net accrual of soil C was
 365 equivalent to 0.88% of the soil C initially present in the native grasslands (Fig. 3). Conversely,
 366 during the last year of trajectory B, soil C accretion corresponded to only 0.17%. Based on the
 367 soil C initially present under native grassland soils (3-chronosequence mean= $125.3 \text{ Mg C ha}^{-1}$,
 368 Fig. 2), these 0.88% and 0.017% values were equivalent to a sizable 1.10 and $0.21 \text{ Mg C ha}^{-1} \text{ yr}^{-1}$,
 369 respectively. It is noted that in each of the two afforestation trajectories of soil C accretion (A
 370 and B), the annual contributions of afforestation to net accrual of soil C began from the soil C
 371 storage estimated by the k-modelled cropland trajectory 50 years prior to soil sample collection
 372 (i.e., 0.728, Fig. 3). This is because we had assumed a lag phase of 5 years as noted above, and
 373 tree planting was 55 years prior to soil sample collection.

374 Based on the assembled k models of cropland-C turnover and simultaneous tree-C
 375 accretion (Fig. 3), of the whole soil C measured beneath the trees at the time of soil sample
 376 collection (Table 1), 25% was estimated to be derived directly from tree-C contributions [i.e.,



(0.945 – 0.711) / 0.945]. This can indicate that although tree-C contributions were substantial, the majority of the C stored in these steppe soils still originated from the initial native grassland before land use conversion to annual croplands.

3.2 Sources and turnover of soil C in afforested croplands: United States cases

Larger accumulation of soil C was consistently found beneath trees relative to the adjacent annual croplands in all three study sites within the Northern Great Plains of the United States. At the shelterbelts in Norfolk (Fig. 4E) and Mead (data not shown) as well as the forest plantation in Huron (Fig. 4F), tree-derived soil C were clearly discernable, in particular within the 0 to 15 cm soil depth increment (Table 2).

At the Norfolk site, soils collected from the 0 to 15 cm depth increment beneath the trees (i.e., within 10 m distance from the center of the shelterbelt) resulted in more than double of the C mass density found in the annually-cropped topsoils that were located farthest from the trees (28 vs. 13 Mg C ha⁻¹; Table 2, Fig. 4A). Concurrently, when comparing the same surface layer and spatial sampling locations, soil δ¹³C sharply shifted from a considerably depleted -25.7 ± 0.1 ‰ beneath the trees to -17.5 ± 0.1 ‰ in the cropped soils north from the shelterbelt (Fig. 4C). As a result, a significant 79% of the soil C storage measured beneath the trees at the time of sample collection was attributed to tree-C contributions (Table 2). This translated into a substantial magnitude of 22 Mg C ha⁻¹ being derived specifically from tree biomass (Table 2). Moreover, as stated above (Method section 2.3), the rest of the soil C was attributed to remaining prairie-C. The existing soil C beneath trees in the Norfolk shelterbelt allotted to remaining prairie-C was only 45.5 ± 0.3 % of the whole soil C typically found in the adjacent annual crop field (Table 2). This indicated that 54.5% of the soil C (equivalent to 13.0 - 5.9 = 7.1 ± 0.4 Mg C ha⁻¹, Table 2) that existed under the long-term annual cropland (i.e., assumed to be at steady state) prior to tree



400 planting has been lost from the topsoil over the 70 years of afforestation at Norfolk. This net
 401 decline in remaining prairie-C is attributable to CO₂ respiratory losses from enhanced biological
 402 activity beneath the trees that gradually accessed, mobilized, cycled and partly mineralized this
 403 legacy prairie-C pool. These results indicated that the turnover of remaining prairie-C in
 404 afforested soils can be even faster than in open cropland fields.

405 Because the crop field south from the shelterbelt in Norfolk was dedicated to perennial
 406 cropping of alfalfa forage – a C3 species, we undertook a mass balance to distinguish and
 407 allocate the C sources as alfalfa-C vs. remaining prairie-C, with a similar approach as in the
 408 afforested areas (i.e., Eq. [4] and [5]). Within the 0 to 15 cm depth increment, soil C storage
 409 under alfalfa (i.e., $13.1 \pm 1.0 \text{ Mg C ha}^{-1}$) was the same as in the annual cropland on the north side
 410 of the shelterbelt (Fig. 4A; Table 2). However, the soil $\delta^{13}\text{C}$ shifted to $-21.6 \pm 0.4 \text{ ‰}$, which
 411 resulted in a $41.2 \pm 0.4\%$ replacement of the whole soil C storage being derived specifically from
 412 recent contributions of alfalfa-C in this perennial forage field.

413 Similar to Norfolk, the shelterbelt at Mead also showed a major contribution of
 414 afforestation to whole soil C storage between tree rows in the 0 to 15 cm depth increment (i.e.,
 415 17 Mg C ha^{-1} , Table 2), which corresponded to 37% of the whole soil C. It is noted that although
 416 the magnitude of original prairie-C lost after 35-yr of afforestation at Mead (i.e., $36.2 - 29.3 =$
 417 $6.9 \pm 0.7 \text{ Mg C ha}^{-1}$, Table 2) was comparable to Norfolk, the proportion of this original prairie-
 418 C lost from afforested soils in Mead was much smaller with only 19% (i.e., $100 - 80.9$; Table 2).
 419 This can indicate that net changes in soil C storage under afforestation simultaneously
 420 encompassing the noted prairie-C losses and the asymmetrically-larger tree-C gains followed a
 421 stoichiometry of absolute capacities, instead of a proportionality of the initial prairie-C.



At the 19-year-old forest plantation at Huron, trees also increased soil C storage while decreasing the $\delta^{13}\text{C}$ signature, in particular in the 0 to 15 cm depth increment (Fig. 4B, Fig. 4D). When contrasting the afforested soils collected at least 17 m away from the plantation boundary vs. topsoils taken within the adjacent cropland from the sampling locations that were farthest removed from the trees, $\delta^{13}\text{C}$ changed from $-20.7 \pm 0.2 \text{ ‰}$ to $-17.3 \pm 0.1 \text{ ‰}$, respectively. Although trees were much younger in Huron than in Mead and Norfolk, a considerable magnitude of tree-C was found (i.e., 12 Mg C ha^{-1} , Table 2). This indicated that direct tree contributions to soil C storage can take place rather quickly, within a few decades following afforestation. However, the changes in the remaining prairie-C pool beneath the trees at Huron apparently differed from what was found in both Mead and Norfolk. While soils beneath trees at both Mead and Norfolk showed declines in remaining prairie-C relative to whole soil C stored in open cropland fields at the time of sample collection, the afforested soil at Huron showed no change in the magnitude of remaining prairie-C in the 0 to 15 cm depth increment. In fact, it is striking how similar the whole C in the cropped soil ($25.6 \pm 0.7 \text{ Mg C ha}^{-1}$) was to the C allocated to the remaining prairie-C pool beneath the trees ($25.3 \pm 0.7 \text{ Mg C ha}^{-1}$, Table 2). What is more, the soils sampled from the adjacent native grassland within the Huron site also returned a very consistent magnitude of soil C storage, with $25.4 \pm 1.9 \text{ Mg C ha}^{-1}$ ($n=3$, data not shown). Provided the uncertainty of field sampling, this evidence strongly indicated that all or nearly all the original prairie-C was retained and still present in the Huron soils under both annual cropping and afforestation (Table 2). Likewise, when examining the 0 to 30 cm soil depth increment at Huron (i.e., aggregating the two sampled soil layers shown in Fig. 4B), we further corroborated this similarity in soil C storage between native grassland and annual cropland (45.4 ± 1.0 vs. $44.2 \pm 1.1 \text{ Mg C ha}^{-1}$, respectively, data not shown)



445 Significant regressions revealed the consistent dependency of C source allocations on
 446 time since tree planting (Fig. 5). Over time following afforestation, tree-C source increased
 447 linearly from an assumed null contribution at planting to become 79% of the whole soil C after
 448 70 years of tree planting in Norfolk ($R^2 = 0.95$, Fig. 5A). We also evaluated changes over time for
 449 the remaining prairie-C in afforested soils relative to the corresponding adjacent croplands within
 450 each study site. Linearity of these prairie-C proportions as a function of time was also observed
 451 when encompassing the three study sites ($R^2 = 0.999$, Fig. 5B). As described above, the more
 452 recently-afforested soils at the Huron site kept the entire prairie-C, while the oldest afforested
 453 soils at the Norfolk site retained less than half of the whole soil C present in the adjacent annual
 454 croplands (45.5%).

455 We focused on estimating the turnover rates of soil C mass density derived directly from
 456 tree-C sources while encompassing the range of conditions in the three study studies. As most of
 457 the beneficial effects of tree planting across the sites in the United States were detected in the 0
 458 to 15 cm soil depth increment, further examination of accretion rates of soil C storage focused on
 459 this specific topsoil layer. Upon assembling the magnitudes of tree-C contributions over time
 460 since afforestation, unified first-order kinetics modelling converged and emerged robustly ($R^2 =$
 461 0.997 , Fig. 6). The k rate constant of $0.0552 \text{ years}^{-1}$ corresponds to a half-life of 12.6 years,
 462 which indicates that more than half of the accrual tree-C occurred within less than two decades
 463 (when accounting for a lag phase of 5 years following tree planting) (Fig. 6). This further
 464 substantiated the rapid contributions of afforestation to increase soil C storage quickly until
 465 reaching a new dynamic equilibrium. This generalized relationship enabled projecting tree-C
 466 accruals in afforested soils within the assessed time range of 70 years. We further implemented
 467 this robust k -progression to simultaneously depict the gains in tree-C while also representing the



declines in prairie-C in afforested soils for each study site separately (Fig. 7). This approach accounts for the C that is being lost from net mineralization of pre-existing C in the remaining prairie SOM (Fig. 7). It was noticeable that the afforested soils at Mead showed faster turnover rate of the remaining prairie-C than the other two sites by approximately two-fold. The k rate constant of net mineralization of prairie-C beneath trees at Mead was 0.145 years^{-1} (Fig. 7B), which corresponded to an MRT of about 7 years. This implies that the average time for prairie-C to be lost from Mead afforested soils was well within one decade, whereas prairie-C in afforested soils in Huron and Norfolk showed longer residence times by about double.

476

4 Discussion

4.1 Carbon contributions from trees to SOM sequestration

Planting trees in croplands creates substantial sinks of atmospheric C in the soil profile (Sauer et al., 2007; Khaleel et al., 2020; Zhang et al., 2020). Current knowledge of this important benefit of afforestation has been deepened and reinforced earlier literature (Post and Kwon, 2000; Hernandez-Ramirez et al., 2011; Chendev et al., 2015b). It is noticeable that having long-term annual croplands as the land use system prior to establishing trees particularly enlarges the soil C sink and replenishment caused by afforestation (Guo and Gifford, 2002; Laganier et al., 2010; Sauer et al., 2012). Overall results indicate that across tree species and local edaphic-climatic conditions at the studied sites, the massive tree-C contributions through decaying roots and litter (Li et al., 2012; Amadi et al., 2016) can saturate the soil with C substrates in surplus to the capacity of microbial decomposition (Li et al., 2018; Deng et al., 2014), which collectively incline the C balance towards net C accrual (Hernandez-Ramirez et al., 2011). Our quantification of these tree biomass-C contributions to newly-accrued soil C further expand this growing body



491 of knowledge. The proportion of new soil C originated from tree biomass were shown to increase
492 significantly with time (Fig. 5A). As abovementioned, the shift from C4-dominated to C3
493 vegetation in the case of the afforested soils in United States enabled us to methodologically
494 apportion the sources of soil C and to identify these direct contributions from trees to increasing
495 soil C storage. In the case of the Russian sites, soil ^{13}C isotope composition does not resolve
496 these C sources because of the lack of shift between C4 and C3 vegetation in the natural history
497 of these landscapes.

498 By contrast to afforestation, long-term annual cropping implies recurrent soil mixing,
499 disruption of any preexistent vertical stratification, microclimate fluctuation, and exposure of
500 SOM to decomposition, which collectively shift the C balance and predispose towards depletion
501 of soil C (Post and Kwon, 2000; Hernandez-Ramirez et al., 2009; Curtin et al., 2014). In addition
502 to this disturbance and exposure of SOM to decomposition, low C inputs and high C removals
503 via harvest are also characteristic of conventional annual cropping systems. A reduction of C
504 inputs in croplands once the native vegetation (roots and aboveground biomass) have been
505 removed (Hu et al. 2013) is also typically followed by alterations in soil physical properties such
506 as decreases in porosity and gas exchange which can become detrimental to plant primary
507 productivity and soil biology (Kiani et al., 2017).

508 Our study explicitly examined and quantified for first time in literature the losses of
509 remaining prairie-C directly beneath trees across afforested soils (Fig. 3, Fig. 5B, Fig. 7). This
510 analyses showed that under afforestation, soil C remaining from original native grasslands
511 continues to be lost from the profile (Fig. 3, Fig. 7), likely via microbial mineralization. This
512 noted decline in remaining prairie-C beneath young afforestation agrees well with a rapid
513 decomposition of SOM in the early stage of tree growth as previously deliberated by Paul et al.



514 (2002), Garten (2002), and Xiong et al. (2020). At the Norfolk site, tree-C contributions
515 effectively replenished and greatly surpassed the gradual losses of remaining prairie-C in the soil
516 (Fig. 7C). In the case of Huron site, trees generated the conservation of the entire prairie-C
517 legacy while also contributing directly to additional tree-C accrued in an overall increasing SOM
518 pool (Fig. 7A).

519 It is noted that although the three US sites (Norfolk, Huron and Mead) shared a common
520 trajectory of tree-C accretion with time (Fig. 5, Fig. 6), their k turnover rates of remaining
521 prairie-C differed (Fig. 7). These apparent divergences are potentially attributable to differences
522 in temperature and moisture regimes across the region. In further details, in the case of accretion
523 of tree-C in afforested soils, this response to land use change seems governed mostly by the
524 change into tree vegetation and the duration of afforestation; therefore, it became feasible for us
525 to establish a unified, robust k model across a range of afforestation ages (Fig. 6). Conversely,
526 loss rates of remaining prairie-C in afforested soils appeared to be mostly contextual and even
527 site specific, likely as a function of local hydrothermal conditions (Chendev et al. 2014, 2015b).
528 Relative to both Norfolk and Huron (Fig. 7), warmer-wetter conditions in Mead could have led
529 to the faster C turnover rate and mineralization of the remaining prairie-C in these afforested
530 soils (Fig. 7B). Overall, these results exemplify how analyzing the compartments of soil C
531 turnover – evaluating separately tree-C contributions and remaining prairie-C, instead of
532 studying only the whole soil C – can provide further insights into SOM dynamics following land
533 use conversions. Future research could address the potential existence of underlying thresholds
534 of heat and moisture availabilities that are conducive to retain and converse pre-existing prairie-
535 C in afforested soils while simultaneously enabling soil C accretion directly from new tree-C
536 contributions.



537 Based on the kinetics-modelled reconstruction of soil C storage over time in the Russian
538 land use chronosequences (i.e., encompassing a range of different duration of cropping since
539 land use conversion), over the 55 years that elapsed since tree planting until soil sample
540 collection, the remaining soil C from the original native grassland was shown to continually
541 being lost (Fig. 3). Our turnover estimations using kinetics modelling suggested that only 1.7%
542 of the initial grassland-C was lost over these 55 years following shelterbelt afforestation (Fig. 3).
543 Mellor et al. (2013) stated that mycorrhizae development in afforested soils can preferentially
544 access and utilized remaining prairie-C beneath trees. This biological effect could contribute to
545 decreases in remaining prairie-C in afforested soils.

546 Chendev et al. (2014, 2015b) further addressed differences in soil C accrual across
547 afforested sites in Russia and the United States, also attributing them primarily to differences in
548 moisture regimes. Within each geographic region as well as in the collective of both countries,
549 they explained that cooler-moister conditions led to increases in overall soil C accrual beneath
550 trees. This postulate is clearly in line with earlier results by Garten (2002). Potential increases in
551 plant primary productivity with increasing moisture as well as reductions in microbial
552 mineralization of the overall SOM with colder conditions can shift and drive the C balance in the
553 soil towards net C accrual.

554 Of the three paired sites in Russia (Table 1), the Yamskaya site is probably the most
555 representative and closely related to the three long-term chronosequences evaluated in this study.
556 This is because Yamskaya and the three chronosequences are all geographically located within
557 the Belgorod oblast, and hence, they share a more similar regional hydrothermal regime. It was
558 striking that the afforested soil at Yamskaya site had a soil C accretion even greater than the
559 native grassland reference, which strongly indicated the high capacity of shelterbelts to sequester



C even beyond the capacity of the corresponding native ecosystem (Table 1). After noticing this finding, it can also be anticipated that although the drier Kamennaya site showed at the present the slowest soil C accretion following afforestation of SOM-depleted croplands (Table 1), it is possible that in the long term, this drier environment can gradually sequestered even more soil C than the moist sites located in Belgorod oblast (e.g., Yamskaya). This is suggested as the Kamennaya site exhibited the highest soil C storage when comparing across all the native grasslands compiled in our study (i.e., 152.5 Mg C ha⁻¹; Table 1 and Fig. 2).

4.2 Turnover rates of soil carbon as a function of land use changes

This study clearly confirms that the long-term dynamics of soil C is consistently nonlinear, either during decline or accumulation of soil C as a function of land use choices. As deliberated earlier by Post and Kwon (2000) and Garten (2002), erroneously assuming linearity in depicting these trajectories of soil C would lead to underestimating the rates of soil C changes during the first decades following a land use conversion as well as overestimating the turnover rates of soil C after multiple decades once the ecosystem has actually reached stability and a balance between their C inputs and outputs. This latter notion essentially applies when long-term cropland or afforested fields have become mature (Hernandez-Ramirez et al., 2011)

Earlier chronosequence and stable isotope analyses by Arrouays et al. (1995) in Southwest France further support that land use effects on soil C changes take place rather quickly. They reported that a new equilibrium in soil C storage was reached within only few decades of a land use change from forest to annual croplands, and with about half of the C loss occurring rapidly within few years (< 10) of beginning cultivation (Arrouays et al., 1995). Similarly, as in our study, only a few decades seems to be required to reach equilibrium when switching from cropland to trees (Richter et al., 1999; Paul et al., 2002; Guo and Gifford, 2002).



Likewise, comprehensive results by Dhillon and Van Rees (2017) depicting soil C accretion caused by afforestation in the Canadian Prairies can be interpreted as net C losses taking place over the first several years after tree planting, and subsequently, an ensuing fast accrual of soil C until tree ages of about 35 years when new equilibria or C sequestration ceilings under afforestation can be reached. In our study, MRT of soil C accretion beneath trees were in general determined to be about two decades (Fig. 6). Furthermore, the evaluation of our two scenarios of asymptotic equilibria of C accrual in afforested soils (i.e., trajectories A and B in the normalized Russian chronosequences; Fig. 3) can provide the boundaries of faster vs. slower C accretion with MRTs of one vs. three decades, respectively.

In the case of long-term annual croplands in Russia, the MRT of 100 years for soil C depletion found in our study (i.e., associated with a k rate constant of 0.010 years^{-1} , Fig. 3) is comparable to findings by Huggins et al. (1998) who registered MRTs of 91 and 143 years in annual cropping systems in Minnesota, but overall longer than a report by Collins et al. (1999) who found a wide range of MRTs between 18 to 96 years for sites with 8 to 33 year-old continuous maize cropping across the Central United States, respectively.

It is noted that the exponential first-order trajectory of soil C turnover in the Russian chronosequences (Fig. 3) was captured better with Eq. [1] than the simplistic $C_{(t)} = C_o \times e^{-kt}$ previously used by Hernandez-Ramirez et al. (2011). While Eq. [1] provided an R^2 of 90% (Fig. 3), $C_{(t)} = C_o \times e^{-kt}$ returned an R^2 of 68% (data not shown). With two fitting parameters (i.e., C_o , C_e), first-order kinetic modelling with Eq. [1] represented reasonably well the assumptions of steady state and new equilibrium at the end of the evaluated time series (Fig. 3, Fig. 6).

Further kinetics modelling efforts of soil C increases in afforested systems can take the form of two functional C pools where inputs and outputs to labile and recalcitrant SOM can be



606 predicted (Arrouays et al., 1995; Garten, 2002; Hernandez-Ramirez et al., 2009; Xiong et al.,
 607 2020). Preferentially accruing C into recalcitrant vs. labile SOM pools in afforested soils can be
 608 interpreted as tree-C contributions towards long- vs. short-term stability of soil C storage,
 609 respectively, with crucial ramifications for mitigation of future climate change (Laganiere et al.,
 610 2010; Hernandez-Ramirez et al., 2011; Deng et al., 2014). Future investigations can also focus
 611 on the protection and stabilization mechanisms of SOM as created by soil aggregate formation
 612 beneath trees (Kiani et al., 2017; Quesada et al., 2020). Once soils subjected to long-term annual
 613 cropping are converted to permanent vegetation, fungal hyphae can become an important means
 614 that mediates C accretion by enhancing soil aggregation (Jastrow et al., 1996; Kiani et al., 2017).
 615 Jastrow et al. (1996) indicated that fungal hyphae could improve macroaggregation and hence
 616 indirectly enhance C accrual. Using phospholipid fatty acid biomarkers, Kiani et al. (2017)
 617 identified a linkage between presence of fungal biomass and increases in hierarchical fractal
 618 aggregation specifically in forest soils, while this association was absent in the adjacent
 619 annually-cropped soils in their study. Furthermore, Quesada et al. (2020) recently discussed the
 620 mechanisms for soil C accretion in tropical forests. In line with earlier findings by Wang et al.
 621 (2016), Quesada et al. (2020) stated that SOM physical protection provided by the formation of
 622 soil aggregates slows decomposition of SOM within aggregates, and hence, it becomes a second
 623 layer of stabilization after realizing the primary SOM stabilizing effects caused by mineral
 624 surfaces of fine soil particles such as silt and clay. Further studies can focus on the effects of
 625 inherent mineralogy and texture as well as clay leaching processes on C dynamics and storage in
 626 afforested soils (Chendev et al., 2020; Quesada et al., 2020).

627

628 **5 Summary**



629 Nonlinear turnover rates of soil C revealed an MRT of a century in long-term croplands
630 as soil C slowly undergoes depletion and losses to the atmosphere. Likewise, when croplands
631 were afforested, nonlinear accretion rates of soil C indicated a MRT of approximately two
632 decades following afforestation. Soil C showed to be rapidly accrued as trees remove CO₂ from
633 the atmosphere and contribute C substrates for SOM accumulation and stabilization. Results
634 collectively substantiated that in addition to other important benefits by trees such as providing
635 air quality, microclimate regulation and erosion control (Sauer et al., 2007; Hernandez-Ramirez
636 et al., 2012), C sequestration in afforested lands is a suitable means to proactively address and
637 effectively mitigate ongoing climate change within a person's lifetime.

638



639 **Data availability**

640 Data are available on request.

641

642 **Author contribution**

643 GHR: Conceptualization; Methodology; Formal analysis; Funding acquisition; Resources;

644 Visualization; Writing the original draft; Review and editing new versions; Corresponding

645 author role.

646 TJS and YGC: Conceptualization; Data curation; Formal analysis; Funding acquisition;

647 Investigation; Methodology; Supervision; Project administration; Resources; Visualization;

648 Review and editing new versions.

649 ANG: Investigation; Methodology; Funding acquisition; Review and editing new versions.

650

651 **Competing interests.**

652 The authors declare that they have no conflict of interest.

653

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664



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Table 1. Soil organic carbon storages and differences within the 0 to 30 cm depth increment under three land uses (i.e., native grasslands, annual croplands and afforestation) in Russia. The nine values of soil C storage across the nine site-land uses were previously presented and discussed in Chendev et al. (2015b) and are repeated here for informing first-order kinetic modelling and estimations of C accretion rates when converting from annual croplands to afforestation as shown in Fig. 3.

Land use or descriptor	Streletskaya Steppe site, Kursk	Yamskaya Steppe site, Belgorod	Kamennaya Steppe site, Voronezh	3-sites mean	Standard error
Soil C mass density (Mg C ha ⁻¹)					
Native grassland (G)	126.2	138.0	152.5	138.9	7.61
Annual cropland (C)	109.3	127.2	123.6	120.0	5.47
Shelterbelt (Trees)	126.4	142.1	125.0	131.2	5.48
Net decrease G-to-C	-16.9	-10.8	-28.9	-18.9	5.32
Net increase C-to-Trees	17.1	14.9	1.40	11.1	4.91
G-to-C / C-to-Trees†	1.01	1.38	0.05	0.81	0.40

†Ratio representing the replenishing of depleted soil C by tree planting. These ratios were calculated as the absolute values of net increase from cropland to shelterbelt (trees) divided by net decrease from grassland to cropland.



Table 2. Soil organic carbon storage within the 0 to 15 cm depth increment contrasting annual croplands and afforestation in United States. Sources of soil C storage beneath the trees were allocated as tree-C versus remaining prairie-C using Eq. [4] and [5] and associated assumptions. Error bars are standard errors of the means.

Site	Tree age (years)	Whole soil C mass density		Tree-C contribution to soil C		Remaining prairie-C		Remaining prairie-C / whole C in cropland [†]
		Annual cropland (Mg C ha ⁻¹)	Beneath trees (Mg C ha ⁻¹)	Mass density (Mg C ha ⁻¹)	Proportion (%)	Mass density (Mg C ha ⁻¹)	Proportion (%)	Proportion (%)
Huron, S. Dakota	19	25.6 ± 0.7	37.5 ± 1.2	12.2 ± 0.8	32.6 ± 1.5	25.3 ± 0.7	67.4 ± 1.5	98.8 ± 2.8
Mead, Nebraska	35	36.2 ± 0.4	46.7 ± 1.5	17.4 ± 1.2	37.2 ± 1.8	29.3 ± 1.9	62.8 ± 1.8	80.9 ± 3.6
Norfolk, Nebraska	70	13.0 ± 1.3	28.0 ± 1.4	22.1 ± 1.1	78.9 ± 1.0	5.90 ± 0.42	21.1 ± 1.0	45.5 ± 0.3

[†] This ratio represents the proportion of remaining prairie-C relative to whole soil C in annual cropland. The magnitudes of both ‘remaining prairie-C’ and ‘whole soil C in annual cropland’ are shown in other columns of this same table. It is noted that balance (e.g. in Mead, 100 - 81 = 19) represents to the proportion of prairie-C lost in afforested soils since tree planting.

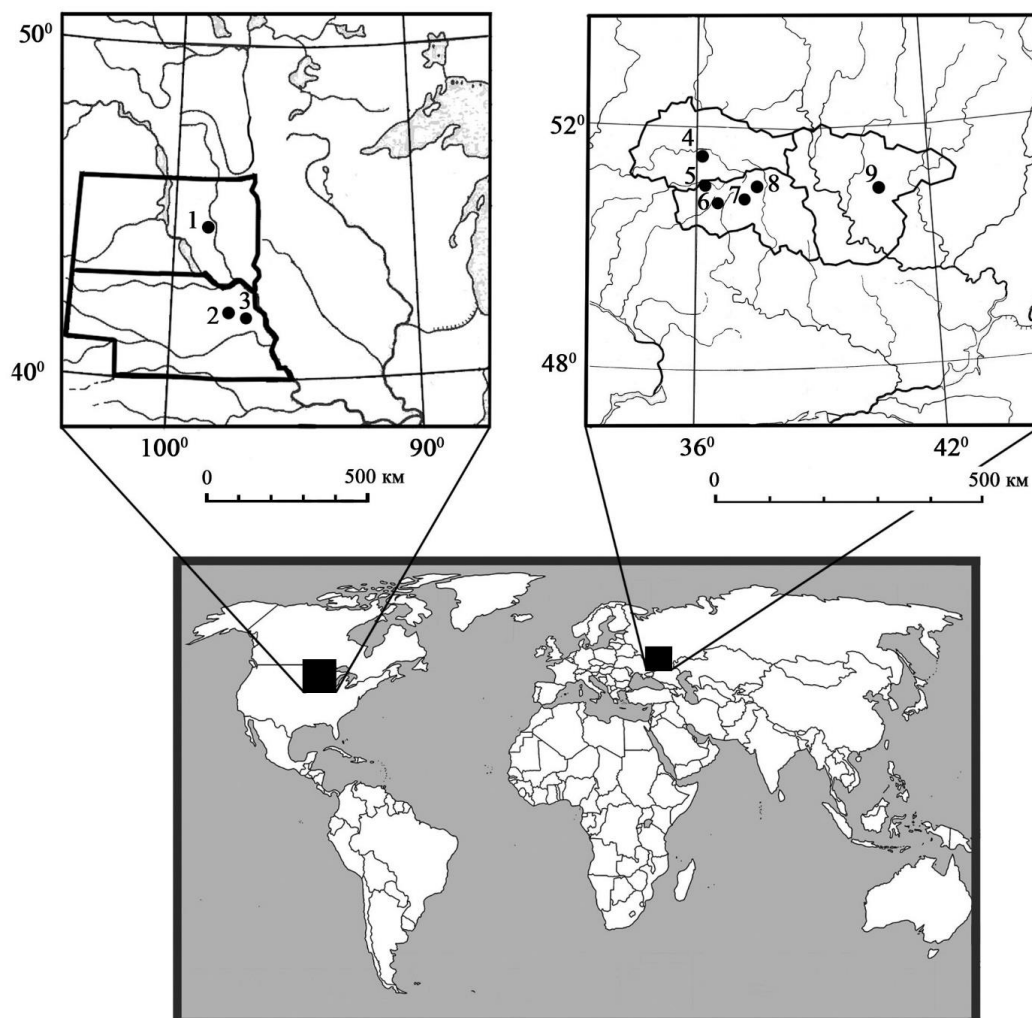


Fig. 1. Geographical location of the nine study sites within the United States (1. Huron, 2. Norfolk, 3. Mead) and Russia (4. Streletsкая Steppe, 5. Ivnyanskiy, 6. Prokhorovskiy, 7. Gubkinskiy, 8. Yamskaya Steppe, 9. Kamennaya Steppe). Within Russia, 5, 6, and 7 are sites with chronosequences of land use conversion, while 4, 8 and 9 are paired sites (native grasslands, annual croplands versus shelterbelts). The three sites in the United States are paired sites (afforestation vs. adjacent annual croplands).

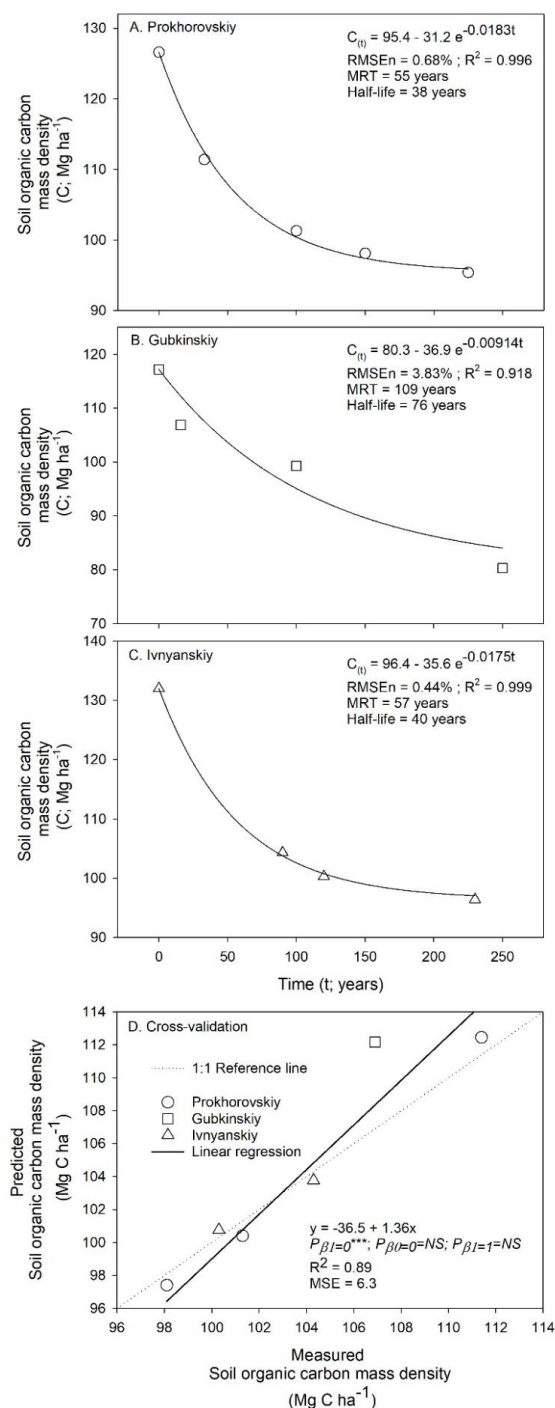


Fig. 2. Land use chronosequences of soil organic carbon storage within the 0 to 30 cm depth increment after converting native grassland to annual croplands in Belgorod oblast, Russia. These showed soil C declines over time. (A) Prokhorovskiy, (B) Gubkinskiy and (C) Ivnyanskiy districts. In Panels A, B and C, first-order kinetic models are described by the solid curvilinear fittings and equations in the form $C(t) = C_e + (C_0 - C_e) e^{-kt}$ where C_e is C at new dynamic equilibrium (inputs = outputs), C_0 is initial C at time of land use conversion (time zero), k is the first-order kinetic rate constant equivalent to turnover rate. As reciprocal of k , MRT stands for mean residence time, while half-life equates to $\ln(2)/k$. Normalized root mean square error (RMSEn) and coefficient of determination (R^2) for each k model are provided. (D) Cross-validation of first-order predicted C versus measured C encompassing the three chronosequences within the age range from 10 to 200 years. Mean squared error (MSE) and R^2 for the cross-validation are provided. The subscripts of the P values denote the null hypotheses for testing the regression coefficient (β_1) and intercept (β_0), where *** is a p-value of <0.001 and NS is non-significant. First-order kinetic modelling was supported by this performance evaluation.

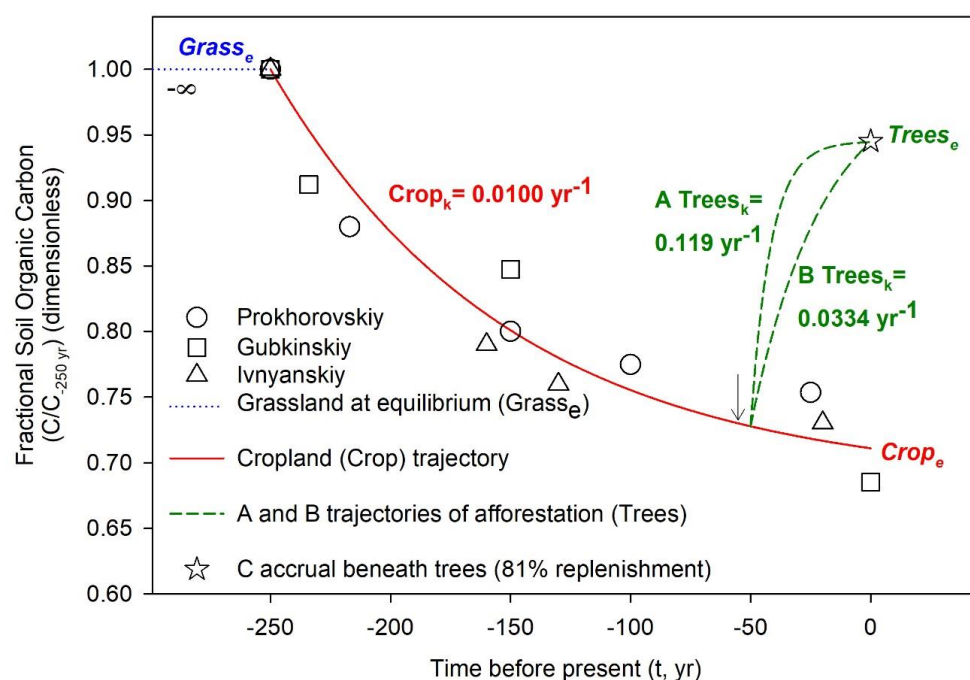


Fig. 3. Reconstruction of soil organic carbon storage within the 0 to 30 cm depth increment following land use conversions from native grassland to annual cropland and subsequently into afforestation with shelterbelts in Russia. This assemblage assumed that native grasslands were at dynamic equilibrium and steady state prior to conversion to annual croplands. Likewise, nonlinear k estimates of turnover rates in croplands and accretion rates under afforestation also required the assumptions of reaching new dynamic equilibria and steady state at zero year (i.e., time of soil sample collection). The cropland trajectory of soil C over time was derived from chronosequence data presented in Fig. 2A, Fig. 2B and Fig. 2C. The soil C accrual beneath trees at time zero (open star) was estimated from measured data presented in Table 1 (i.e., of the soil C that had been depleted by cropping, afforestation replenished 81%, based on 3-sites mean). Note that although trees were planted on year -55 (vertical arrow \downarrow), tree-C contributions to soil C accrual were accounted for starting from year -50 based on a literature review by Paul et al. (2002) that suggested a lag phase of 5 years. The trajectory ‘B Trees’ (dashed red line with $k = 0.0334 \text{ yrs}^{-1}$) assumed that the soil C storage had asymptotically reached 95% of a theoretical equilibrium (i.e., ‘Trees_e’ / 0.95). First-order kinetic modelling was used to derive these three nonlinear trajectories of soil C with the general form $C(t) = C_e + (C_o - C_e) e^{-kt}$. With the aim of integrating information from the three available chronosequences (Fig. 2), all soil C storage data were normalized (i.e., zero to one) and presented here as fractions of C storage at time of conversion from native grassland to annual cropland (shown as -250 years before time of soil sample collections).

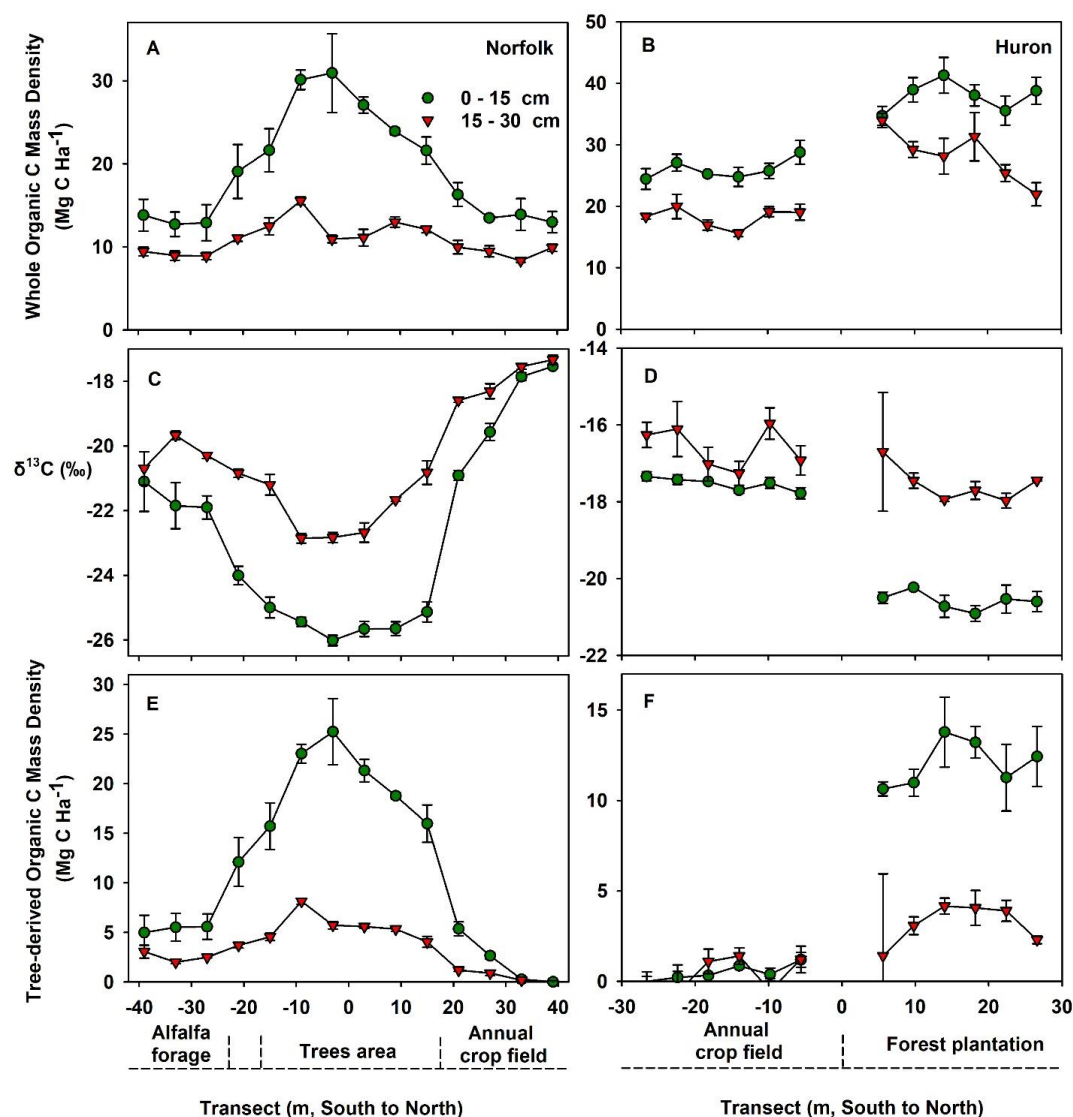


Fig. 4. (A, B) Soil organic carbon mass storage, (C, D) stable isotope ratios of organic C ($\delta^{13}\text{C}$), and (E, F) organic C mass derived from C3 plants across transects at Norfolk shelterbelt (left panels) and Huron forest plantation (right panels) for the 0 to 15 and 15 to 30 cm soil depth increments. Adjacent cropped fields were also included. Within the afforested areas in panels E and F, the reported organic C masses are primarily attributed to direct tree contributions. Contributions of tree-C to soil C storage were clearly discernable within the 0 to 15 cm depth increment. Note the difference vertical scales across panels. Error bars are standard errors of the means, with sample sizes of 4 for Huron and 3 for Norfolk.

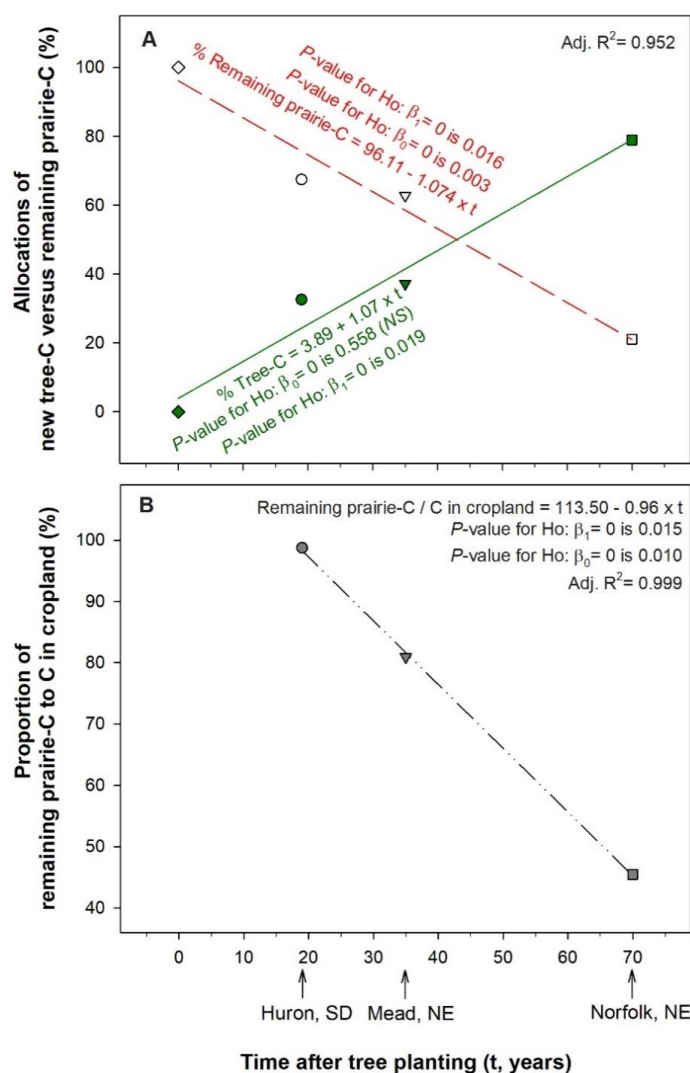


Fig. 5. Proportions of soil organic carbon within 0 to 15 cm depth increment (A) from tree-C versus remaining prairie-C relative to whole soil C stored directly beneath trees, and (B) remaining prairie-C in afforested soils relative to whole soil C in the adjacent croplands. It was inferred that all soil C different from new tree-C was preexisting soil C attributable to remaining prairie-C. (A) Mead data was recalculated from Hernandez-Ramirez et al. (2011) as compiled in Table 2. The tree-C source increased to become 79% of the whole soil C over 70 years after tree planting. (B) The proportion of prairie-derived C beneath trees declined over time to become less than half (45%) of the whole soil C in the adjacent annual croplands, which were assumed to be at steady state.

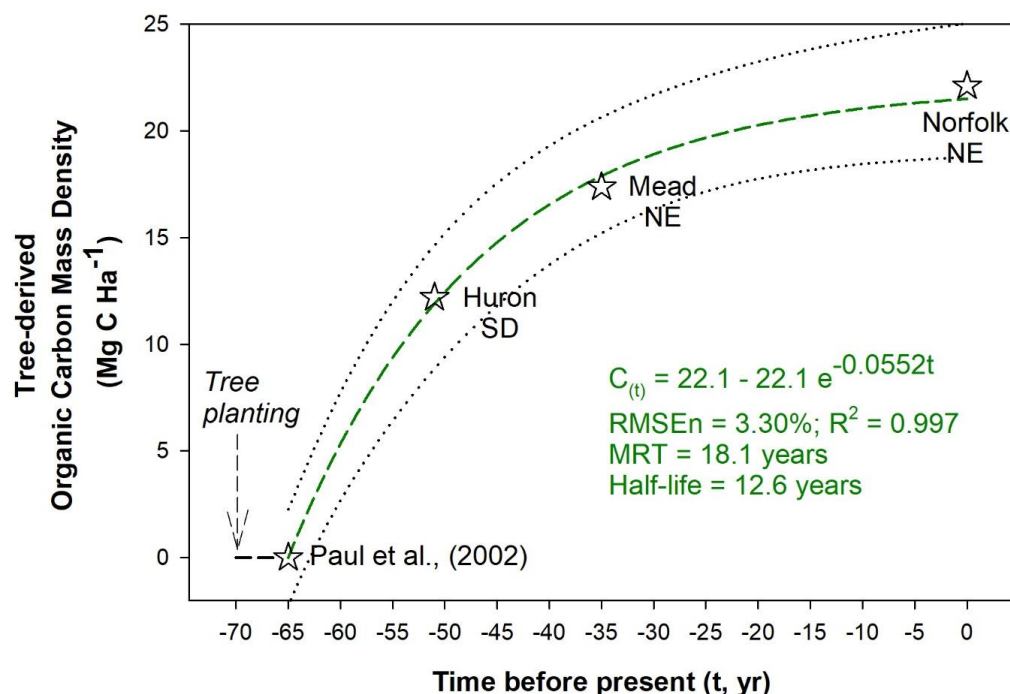


Fig. 6. Reconstruction of soil organic carbon storage within the 0 to 15 cm depth increment following land use conversion from annual cropland to afforestation United States. The arrow indicates the time of tree planting (-70 years). Based on a literature review by Paul et al. (2002), we included a lag phase of 5 years following tree planting. Huron and Norfolk data were derived from results presented in Fig. 4. Mead data was recalculated from Hernandez-Ramirez et al. (2011) as compiled in Table 2. This assemblage supports that afforested soils were approaching steady state at nearly 70 years after tree planting, as required for first-order kinetic modelling. The first-order kinetic model is depicted by the solid curvilinear fitting and equation in the general form $C(t) = C_e + (C_o - C_e) e^{-kt}$, where k is the first-order kinetic rate constant equivalent to accretion rate under afforestation, and MRT stands for mean residence time. Normalized root mean square error (RMSEn) and coefficient of determination (R^2) for the k model are also provided. The 95% prediction bands of this k model are provided as dotted lines. This nonlinear trajectory describes and highlights the contribution of trees to soil C accrual.

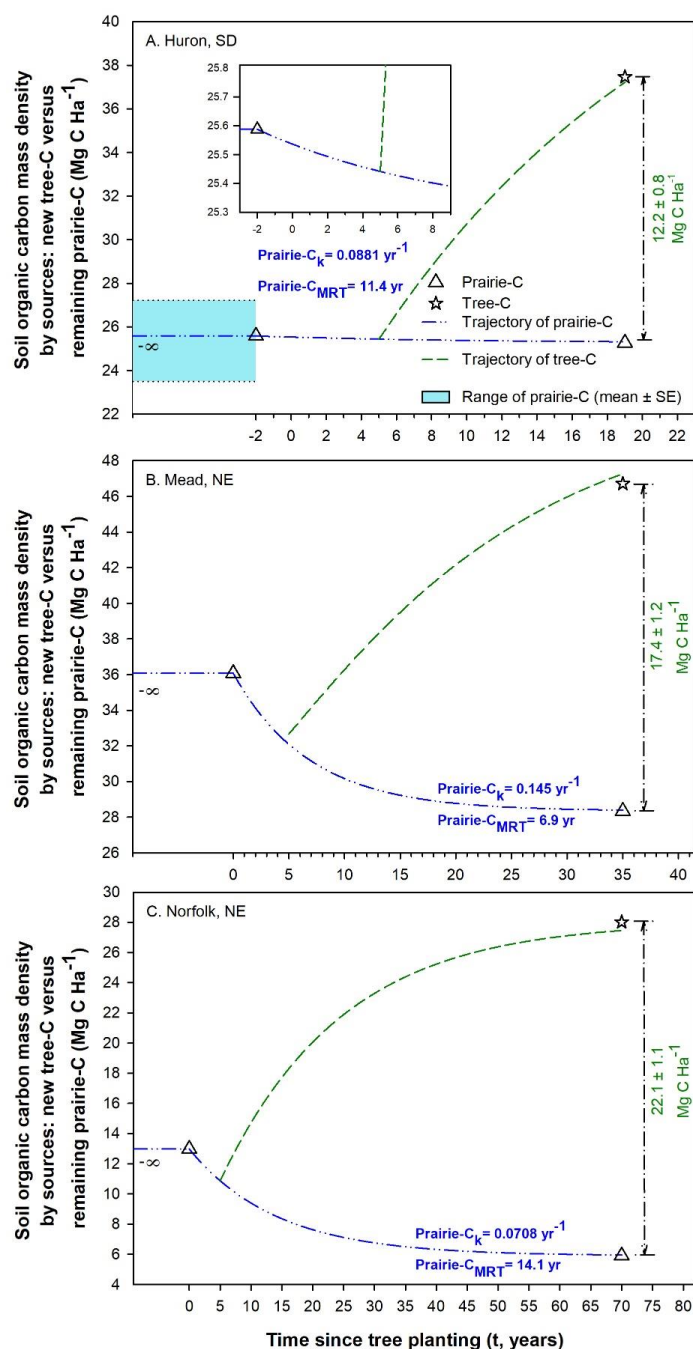


Fig. 7. Dynamics of net carbon accretion in afforested soils caused by simultaneous C gains from substantial tree-C contributions and smaller C losses from mineralization of remaining prairie-C. It was inferred that all soil C different from new tree-C was preexisting soil C attributable to remaining prairie-C. Time of tree planting was set at the year zero. Based on a literature review by Paul et al. (2002), we included a lag phase of 5 years following tree planting. The accretion trajectories of tree-C presented within each Panel were projected using the unified first-order model developed in Fig. 6. The magnitude of tree-C gains as well as the turnover rates (k) and mean residence times (MRT) of prairie-C trajectories are provided within each panel. The Huron site (Panel A) had an adjacent native grassland available, which here is provided as a reference and plotted prior to the time of conversion into annual cropland. Note the different x and y scales across panels.