University of Sopron

Faculty of Forestry

Theses of doctoral (PhD) dissertation

Effects of sessile oak and black locust forests on soil-aggregate dynamics and SOM sequestration.

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1. Introduction and aims of the research

On a global scale, soils (the pedosphere) are the third largest carbon (or organic matter) sink after vegetation and oceans, and are also a fragile, conditionally renewable natural resource. Well-functioning ecosystems capable of providing many functions and fertile sites, i.e. well supplied with nutrients and moisture imply a morphologically diverse and complex structure. The focus of our research has therefore been on structural components beyond soil organic matter: micro- and macroaggregates.

Based on the preliminary study with black locust (BSc thesis), with the National Forest Inventory Database another species the sessile oak was selected, which is planted in Hungary on similar sites and climate conditions (also in monoculture) as black locust. A review of the international literature has revealed several articles on SOM sequestration in forest soils. One of the most frequently described phenomena is that with higher plant diversity (i.e. a higher number of tree species in a given area, but also a higher development of undergrowth), the carbon stock sequestred in the soil tends to increase and the overall soil properties improve. It should be noted, however, that these effects can vary greatly in the accumulation and mineral horizons; the effect is more pronounced under coniferous forest stands. The effect of specific tree species on the permanently sequestrated carbon stocks of their sites has been little studied.

There are also few studies that analyze the effects of woody vegetation on aggregate formation and stability, and these do not confirm a clear tree species influence or report significant spatial variations within specific soil profiles. There are considerably more papers available on the influence of field crops (e.g. maize, peas, wheat) on soil aggregates.

To enable the investigation of the above discussed correlations within a realistic (feasible) framework, a special chronosequence, space-for-time substitution was used. Thus, we were able to compare the soils of first- an n generation (more logging, de- and reforestation) black locust and oak stands with unforested control sites. The method allowed us to study simultaneously the influence of the longer or shorter presence of tree species on a range of sites (with uniform sites conditions) on soil development. To improve the generalizability of the conclusions, this experimental scheme was applied in two regions of the country (Somogy and Nógrád counties), with two independent but matched sets of measurements.

For our work, we used both existing and novel test methods. In the latter case, we also aimed to test the applicability and suitability of the new method for forest soils. To this purpose, we carried out parallel measurements on a large number of samples.

Specific aims:

- Laboratory analysis of soil samples from a total of 16 sites. Determination of physical and chemical properties relevant for aggregate formation and SOM sequestration. The data obtained allowed a basic comparison of the different sites and soil development conditions.
- Examination of the percentage distribution of primary soil particles (mineral components of aggregates) by size class at all research sites. This way we obtained information about the initial state and conditions of soil development.
- Electro-hydrostatic testing of the fine fractions (clay and silt fractions) and the evaluation of the efficiency and applicability of this method.
- The fractionation of soil organic matter (SOM) according to the strength (persistence) of the sequestration. This method was used to identify/measure the role of the studied tree species in the short-, medium- and long term soil carbon sequestration.
- Investigate the size distribution, morphology and stability of aggregates, thereby the influence of the studied tree species on soil aggregate dynamics.

- Structure the measured physical and chemical parameters chronosequentially and thus construe the carbon sequestration, aggregate formation (ultimately soil development) process, as well as to reveal the influence of individual tree species.
- Provide new data on the carbon sequestration and aggregate dynamics on Cambisols in Hungary, with the aim of closing the knowledge gap and clarifying possible errors or inconsistencies.

2. Material and methods - Sampling- and analytical methods

2.1. Sample collection and processing

For the study, organic and mineral horizons of Cambisols were sampled in the Külső Somogy and Középső-Cserhát-vidék forest landscapes in the 'turkey oak-sessile oak climate' on loess bedrock, at uniform depths of 0-5 cm, 5-10 cm and 10-30 cm. Sampling was carried out at a typical (representative) location of the site, taking into account local characteristics (topography, vegetation, absence of disturbance). Sample preparation was carried out according to ISO 11464. For the analyses of mineral horizons samples were air-dried at 40°C, structural and root fragments were removed and base material were sieved (<2 mm). The bulk density was determined from soil samples collected with 100 cm^3 volume cylinders.

2.2. Measured parameters and analytical methods

pH measurement. Specifying the acidity or basicity of soil samples was performed by potentiometric measurement of distilled aqueous and potassium chloride soil suspensions according to ISO 10390.

Determination of organic and inorganic carbon content and the nitrogen content. The organic and inorganic carbon content was determined by catalyzed dry combustion according to ISO 10694. The nitrogen content of the samples was also determined in the same way and on the same equipment (Elementar Vario EL cube device) in a single run with the carbon content determination.

Determination of soil texture. The mass distribution of primary soil components was determined by combined sieving-sedimentation tests. Fine fractions (clay and silt fractions) were determined by both Köhn-pipetting (ISO 11277) and electro-hydrostatic principles (Pario® instrument) by parallel measurements, while sand fractions were separated uniformly by wet sieving. The sample preparation (H_2O_2 digestion + chemical ($Na_4P_2O_7$) and gentle physical dispersion) was identical in all cases.

Density difference-based SOM fractionation. In this study, the procedure developed by Golchin et al. (1994) was used. The

density of the fractionation medium (3 Na_2WO_4 - 9 WO_3 - H_2O) was ρ = 1.6 g · cm $^{-3}$. After measuring the dry mass of the separated fractions (fLF, oLF and HF), their carbon and nitrogen contents were determined.

Digital dynamic image analysis. To investigate the size distribution of aggregates and primary mineral soil constituents, as well as aggregate stability, we used the QicPic automatic image analyzer system in combination with Lixell - Sucell modules. The samples were fed into the device as aqueous suspensions. Thus, in addition to the size, geometric characteristics were determined in the range 15 - 1100 µm, and the data were grouped according to the smallest Feret diameter ($Feret_{min}$) and particles with sphericity lower than S = 0.95 were excluded from the measurement. To investigate aggregate stability, the ultrasonic unit of the device was used to impart ~0.30 Joule · ml^{-1} of destructive energy in two steps, followed by repeated measurements.

Statistical evaluation. The data and measurement results discussed in this dissertation were evaluated using the statistical platform R and R-Studio (version 4.1.2). To test for significant differences, we used the Kruskal-Wallis test (H-test), supplemented by 'post-hoc' paired tests (Dunn's test), where p-values were adjusted using the Bonferroni method. Two-tailed variance between SOM-fractions was tested using Wilcoxon-

Mann-Whitney test (U-test) and linear mixed-effects models (LMMs) were used to reveal the factors affecting soil carbon sequestration.

The confidence level used in the dissertation is $(1 - \alpha) = 0.95$.

3. Results and conclusions – theses

Summary of the new scientific results obtained from the research in thesis points:

1. The results obtained with the 'Pario classic'method, which is a more efficient tool for testing the texture soil samples, do not match the results obtained with the Köhn-pipette, especially for higher clay contents (The middle silt fraction was an exception from this.). With the (semiempirical) linear regression functions developed by us, the fractions of faulty silt and clay fractions can be corrected with an error below <5% [m/m] (Figure 1.).
2. Our results demonstrated significant deviation in both the temporarily bound free light fractions (fLF) and the longer-term bound occluded light fractions (oLF) and heavy fractions (HF), both between sample and control sites and between first- and n-generation stands (Figures 2. a–b.). Although the influencing site factors and the influence of individual tree species on the 'stock change' differed

somewhat by fraction, depth level and between the two regions, the trends were still similar.

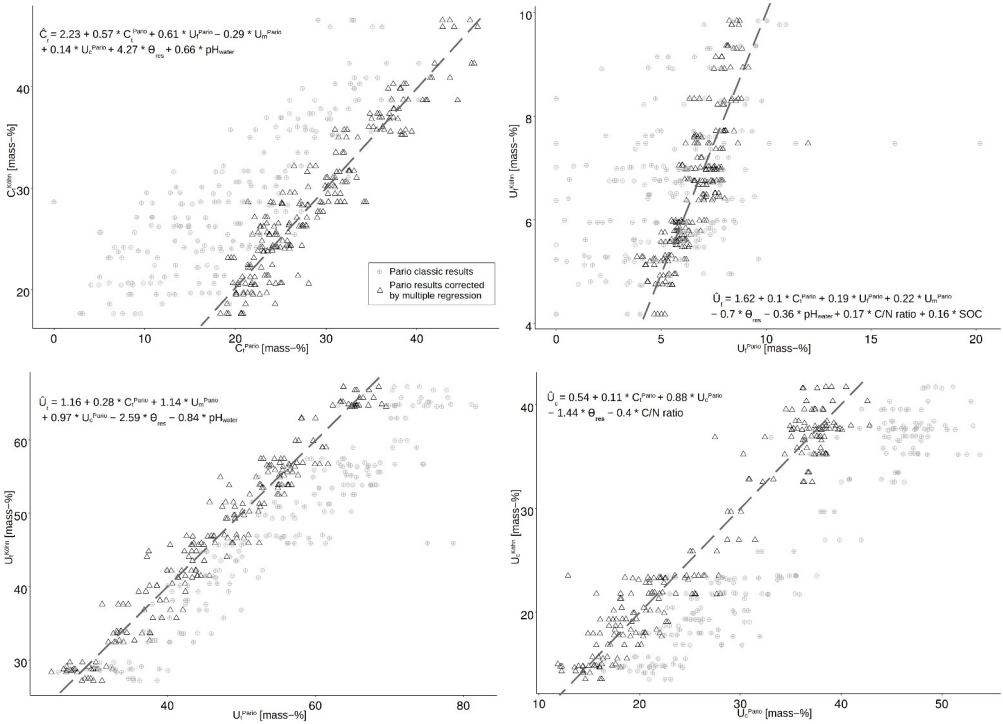


Figure 1. Correlation of original (grey cross) and corrected (black triangle) Pario measurements (x axis) with reference (Köhn-pipetted) measurements (y axis). From top left to right, in order: clay, fine silt, total silt, coarse silt fractions; and the regression equations.

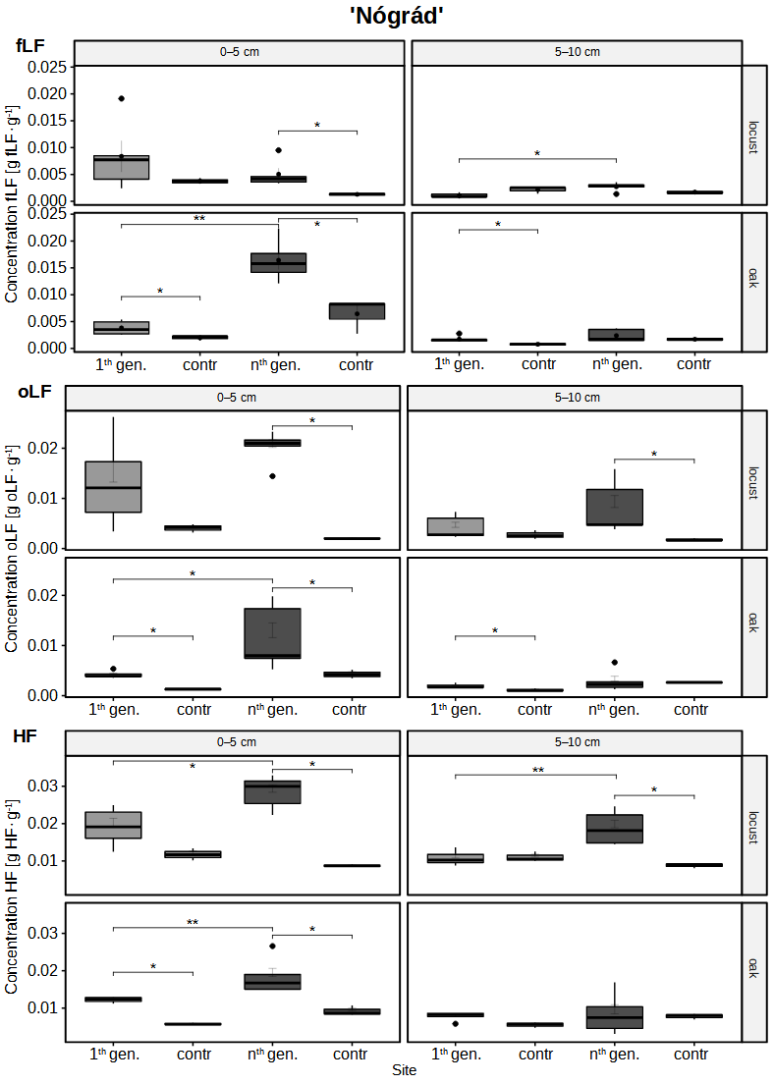


Figure 2. a. Quantitative development of temporarily, medium- and long-term sequestrated SOM in the different fractions in the 'Nógrád' region at depths of 0-5 cm and 5-10 cm. Significant differences (based on U-test) are indicated by brackets, significance level by asterisks: p-value: '***' p<0.001; '**' 0.001<p>0.01; '*' 0.01<p>0.05.

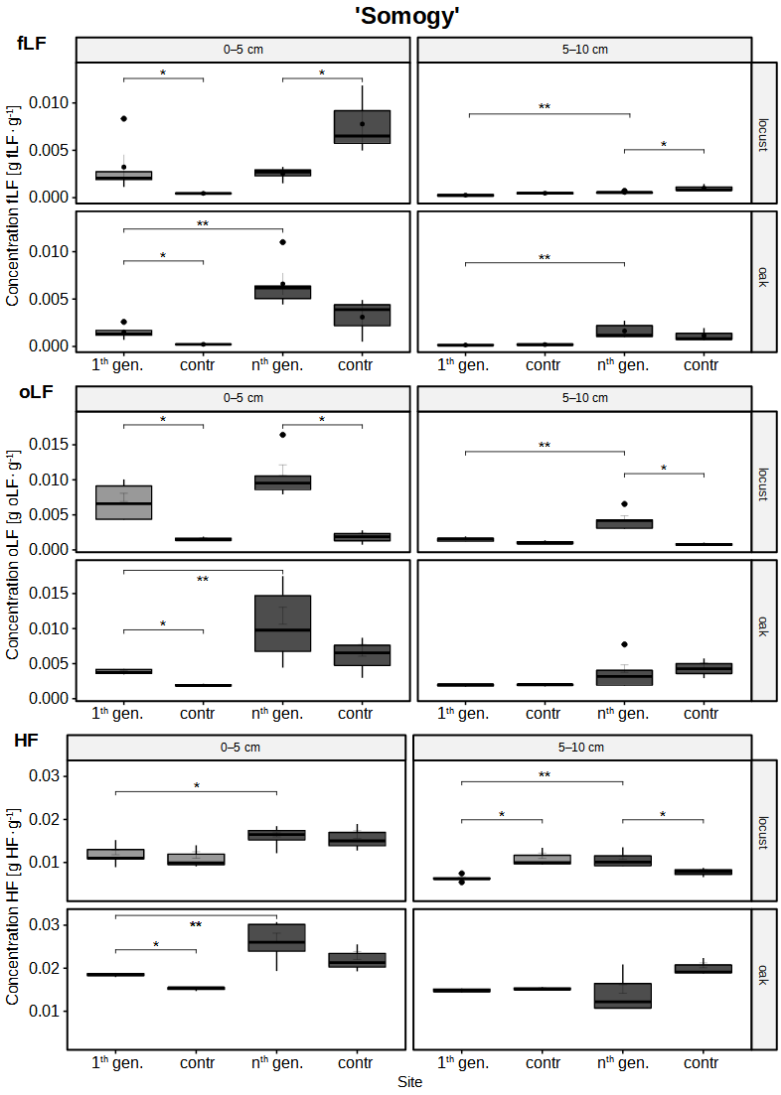


Figure 2. b. Quantitative development of temporally, medium- and long-term sequestrated SOM in the different fractions in the 'Somogy' region at depths of 0-5 cm and 5-10 cm. (For the legend on significance see figure 1.a.)

3. Using linear mixed-effect models, we demonstrated a significant positive influence of sessile oak compared to unforested control sites, both in the 5-10 cm mineral horizon for temporarily and long-term bound organic matter (fLF and HF fractions). In the case of black locust, a more complex relationship was observed with a similar tendency as in the previous cases. In the 0-5 cm layer, black locust had a significant negative impact on the amount of the free light fraction (fLF). But significantly higher amounts of occluded light fraction (oLF) were measured in the presence of black locust than on unforested sites, both at depths of 0-5 cm and 5-10 cm.

4. The modeling has shown that the quantitative development of some particular fractions in the soils investigated does not have a significant influence on the amount of other fractions. The exception is the heavy fraction (HF), which is significantly influenced by the amount of the free light fraction (fLF). Conclusion: the SOM sequestration process does not (necessarily) follow the expected path (fLF → oLF → HF).

5. A comparison of SOM fractions with the overall C/N ratios of the sites gives an indication of the mobility of organic matter on the specific site by means of the degree of microbial degradation (composting) (Figure 3.). Our measurements

show that the C/N ratios of both tree species (with the exception of the black locust in 'Somogy') in the plots of the n generation sites differed significantly from the control sites. The influence of the first generation forest stands is moderate and not significant compared to the control sites. In areas covered with woody vegetation, the regular input of forest litter (low-degradation SOM) was generally reflected in higher dispersion of the rates. For the light fractions (fLF, oLF), higher ratios than the reference C/N ratio were observed, indicating a higher proportion of lower degraded, 'fresh' dead organic matter in these fractions. The fraction with the lowest C/N ratio was in all cases the heavy fraction (HF), which is highly mineralised, indicating that in this fraction's organic matter is stored for a long time with little/no accessibility to saprobionts.

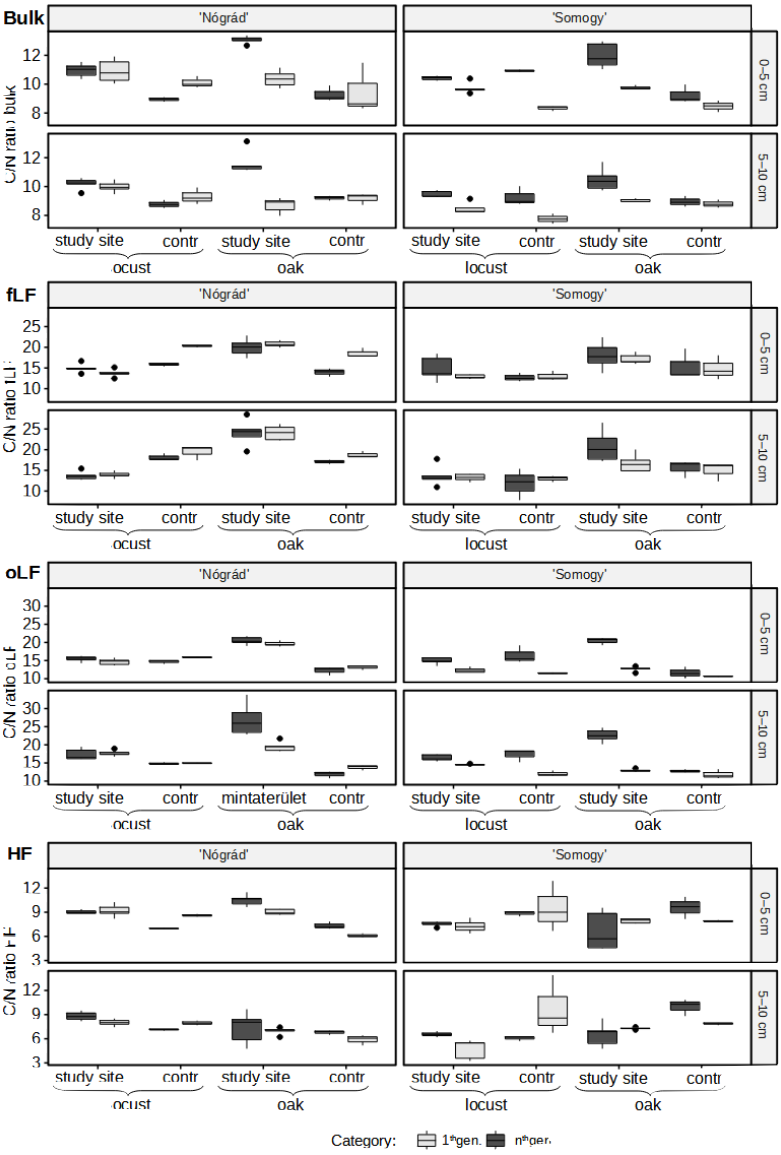


Figure 3. Development of the C/N ratio on the study sites and control plots; in the bulk sample (top) and in each SOM fraction (fLF, oLF and HF) at depths of 0-5 cm and 5-10 cm.

6. The size distributions of aggregates differed between the two regions and between tree species, generations and mineral horizons (Figures 4. a-b). The unforested control sites showed more homogeneous distributions than the sample sites with woody vegetation. More microaggregates (<250 μm) were detected in the 'Somogy' region than in 'Nógrád', which is also due to the higher silt and clay content of the 'Somogy' samples (more 'smaller' primary grains). According to aggregate size the ascending order was unforested sites<black locust sites<oak sites.

7. The aggregate stability tests (Figures 5 a-b) basically confirmed the already known inverse ratio correlation, that increasing size results in decreasing stability. But the stability of larger aggregates was somewhat higher in the oaks of the 'Nógrád' region.

8. The results confirmed the influence of the species of sessile oak and black locust on soil aggregate dynamics at the 0-30 cm mineral horizon.

9. The space-for-time substitution (special chronosequence) can be successfully applied to study carbon sequestration, aggregate formation (ultimately soil development) processes and to study the influence of individual tree species in these processes.

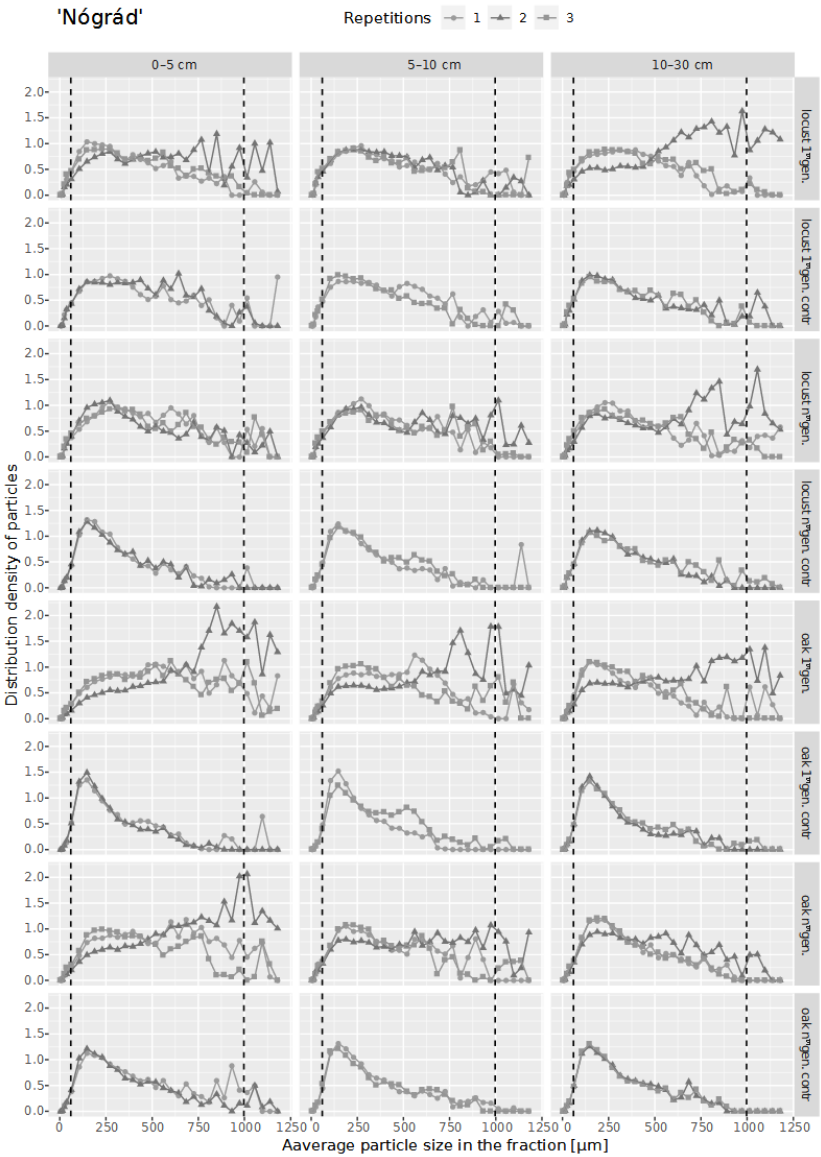


Figure 4. a. Size distribution of aggregates in the 'Nógrád' region at depths of 0-5 cm, 5-10 cm and 10-30 cm.

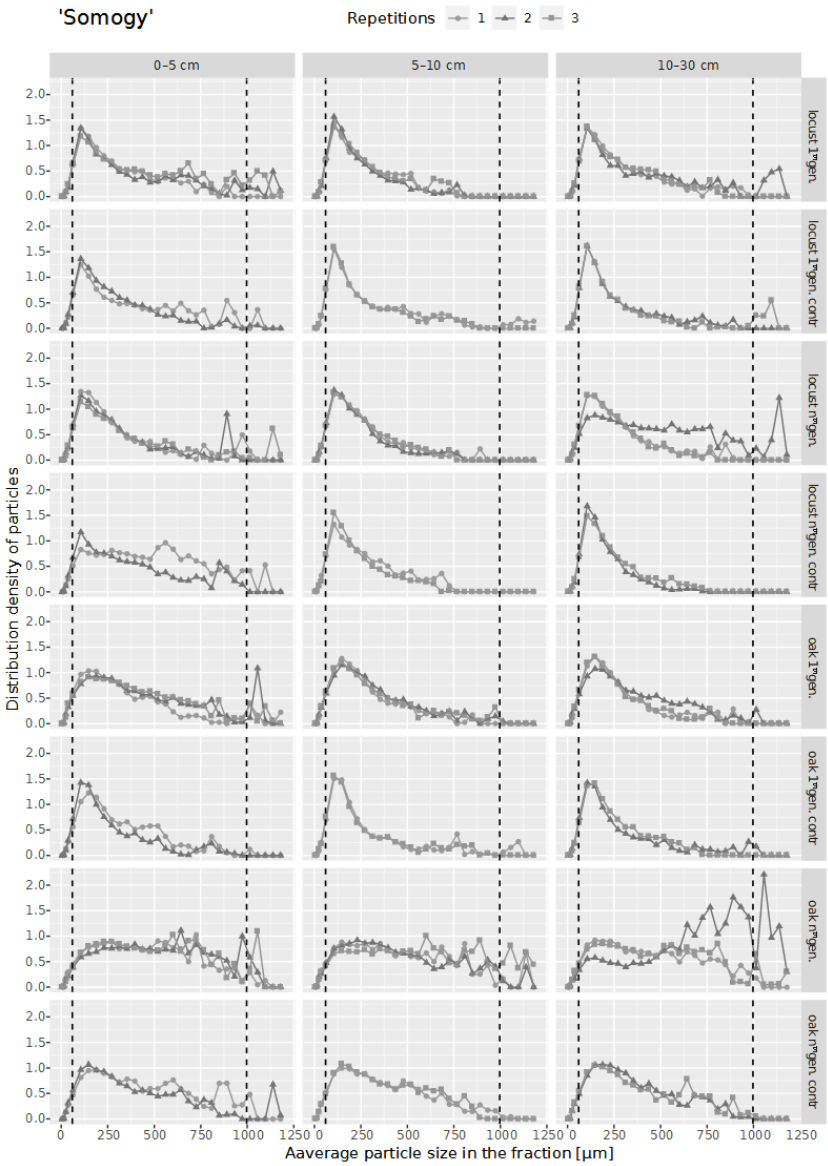
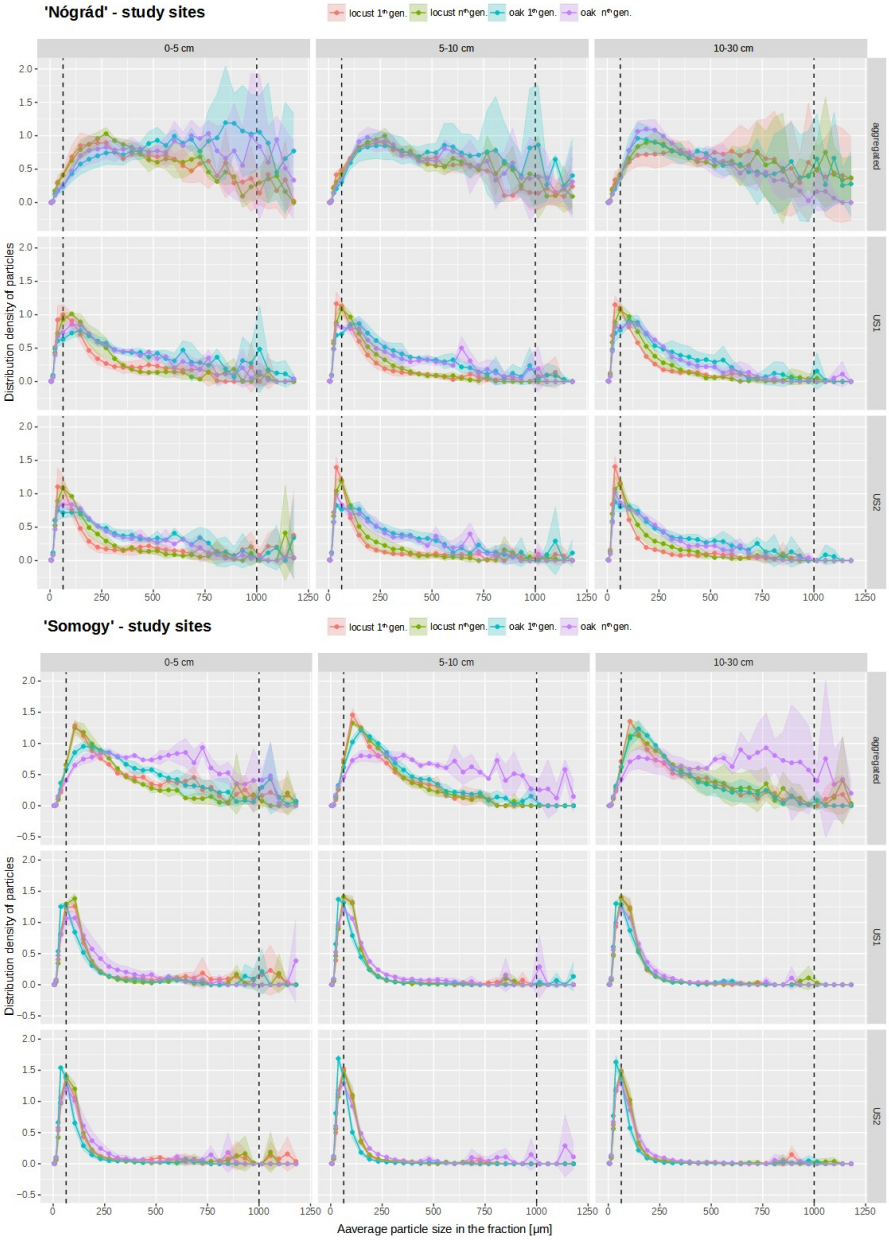


Figure 4. b. Size distribution of aggregates in the 'Somogy' region at depths of 0-5 cm, 5-10 cm and 10-30 cm.



Figures 5. a-b. Grain size distribution by frequency for aggregated and ultrasonically treated (US1 and US2) samples in the 'Somogy' and 'Nógrád' regions (forested sample sites) in the three analysed horizons.

4. Bibliograpy of the author's publications considering the dissertation

Publications in journals

Mátra, Z. S., Heil, B., Kovács, G., Lang, F., Schack-Kirchner, H. (2022): Increased accuracy and precision of hydrometric soil texture measurements ("Pario classic method") with semi-empiric correction functions. Journal of Plant Nutrition and Soil Science, 185, 417– 426. (Impact factor: 2.426)
https://doi.org/10.1002/jpln.202100213

Conference presentation

Mátra Z.S., Graf-Rosenfeller M., Lang F., Heil B., Kovács G. (2017): The impact of black locust (*Robinia pseudoacacia*) on soil development and biomass (C) fixation. HU-DE workshop on Carbon-Metabolism, Sopron. 11-12.03.2017