Chapter 4

Control of Soil pH, Its Ecological and Agronomic Assessment in an Agroecosystem

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Additional information is available at the end of the chapter

http://dx.doi.org/10.5772/intechopen.75764

Abstract

Lithuania is located in the humid zone, where mean annual precipitation exceeds mean evapotranspiration and soil acidification is an ongoing natural process encouraged by anthropogenic activities. Traditionally, the process may be controlled by different intensity liming. The chapter summarizes the data on long-term liming and fertilization experiments made in Western Lithuania. The object of the investigation is the naturally acid soil, Bathygleyic Dystric Glossic Retisol (texture: moraine loam with clay-sized particles content of 12–14%), and the same soil exposed for more than half a century to different liming and fertilization intensity. Our systematic analysis shows that it is impossible to reach appropriate moraine loam soil conditions for organic matter decomposition, carbon sequestration, soil aggregation, nitrogen fixation, nutrient accumulation, and plant growth by using intensive liming only. It is necessary to co-ordinate proper liming and organic fertilizing. The soil acidity was neutralized (pH_{KCl} 5.9 ± 0.1) and mobile aluminum abolished in the topsoil and subsoil to a 60 cm depth; moreover, the highest amount of soil organic carbon (1.91%), water stable aggregates (59%), intense nitrogen fixation, and highest grain yield was established in the periodically limed (with 1.0 rate $CaCO_3$ every 7 years) soil with 60 t ha^{-1} farmyard manure (FYM) application.

Keywords: soil pH, liming, manuring, microbial activity

1. Introduction

Soil is an integral part of nature. This means that the soil body is not only a recipient but also a donor of its own products and original materials like nitrogen, phosphorus, and carbon to other natural resources—mainly air and water [1, 2]. Modern intensive agriculture based on the use of substances of non-natural origin poses a major threat to nature and primarily to soil. It results in the disturbance of the vital ecological functions of the soil, including chemical buffering capacity, nutrient cycling, biodiversity, organic matter decomposition, as well as esthetic functions. The degradation of soil generally manifests itself by the loss of humus, decrease of biological activity, destruction of structure, increasing compaction, leaching of nutrients, runoff, and acidification [3, 4]. Soil acidification can be accelerated by intensive farming or prevented by sustainable management practices. Soil acidification management includes both neutralization of soil acidity and regulation of the acidification of limed soil. A key aspect in managing acid soils for plant growth is the pH amendment of the soil. Application of lime materials and organic substances could be an effective measure for improving soil chemical properties and the changes of their indexes depend on the amount of liming materials and frequency of application [5, 6]. Spatial patterns of topsoil pH in Lithuania are fundamentally controlled by the soil parent material. Topsoil, subsoil, and parent material collectively influence soil reaction and should not be treated as separate components [7]. Analysis of the pH dynamics in Lithuanian soils over half a century under the influence of hypothetical minimal liming level conducted 50 years ago, suggests that soil acidity neutralization must be an uninterrupted process. Once soil loses its chemical balance, it is less resistant to acidification than initially acidic soil. Liming is a strategic tool for improving acid soils, if the chosen liming intensity allows maintaining soil pH_{KCl} value at a level of 5.7–6.2, which is optimal for many crops. In Lithuania, a long liming tradition has led to a problem of overliming (topsoil pH > 7.0) of moraine loam soil, which results in morphological changes in the soil profile and increased calcium leaching, deterioration of topsoil structure, and decreasing crop yield [8]. This shows that soil pH is a critical parameter that influences the plant-soil-water interfaces. Soil pH impacts on a number of factors affecting microbial activity, like solubility and ionization of inorganic and organic soil solution constituents, and these will in turn affect soil enzyme activity. The soil bioavailability of many nutrients and toxic elements and the physiology of the roots and rhizosphere microorganisms are affected by soil pH [9].

Soil acidity regulates the rate of organic matter mineralization, impacting the release of carbon (C) and nitrogen (N) from soil organic matter. Also, soil acidity has a deleterious effect on the symbiotic relationship between rhizobia and legumes, reducing N fixation and the subsequent supply of N for soil-derived GHG (greenhouse gases) [10]. Changes in the environmental conditions occur in line with alterations in agrophytocenoses. Soil pH influences the changes in the species composition, structure, and plant and environment interaction of the agrophytocenoses.

In summary, the soil genesis and texture, the type of lime materials and their exposure time as well as climatic conditions are very important factors for the liming efficiency on soil pH, structure, and stocks of carbon and nitrogen. There exist opinions that efficiency of agricultural practices on soil pH changes depends on the geochemical environment in which these practices are applied.

In this chapter, we will briefly consider some aspects of the naturally acid moraine loam soil pH management practices and soil-plant interactions at various pH levels under the climatic conditions of West Lithuania.

2. Description of the experiment

2.1. Site

The chapter presents the scientific achievements of half a century research carried out in the Vezaiciai Branch of Lithuanian Research Centre for Agriculture and Forestry. The site of the study was Vezaiciai Branch, Lithuanian Research Centre for Agriculture and Forestry (LAMMC) located in West Lithuania's eastern fringe of the coastal lowland (55°43′N, 21°27′E). The present research was conducted in the crop rotation field devoted to the experiment on long-term liming and fertilization carried out since 1949.

2.2. Soil

Long-lasting geological processes have formed the soils of Lithuania. Prevailing in the country are Luvisols (21%) and Retisols (20.4%). The soil of the experimental site is Bathygleyic Dystric Glossic Retisol (WRB—World Reference Base, 2014) (texture: moraine loam with clay-sized particles content of 12–14%). According to the content of clay particles, the soil profile is differentiated into alluvial and illuvial horizons with diagnostic horizons: Ah-ElB-ElBt-BtEl-BCg. The soil is very acidic (pH_{KCl} 3.9–4.2) in its whole profile up to 160 cm depth and the amount of toxic mobile aluminum is very large, both in the topsoil and subsoil (respectively 100 and 300 mg kg^{-1}), the occurrence of calcareous rock were found up to more than 2 m depth. According to its profile differentiation and all acidity parameters, the soil under study is in priority in terms of the need for liming. The deficiency in clay (<0.002 mm), cations of Ca and Mg and organic colloids is the main factor that influences the low stability of acid topsoil aggregates; hence the soil structure is poor and changeable under various climatic and anthropogenic factors.

2.3. Climate

The climate is moderately warm and humid. Lithuania is characterized by mild winters with frequent thaws, relatively warm springs, moderately warm summers, and warm and wet autumns. The country experiences approximately 70 anticyclones and 100 cyclones annually. Anticyclones predominate in the winter and spring, while cyclones occur in the autumn and summer periods. Lithuania is located in the humid zone, where mean annual precipitation (748 mm) exceeds mean evapotranspiration (512 mm). The mean annual amount of precipitation is more than 800 mm and the average annual air temperature is 6.7 °C in the region of West Lithuania. This region is strongly affected by the maritime climate, because of which it receives the greatest annual amount of precipitation, averaging 923 mm over the last 40 years, compared with the other regions of the country. However, the average annual amount of precipitation has decreased by 7.9% over the past 40 years. The amount of rainfall that falls during the warm period (April-October) is also decreasing (17.9%). The amount of rainfall is particularly important during the spring period when plants start their vegetative/growing season. The variation of precipitation amount in recent years has shown a trend toward reduction in the spring period.

2.4. Experimental design

The investigations were performed at the two stationary liming and fertilizing field trials:

1. The effects of long-term liming on the topsoil properties were estimated using the following experimental design shown in **Table 1**.

2.5. Object of investigation

The naturally acid soil and the same soil exposed to different liming intensity were the objects of the investigation. This long-term field experiment was started in 1949. Applying the long-term liming system (primary, repeated, and periodical liming) in the period 1949–2005 formed the different soil pH levels (**Table 1**). Before liming with 0.5 rate every 7 years during the study period, the soil pH_{KCl} was 5.4–5.9, and in the soil limed with 2.0 rates every 3–4 years, it was 6.4–6.8. The study on the soil structure and organic carbon compounds was conducted in soils that significantly differed in pH.

2.6. Trial history

Pulverized limestone (92.5% of $CaCO_3$) was used for periodical liming on the background of primary and repeated liming with slaked lime. Minimal organic fertilizing (40 t ha^{-1} manure during a seven-course rotation) was undertaken with traditional tillage and intensive crop rotation: sugar beet, spring barley with undersown grasses, perennial grasses (for 2 years), winter wheat, vetch-oats mixture for grain, and pea-barley mixture for forage. In 2008, the long-term experiment was adjusted. In 2005–2013, the soil was not limed. The crop rotation was shortened to four courses: spring barley with undersown grasses, perennial grasses (for 2 years), winter wheat, and spring oilseed rape.

The mineral fertilizing in crop rotation was background ($N_{60}P_{60}K_{60}$), and the fertilizing with organic fertilizers was minimal (40 t ha^{-1} farmyard manure during crop rotation and traditional soil tillage).

Liming intensity	**Amount of $CaCO_3$ applied, t ha^{-1}**		**Total amount of $CaCO_3$ applied, t ha^{-1}**
	1949–1998	**1998–2005**	**1949–2005**
1. Unlimed (**pH_{KCl} 4.0–4.1**)	—	—	—
2. Periodical liming using ×**0.5** of the liming rate calculated based on the soil hydrolytic acidity (3.3 t ha^{-1} $CaCO_3$) **every 7 years** (**pH_{KCl} 5.4–5.9**)	18.1	—	18.1
3. Periodical liming using ×**1.0** of the liming rate calculated based on the soil hydrolytic acidity (15.0 t ha^{-1} $CaCO_3$) **every 3–4 years** (**pH_{KCl} 5.9–6.3**)	46.8	7.5	54.3
4. Periodical liming using ×**2.0** of the liming rate calculated based on the soil hydrolytic acidity (15.0 t ha^{-1} $CaCO_3$) **every 3–4 years** (pH_{KCl} **6.4–6.8**)	89.9	15.0	104.9

Table 1. Experimental design.

2. The effects of long-term liming in combination with organic fertilizers (manure and green fertilizers) on the topsoil properties were estimated using the following experimental design:

Factor A. Soil acidity (pH_{KCl}): (1) unlimed soil (pH_{KCl} pH 4.1–4.3); (2) limed soil (pH_{KCl} 5.8–6.0).

Factor B. Organic fertilizers: (1) without organic fertilizers (control treatment); (2) green manure or plant residues; (3) farmyard manure 40 t ha^{-1}; (4) green manure (on a background of 40 t ha^{-1} farmyard manure (bkgd of FYM 40)); (5) farmyard manure 60 t ha^{-1}; and (6) green manure (on a background of 60 t ha^{-1} farmyard manure (bkgd of FYM 60)).

In a long-term experimental trial of farmyard manure rates starting from 1959 to 2005, 80 and 120 t ha^{-1} of farmyard manure were incorporated in two applications divided into equal parts for the seven-course crop rotation (for winter wheat and fodder beet). After the reconstruction of the trial in 2005, 40 and 60 t ha^{-1} of farmyard manure were incorporated in a single application (for winter wheat) in the five-course crop whereas in the fourth and sixth treatments, manure was not applied. Solid cattle manure was used, containing 14.53% of dry matter, 17.83% of organic matter, 0.42% of total nitrogen N, 0.27% P_2O_5, 0.67% K_2O, 2668 mg kg^{-1} Ca, 692 mg kg^{-1} Mg, pH_{KCl} 8.5.

The following alternative organic fertilizers were employed: in 2010, the aftermath of swards was disked in at 15 cm and plowed in at 20 cm depth. In 2011, after wheat harvesting, the straw was chopped, incorporated at 15 cm and plowed in at 20 cm depth. In 2012, the green mass of lupine and oats was disked in at 15 cm and plowed in at 20 cm depth after lupine pods had reached milk maturity. In 2013, after rape harvesting, the stubble and straw were chopped and incorporated by a cultivator at 15 cm and plowed in at 20 cm depth.

On limed background, in 2010, liming was applied repeatedly using pulverized limestone at one rate according to the hydrolytic acidity. All treatments were equally fertilized with mineral fertilizers (background fertilization). Fertilizer $N_{60}P_{60}K_{60}$ have been applied for winter wheat and spring barley stands, $N_{30}P_{60}K_{60}$ for lupine-oats mixture, and $N_{60}P_{90}K_{120}$ for winter rape stands. Fungicides and insecticides were used in case of necessity; herbicides were not used at all. Conventional soil tillage was applied. The acidic soil had been periodically limed and manured for a period of 47 years. During the period of the study, the soil received: 38.7–36.5 t ha^{-1} $CaCO_3$; 840 t ha^{-1} cattle manure, 2740 kg ha^{-1} mineral nitrogen; 3030 kg ha^{-1} phosphorus, 3810 kg ha^{-1} potassium.

3. Management of soil acidity

Acidification of soil is a continuous naturally occurring process in soil formation. It is promoted by natural and anthropogenic factors. Soil acidification management involves both neutralization of soil acidity and regulation of the acidification of limed soil. The pH level in agricultural soils is strongly influenced by the fertilizers and pesticides applied, crops grown and their sequence in a crop rotation, as well as by tillage intensity. Liming is the most efficient measure used to neutralize soil acidity. Liming materials alter the mobility of some biogenic elements and their buildup in the soil. Application of lime materials and organic substances could be an effective measure to improve the chemical properties of moraine loam soil, but the changes in their values depend on the amount of liming materials and frequency of application. Studies conducted in Lithuania have shown that the effect of initial liming lasts for over 30 years. The published data indicate that the change in soil acidity indicators after liming primarily depends on whether the liming is performed for the first time or repeatedly. Within a period of 50 years after primary liming, the pH values return to the initial ones and practically do not differ from those of acid soils; however, it was noticed that the soil limed once even with the lowest 0.5 rate (according to hydrolytic acidity) tended to acidify more rapidly compared with the soil that had never been limed. This suggests that once the chemical balance of the soil has been disturbed, it becomes less resistant to environmental impacts than a naturally acid soil. Therefore, from the soil conservation viewpoint, liming should be an uninterrupted and systematic process in the agroecosystem. The results of such systematic (primary, repeated, and periodical) liming are significantly changed pH values in the upper and deeper soil horizons and alterations in the mobile Al contents (**Figures 1** and **2**).

Regular liming (during a 35-year period) resulted in a decrease in the hydrolytic acidity and mobile aluminum in a loam soil and an increase in the pH in the soil profile up to 100 cm depth. Periodical liming every 7 years at a rate of 0.5 by hydrolytic soil acidity (3.8 t ha^{-1} $CaCO_3$) allowed maintaining the soil at a medium acidity level (pH_{KCl} 4.8–5.1); when the soil was intensively limed at a rate of 1.0 every 3–4 years, mobile aluminum was abolished and pH_{KCl} in the upper layers of the soil reached close to neutral, 6.4–6.7. Long-term regular soil liming changes the acidity not only in the topsoil, but, because of the migration of calcium and magnesium cations, the chemical properties in the subsoil horizons change as well. When liming is intensified to 2.0 rates every 3–4 years, the pH_{KCl} value in the topsoil reaches 6.9–7.2, i.e., the acid soil becomes neutral and acidification is hindered in the ElB horizon up to 40 cm depth (**Figure 2**).

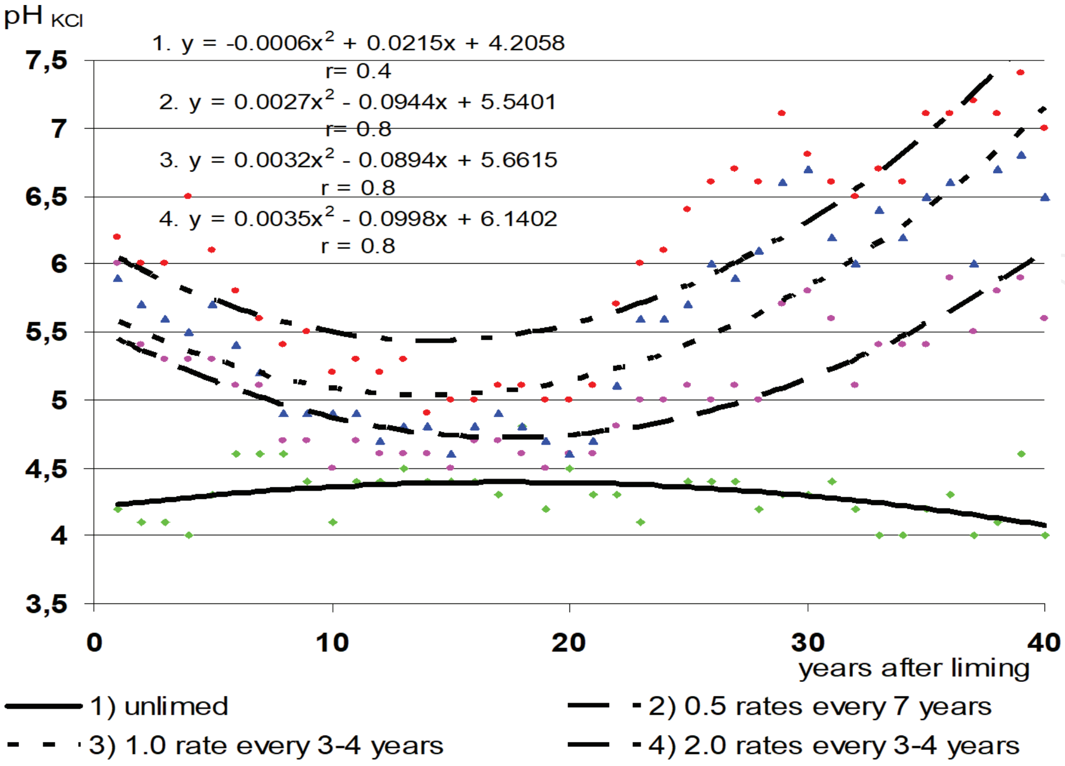

Figure 1. Long-term liming effect on topsoil pH changes.

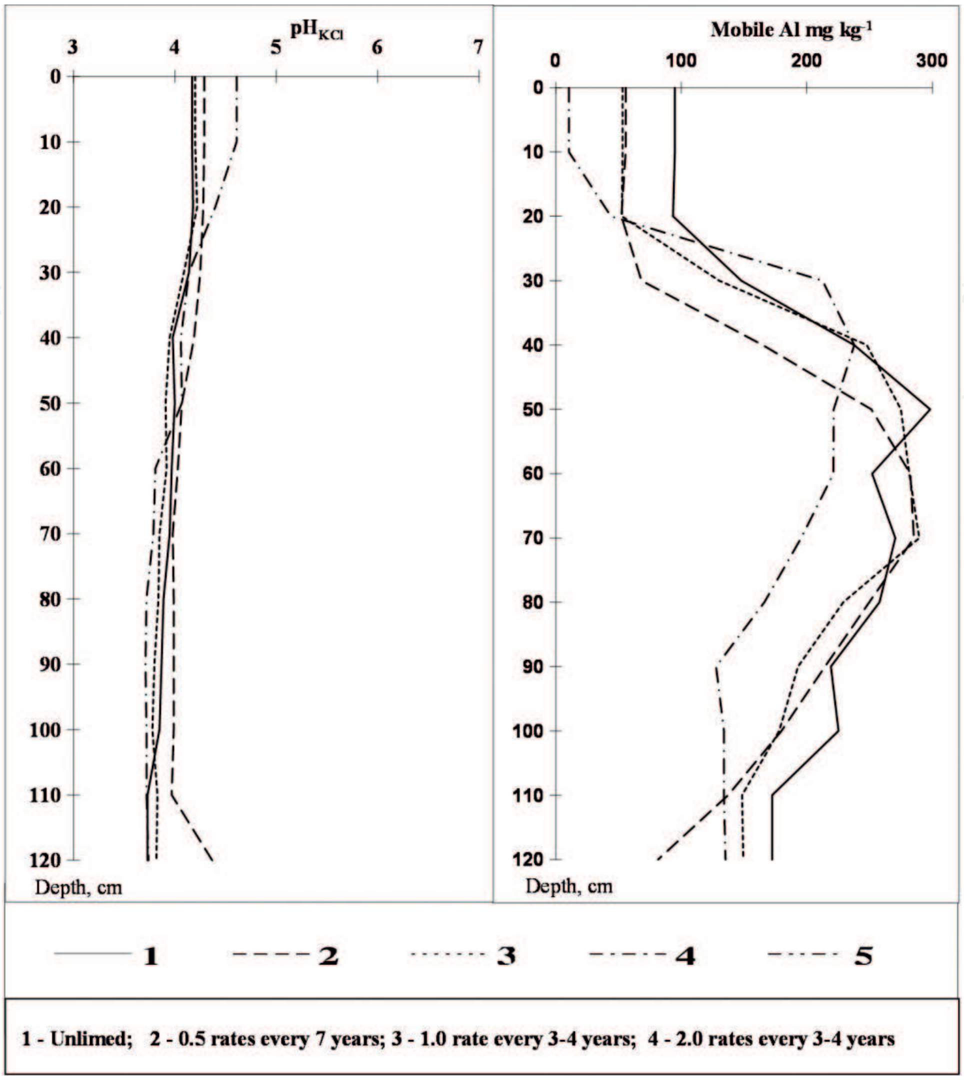

Figure 2. Long-term liming effect on pH and mobile Al changes in soil profile.

Long-term intensive liming causes significant morphological changes in the soil profile, and increases the area of podzolic veins in the profile wall. Ozeraitiene has documented that the highest leaching of calcium occurs and soil structure deteriorates in intensively limed soil [8]. This shows that it is impossible to reach optimal soil conditions for plant growth by using intensive liming only. It is necessary to combine liming with organic fertilizing. A number of different mechanisms have been proposed to explain the positive effect of organic residues, and their decomposition products in raising soil pH and/or complexing phytotoxic Al and thus improving the fertility of acid soil [11–14].

According to Mokolobate and Haynes, the addition of organic residues to acid soils is potentially a practicable low-input strategy for increasing soil pH, decreasing concentration of phytotoxic Al, and reducing lime requirements [14]. This statement was based on our data obtained in naturally acid soil with incorporation of farmyard manure. A systematic fertilization of naturally very acidic soils with farmyard manure at a 60 t ha^{-1} rate every 3–4 years for over five decades resulted in decrease in mobile Al by 2.3 times, i.e., from 117.0 to 50.5 mg kg^{-1} and in an increase in the pH in the topsoil by 0.2 units (**Figure 3**).

The soil acidity neutralizing effect of solid manure showed up only after 20 years of its application, but its significant effect manifested itself the next year after application, when pH_{KCl} values reached 4.8–5.3 and mobile Al content decreased to <50 mg kg^{-1} (**Table 2**).

A combination of farmyard manure application (60 t ha^{-1} every 3–4 years) with periodical liming (1.0 rate of $CaCO_3$ according to hydrolytic soil acidity every 5–7 years) stabilizes the acidification process (a pH of 5.6–7.1 is maintained) in the arable layer. The findings of the long-term experiments indicate that liming alone was less efficient for soil acidity indicators than its combination with farmyard manure. The obtained results substantiate the research evidence reported by Teit that liming materials and farmyard manure are agronomic practices that cannot replace each other but can complement each other [15].

Liming materials and their combinations with farmyard manure (FYM) reduce the amount of mobile aluminum to a level that is non-toxic to plants (1.17–1.70 mg kg^{-1}). An analysis of the long-term data revealed that the amount of mobile Al after liming and FYM fertilizing decreased to 0.0–0.3 mg kg^{-1}, while after 6–7 years, an increase to 1.3–6.0 mg kg^{-1} was determined.

The neutralization of soil acidification in the topsoil and subsoil up to 60 cm depth was achieved by periodical liming with 1.0 rate every 7 years in combination with the application

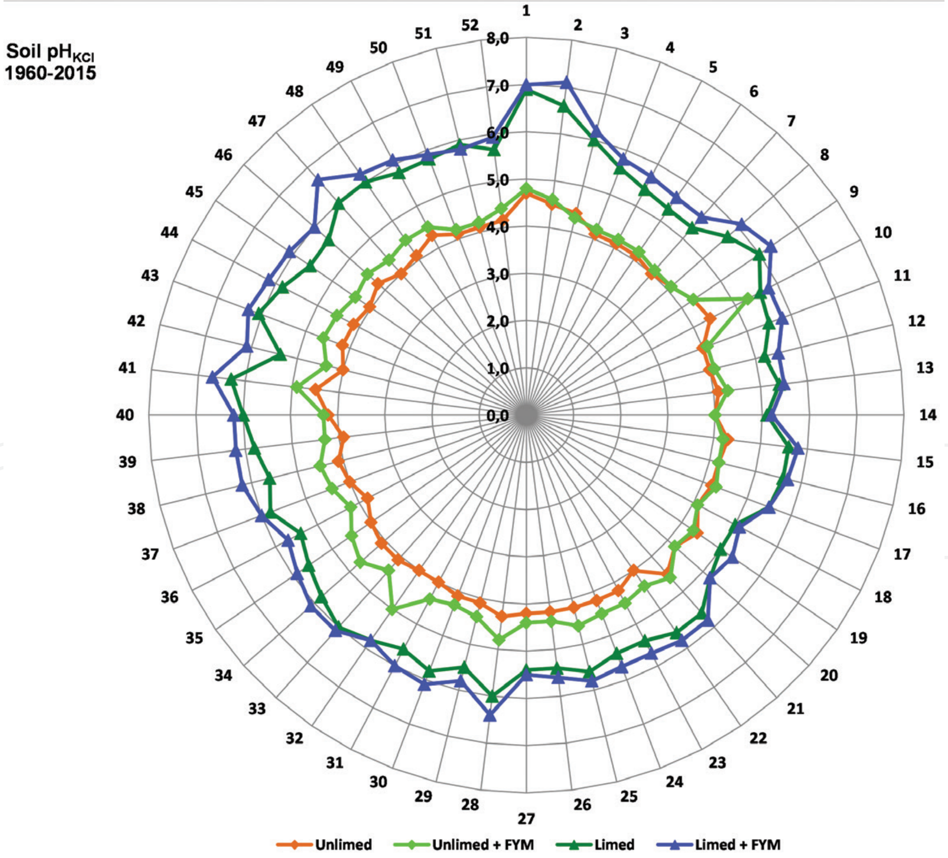

Figure 3. Long-term liming and manuring effect on topsoil pH changes.

Treatment.	Arithmetic mean $\bar{x}$	Standard error of the mean $S\bar{x}$	Minimal value of indicators Min.	Maximal value of indicators Max.	Standard deviation SD	Coefficient of variation V%
pH (units)						
Unlimed	4.16	0.02	3.80	4.70	0.17	4.06
Unlimed + FYM	4.40	0.03	4.00	5.30	0.25	5.62
Limed	5.66	0.05	5.00	6.90	0.36	6.34
Limed + FYM	5.93	0.06	5.10	7.10	0.41	6.87
Mobile Al (mg kg^{-1})						
Unlimed	117.0	2.84	63.90	169.7	20.28	17.34
Unlimed + FYM	50.5	4.33	13.10	107.5	30.90	61.20
Limed	1.70	0.22	0.00	7.60	1.55	91.41
Limed + FYM	1.17	0.18	0.00	6.0	1.25	107.1

Table 2. Results of statistical analysis of agrochemical (pH and mobile Al) indicators.

of 60 t ha^{-1} manure every 3–4 years in the seven-course crop rotation system (**Figure 4**). The effects of long-term (1959–2005) liming in combination with cattle manure application on soil pH and mobile aluminum were investigated in the whole soil profile up to a 100 cm depth. Acid soil had been periodically limed and manured at different intensities for 47 years. During the entire period of the study, the soil received 38.7–36.5 t ha^{-1} $CaCO_3$; 840 t ha^{-1} cattle manure; 2740 kg ha^{-1} mineral nitrogen; 3030 kg ha^{-1} phosphorus; 3810 kg ha^{-1} potassium. The findings suggest that long-term (47 years) periodic liming in combination with the application of cattle manure significantly altered the chemical properties within the entire soil profile.

In the topsoil and subsoil, up to 60 cm depth, the soil acidity was neutralized by systematic liming at 1.0 rate every 7 years in combination with the application of 60 t ha^{-1} manure every 3–4 years. Long-term periodical liming in combination with manuring improved the chemical properties of acid soil profile in the ElB and ElBt horizons.

The rate of acid soil neutralization depends on the particle size of the liming materials. According to the size, liming materials can be classified as powdered, granulated, pelletized, etc. Powdered liming materials show the most rapid action; however, their application is rather complicated because special machinery is required in order to spread them evenly. Granulated liming materials react longer with soil and the duration of their action is longer than that of powdered ones [16]. In Lithuania, acid soils for more than three decades (1964–1994) were systematically limed every 5–7 years with conventional liming material—powdered limestone [17].

Recently, granulated liming materials have been increasingly used in European, including Lithuania, and other countries for maintenance liming of soils [18, 19].

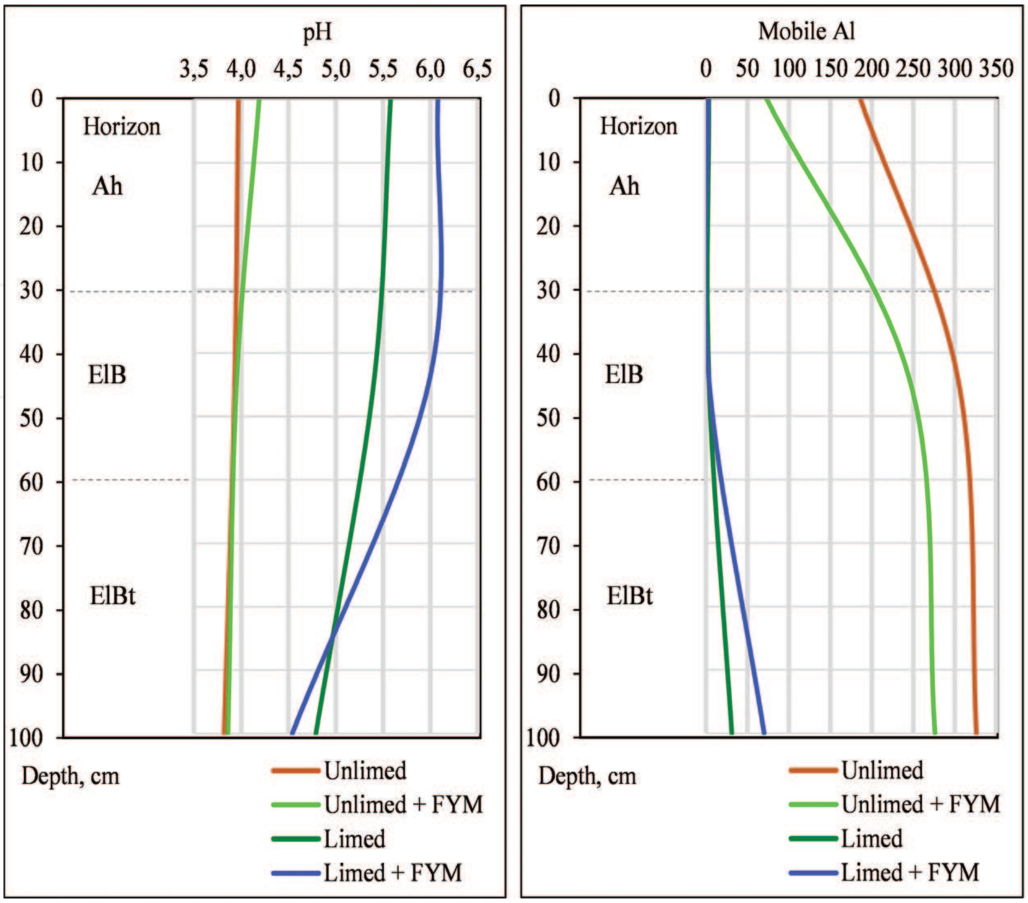

Figure 4. Long-term liming and manuring effect on pH and mobile Al changes in soil profile.

4. Soil pH and microbial prevalence

The provision of energy to the microbial community by root exudates, dead roots, and intense microbial metabolic activity benefits mineralization capacity, improved soil structure, and plant growth conditions. Mineral nutrients, organic matter (including fresh plant material), and growth factors exist within appropriate temperature, moisture, redox potential and pH ranges for microbial growth [20–22].

The majority of soil systems tend to fall in the soil pH range fall below 5.5 in the agroecosystem. Yet, acidic soils constitute a major portion of the world soil resources. West Lithuania also falls in the region of acidic soils. Acid conditions present a stressful environment not only for plants but also for the microbial community. In West Lithuania, albeluvisol (Bathygleyic Dystric Glossic Retisol (WRB 2014) has been investigated for nearly six decades. Soil liming and nutrition with various organic fertilizers: incorporation of plants residues (treatments 2, 4, 6) and 40 t ha^{-1} (treatments 3, 4), or 60 t ha^{-1} (treatments 5, 6) of farmyard manure created different conditions for the functioning of soil microorganisms and carbon sequestration in the soil. The experimental findings suggest that a significant increase in the pH value was achieved due to liming, and the concentration of C significantly increased when farmyard manure together with the incorporation of plant residues had been used for crop nutrition. During the experimental period, soil chemical indicators changed and microorganism communities, which determined the biochemical processes in the soil, formed. The following processes were analyzed: greenhouse gas release—CO_2 emission from the soil—and groups of microorganisms participating in the organic matter decomposition processes and being able to assimilate mineral nitrogen [23]. The CO_2 emission from the soil is also an indicator of the general biological activity of soil microorganisms. Our study showed that with a significant reduction in soil acidity resulting from liming, a significant increase in CO_2 emission occurred. An increase in the pH value (factor A) by an average 1.62 units significantly increased the content of ammonifying, mineral nitrogen assimilating, and spore forming bacteria and was essential in reducing the number of fungi, because more compounds are formed in the soil, which are available to bacterial microflora (**Table 3**). According to the averaged results, increasing concentration of carbon (factor B) in the soil enhanced CO_2 emission; however, a more detailed analysis revealed that in acid soil, CO_2 emission increased in all organic fertilization cases, but when soil acidity decreased, organic fertilization was no longer an effective means. As a result of uneven fertilization, higher concentration of carbon accumulated in the soil and a significant increase was observed in the abundance of ammonifying and mineral nitrogen assimilating microorganism populations, indicating more favorable conditions for the functioning of the microbial community in the soil. The same regularity was established for sporogenes bacteria, which are also very active in the organic matter mineralization processes. However, the abundance of fungi populations was substantially higher only with the integrated incorporation of manure and plant residues in the soil.

Such complex fertilization introduces a larger amount of more complex compounds into the soil, which can be mineralized by the complex enzyme system of fungi.

Symbiotic biological nitrogen fixation is one of the processes that describe the ecological balance of soils. Acid soils are characterized by poor diversity and activity of microorganisms [24]. None or insignificant amounts of *Azotobacter chroococcum* and *Trichoderma lignorum* are found. Nitrification process, which ensures the availability of nitrogen compounds to plants and other microorganisms, reaches the highest intensity when soil acidity was diminished to pH 6.7. Long-term studies indicate that the optimal pH indicator value for various physiological groups of microorganisms is different: for ammonifying and sporogenes bacteria, it is 6.2; for streptomycetes bacteria, 6.7; and for mineral nitrogen assimilating bacteria and micromycetes, it is 6.0 [25].

Soil reaction is one of the most important factors influencing legumes and *Rhizobium* symbiosis. Greater concentration of H^+ ion increases the solubility of Al, Mn, and Fe; these elements may become toxic to plants. However, it is known that *Rhizobium leguminosarum bv. trifolii* can survive in very acidic soils due to low acidity microzone formation; hence, rhizobia can be isolated from these soils [26]. All living organisms require nitrogen to make proteins, enzymes, and other cellular components. Seventy-eight percent of the atmosphere contains nitrogen that is required by all living organisms, but the gaseous form is unavailable for most organisms. Those that can take up nitrogen gas, do so through a process of nitrogen

Treatment	**Soil pH and carbon (%) variation in soil (mean indices)**	**CO_2 emission, mg g^{-1} abs. dry soil day^{-1}**	**Ammonifying bacteria, CFU × 10^6 g^{-1} abs. dry soil**	**Nitrogen assimilating bacteria, CFU × 10^6 g^{-1} abs. dry soil**	**Micromy-cetes, CFU × 10^4 g^{-1} abs. dry soil**	**Sporogenes bacteria, CFU × 10^4 g^{-1} abs. dry soil**
Soil pH, liming (factor A)						
Unlimed soil (pH_{KCl} 4.1–4.3)	4.24–4.53 (4.35)	0.0271	5.9	4.6	4.3	3.1
Limed soil (pH_{KCl} 5.8–6.0)	5.87–6.15 (5.97**)	0.0288*	7.9*	7.7*	2.7*	7.6*
$LSD_{0.05}$		0.00095	0.47	0.268	0.107	0.23
C_{org}, organic fertilizers (factor B)						
Without organic fertilizers	1.328–1.459 (1.41)	0.0272	5.8	4.8	3.5	3.9
Green manure or plant residues	1.408–1.528 (1.47)	0.0273	6.8	4.8	3.5	3.8
Farmyard manure 40 t ha^{-1}	1.475–1.657 (1.58**)	0.0298	7.1*	6.8*	3.5	6.4*
Green manure (bkgd of FM 40)	1.493–1.718 (1.62**)	0.0273	7.1*	6.9*	3.2	5.4*
Farmyard manure 60 t ha^{-1}	1.478–1.649 (1.59**)	0.0277	7.4*	6.9*	3.6	6.8*
Green manure (bkgd of FM 60)	1.382–1.540 (1.48)	0.0284	7.2*	6.8*	4.1*	6.1*
$LSD_{0.05}$		0.00213	1.057	0.599	0.239	0.514

*Significant at $P \leq 0.05$.

**Significant at $P \leq 0.01$.

CFU, colony forming unit.

Table 3. The effect of different soil pH (liming) and supply of carbon (organic fertilization) on microbiological indicators.

fixation. Many species can fix carbon though photosynthesis (all green plants, algae, some bacteria) but only few organisms can fix nitrogen. Nitrogen fixing organisms can be free-living or symbiotic. Symbiotic associations developed after the evolution of terrestrial green plants. Free-living nitrogen fixers are among the most ancient organisms. Symbiotic associations between species of nitrogen-fixing bacteria and green plants result in considerable amounts of nitrogen being fixed. Among the best studied of these symbiotic associations are those among various species of *Rhizobium* and other species of legumes. These associations are very specific. Phylogenetic analysis of the rhizobia based on the 16S rRNA subunit places the species into three genera: *Rhizobium*, *Bradyrhizobium*, and *Azorhizobium*. Metagenomic analysis of differently fertilized and tilled Bathygleyic Distric Glossic Retisols suggest that Rhizobium accounts for 0.3–0.4%, i.e., 39,358–64,767 operation taxonomy units (OUT) of the 131,194–161,917 identified OUT. Research on symbiotic nitrogen fixation has been carried out for 50 years in West Lithuania. Experimental results obtained by various researchers were comprehensively presented in various scientific publications and summarized by Lapinskas [27].

Symbiotic nitrogen fixation depends on the occurrence and survival of rhizobia in the soil and also on their efficiency. Soil reaction is one of the most important factors that influence legume and *Rhizobium* symbiosis. The data of long-term research evidenced that additional inoculation with rhizobia bacteria resulted in 6–16% increase in dry matter yield, and for some species, for example, goat's rue and soy, the dry matter yield increase amounted to 119–165% [28]. Such differences are primarily determined by the occurrence of the specific rhizobia and the activity of their enzymes in the soil. This is especially relevant in soils, which, due to the intensive use of mineral fertilizers, acidify, thus contributing to the survival and proliferation of atmospheric nitrogen fixing bacteria. *Sinorhizobium meliloti* and *Rhizobium galegae* bacteria, which form symbiosis with certain plants, for example, lucerne and Caucasian goat's rue, are highly sensitive to acid soil and soluble Al. The acidification inhibits the root-hair infection process and nodulation. The prevalence of the main species of rhizobia (*Rhizobium leguminosarum bv. trifolii* and *Rhizobium bv. viciae*, *Sinorhizobium meliloti* and *Rhizobium galegae*) was established in 400 different soil samples in Lithuania [28]. A dilution method was used for legume inoculation in sterile conditions (**Figure 5**). Estimation of the rhizobia population abundancy suggested that the range of the pH value is not the same for different bacteria: in very acid soil (pH 4.1–4.5), *Sinorhizobium meliloti* was not detected, while *Rhizobium galegae* bacteria were detected only when the soil pH value ranged from 5.1 to 5.5. Such low pH was best tolerated by *Rh. leg. bv. trifolii* (14.2×10^3) and *Rh leg. bv. viciae*, but even their populations were not rich. The optimal pH range for *Rh. leg. bv trifolii* was 5.6–7.0, for *Rh leg. bv viciae* 6.1–7.0. *Sinorhizobium meliloti* was the most numerous at a pH range of 6.6–7.0 and only *Rhizobium galegae* were most abundant at pH > 7.0. It is known that the symbiotic efficiency of different legumes and *Rhizobium* bacteria strains depends on the environmental conditions, mineral nitrogen concentration, and soil pH changes, which essentially determine the activity of nitrogenase enzyme [29]. The activity of nitrogenase was best investigated in the red clover association (**Figure 6**). The gas chromatography assay, used to determine the activity of nitrogenase enzyme, responsible for the biological nitrogen fixation in the red clover and *Rh. leg. bv. trifolii* association suggested that in an acid soil, the change of the pH value from <4.75 to 6.0 was insignificant for nitrogenase activity, but an increase in the pH value above 6.25 resulted in a nearly 3-fold increase in nitrogenase activity. During the summer period, in the same soil, even at a soil pH < 4.75, nitrogenase fixed biological nitrogen 10 times more intensively compared with the autumn period. As the pH value increased to 5.25, nitrogenase

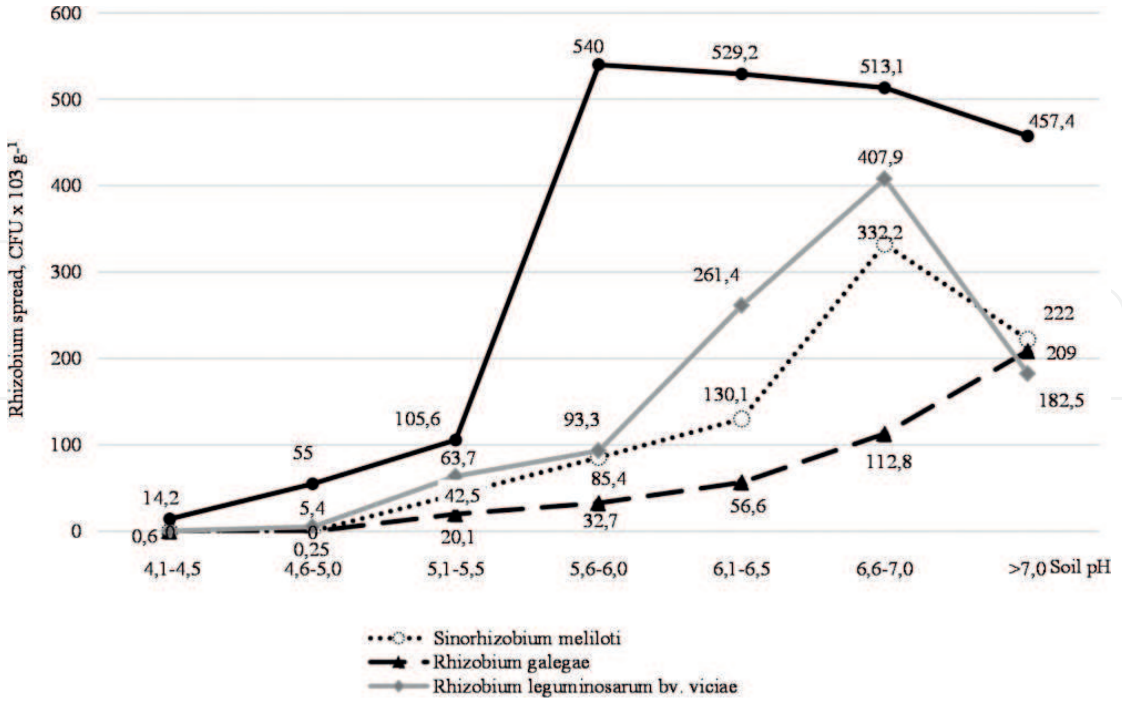

Figure 5. The impact of the soil pH on *Rhizobium* spread (×10^3 CFU g^{-1}).

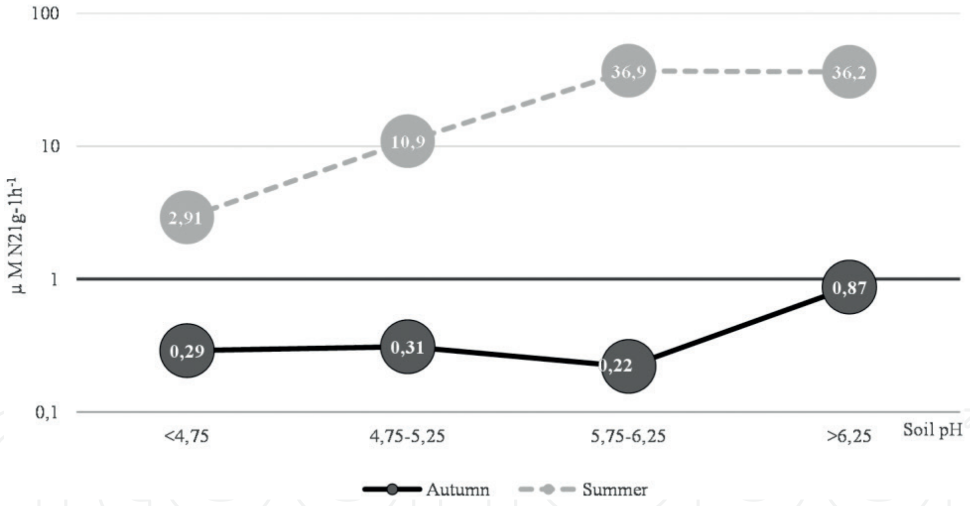

Figure 6. The activity of nitrogenase as influenced by soil pH and season.

activity was 10.9 μ M $N_2$1 g^{-1} h^{-1} and was 35 times higher than in autumn. With the change of the pH value from 5.75 to >6.25, nitrogenase activity increased more than 40-fold. It was noted that soil acidity was a more stressful factor for nitrogenase activity in autumn than in summer.

This determination of the tolerance of different rhizobia species for low pH is an opportunity to use biological preparations more efficiently and to select plants that are resistant to soil with a lower pH [27]. Biological fertilizers have recently become a very popular product used to improve the vital functions of soils. This is especially relevant in ecologically sensitive agro-climatic regions, like West Lithuania, which receive a lot of rainfall and where naturally acid soils predominate. Application of various biological measures, including additional incorporation of humic and amino acids, trace nutrients, and various species of microorganisms that supplement the complex of soil microorganisms, is a common practice [29, 30].

5. Soil pH effect on organic carbon and water stable aggregate content

Soil organic matter is the chief indicator of soil quality and ecological stability. Consequently, carbon accumulation in stable forms not only supports and enhances the organic matter content in the soil, but also exerts a positive impact on the soil quality and the entire ecosystem [31]. Taking into account the environmental conditions and land use, soil can be the source of both carbon accumulation and release. The directions of these processes depend on the intensity of the anthropogenic load present in the specific natural conditions. Soil liming and organic fertilization are one of the most common ways to improve carbon sequestration in the soil [32, 33]. Negative statistically significant effect of periodical liming at 0.5 and 2.0 rates on SOC (soil organic carbon) content was determined (**Figure 7**). In the unlimed treatment, the SOC content was 1.45%, while in the periodically limed treatment at 2.0 rates every 3–4 years, it was by 0.18% lower. A positive statistically significant effect of fertilization on soil organic carbon content in the soil was established. The content of SOC was 1.47% in the non-fertilized treatment and in the fertilized treatments, it varied from 1.59 to 1.91%. The highest amount of soil organic carbon (1.91%) was obtained in the limed soil applied with FYM. A similar content of SOC was determined after incorporation of alternative organic fertilizers (wheat straw, oilseed rape residues, roots, stubble, and perennial grasses). However, in contrast to incorporated farmyard manure, the largest part of alternative organic fertilizers is composed of nitrogen compounds that are rapidly mineralized in the soil.

Decomposition of FYM includes longer phases of mineralization and nitrogen immobilization, thus increasing the amount of stable organic compounds and accumulation of humus in comparison with the use of plant residues. SOC and dissolved organic carbon (DOC) are important indices for soil organic matter (SOM). Dissolved organic carbon (DOC) is a key component of the active SOM pool and serves as a source and sink of soil nutrients and/or as an ecological marker to understand soil fertility. Soil liming with or without fertilization could affect the content of DOC, leading to alterations in the formation of complexes between organic ligands and metals, and SOM sequestration. On the other hand, increased decomposition of stabilized material induced by addition of fresh organic material triggering microbial activity can result in higher C losses from soil. These indices are influenced by agricultural practices [34, 35]. The content of DOC is closely related to soil- and management-associated factors. Liming at different intensities did not show any statistically significant effect on the accumulation of humic acids in the soil. However, intensive soil liming (at 2.0 liming rate) tended to increase the content of humic acids (**Figure 8**). Fulvic acids predominated in unlimed soil. This treatment was found to have the highest amount of fulvic acids (0.562% of C). It could be associated with a slow humification processes in the soil, due to the

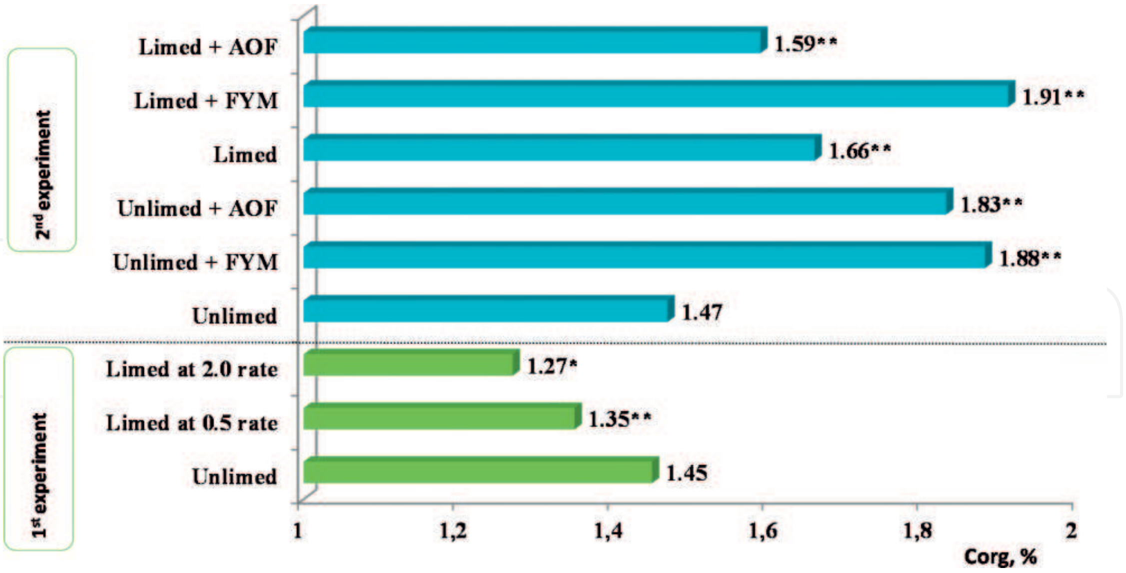

Figure 7. Effect of soil liming and fertilization on the amount of organic carbon in the topsoil, 2011–2013 average data. *Note:* * and ** Significantly different from control (unlimed) ($P < 0.005$) and ($P < 0.001$); FYM, farmyard manure; AOF, alternative organic fertilizers.

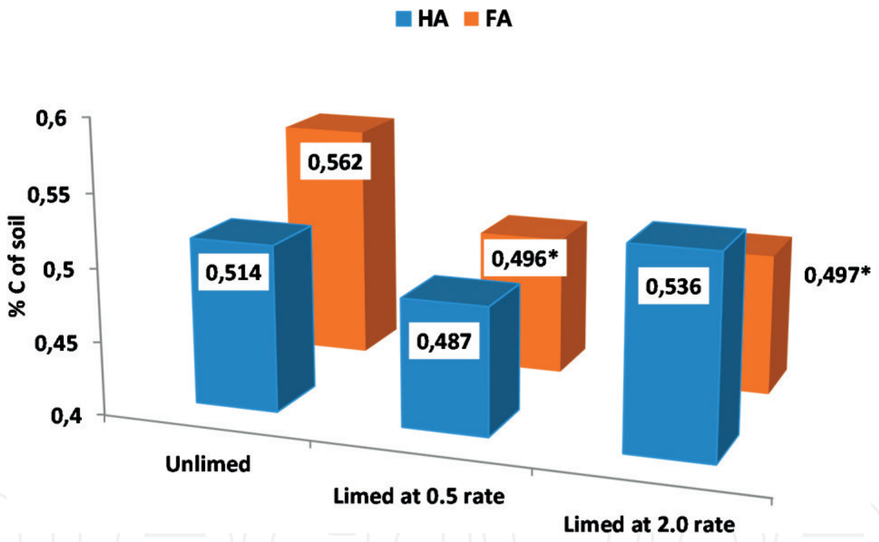

Figure 8. Liming effect on the total content of humic and fulvic acids in soil, % C (where HR, humic acids; FR, fulvic acids). *Note*: * Significantly different from control ($P < 0.005$).

low carbon content, when the content of fulvic acids is always considerably higher compared with humic acids in the soil. Soil liming significantly decreased the amount of fulvic acids. Soil amendment with organic fertilizers significantly increased the amount of humic acids, which indicates an increase in organic matter fixation and its stability in the soil. Seeking to assess the relationship between labile and stable humic acid amount and soil pH, we conducted a correlation and regression analysis. Mobile humic acid fraction (HR1) was assigned as labile humic acids, while humic acids fractions bound with calcium (HR2) and soil minerals (HR3) were stable fractions. It was determined that increasing soil pH decreased labile and increased stable humic acid amounts in the soil (**Figure 9**).

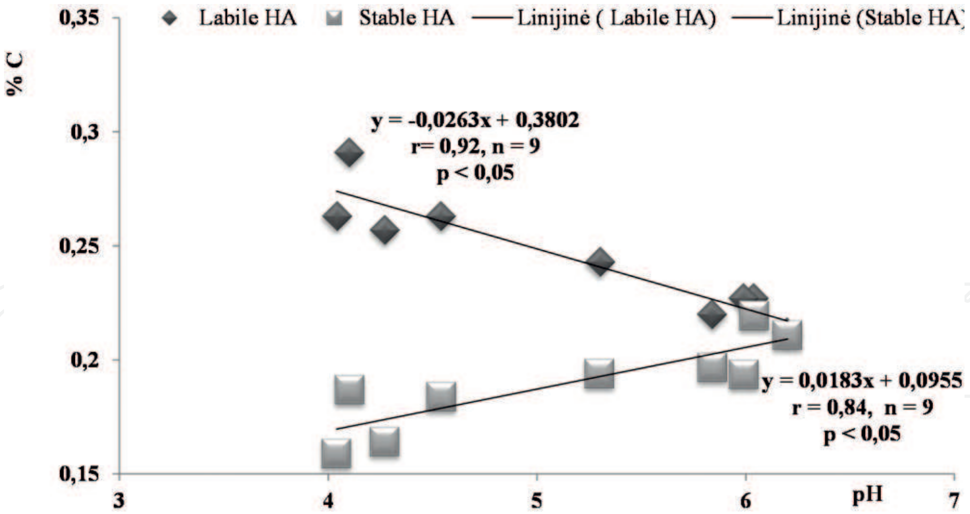

Figure 9. The relationship between labile and stable humic acids amount and soil pH.

Soil aggregates stability is a crucial soil property affecting soil sustainability. Physical protection of soil organic matter by aggregates is considered to be an important mechanism for soil carbon stabilization. Due to the small amount of water-stable aggregates, the structure of acid moraine loam soil is poor and depends on various climatic and anthropogenic factors. The findings of our study evidenced that long-term (for 56 years) systematic liming by 0.5 rate every 7 years and 2.0 rates every 3–4 years on the background of minimal organic fertilizing and traditional tillage affected the structure of moraine loam soil (**Table 4**). According to the data obtained over a period of 11 years, the largest amount (68.4–72.1%) of water stable aggregates (>0.25 mm) was present in the intensively limed soil under perennial grasses and winter wheat in 2002–2003.

A 28–34% increase in the content of water stable aggregates was recorded in limed soil compared with unlimed soil. These data suggest that perennial grasses grown in limed soil can effectively improve the structure of moraine loam soil. During the study period, a decrease in water stable aggregates was determined in both acid (from 57 to 25%) and limed (from 72 to 29%) soil. These findings indicate that the climatic conditions (swelling of clay under warm and wet conditions in autumn–winter periods and drying and rewetting cycles in spring-summer periods) have a significant negative impact on the formation of the aggregates. The negative effect of these weather conditions on soil aggregates are observed by other researches [36]. Also, the decrease of water stable aggregates in moraine loam soil is caused by the intensive anthropogenic activity inducing a decrease of organic carbon. Any agricultural activity which increases organic matter content in the soil has a direct effect on the increase of soil aggregate stability. The data of the soil structure in the topsoil and subsoil showed that long-term manuring had a positive effect on water stability of soil aggregates (**Figure 10**).

Under the influence of long-term application of farmyard manure, the content of water stable aggregates >0.25 mm increased by 48.1% and amounted to 52.3%, compared with the treatment with no manure. An 81.6% increase in the content of aggregates was determined in soil that had been fertilized with manure for a long time and optimal pH had been maintained by periodic liming.

Treatment	Investigation year and vegetation type										
	1996 W	1997 V + O	1998 P + B	2002 G	2003 W	2004 P + B	2005 V + O	2008 B	2009 G	2010 T	2011 R
Unlimed	57.3	50.4	56.5	56.2	50.9	40.1	42.0	34.2	37.7	26.0	24.8
0.5 rate every 7 years	51.5	48.9	53.4	64.7	63.8*	53.5*	51.4*	47.1*	43.8	32.0	29.8
2.0 rate every 3–4 years	56.0	55.9*	58.4	72.1*	68.4*	50.5*	50.6	42.4	53.2*	30.5	28.8

*Significantly different from control ($P < 0.005$).

W, winter wheat; V + O, mixture of vetch and oat; P + B, mixture of peas and barley; G, perennial grasses; B, spring barley; T, winter triticale; R, spring rape.

Table 4. The effect of periodical liming on the content (%) of water stable aggregates (>0.25 mm).

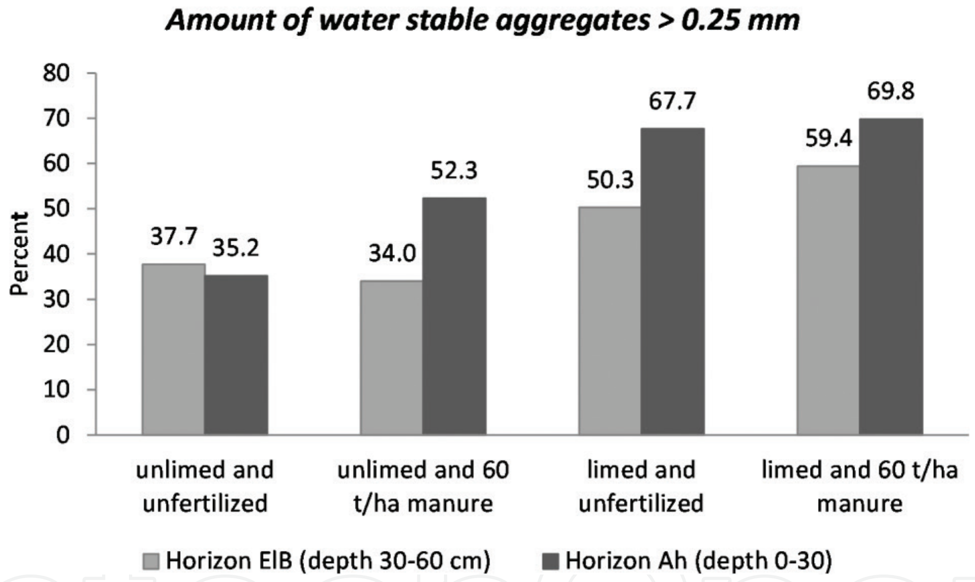

Figure 10. Effects of long-term liming, manuring, and their combination on the change of water stable aggregates in the soil profile.

The content of water stable aggregates both in the naturally acid and in limed soil was 30.3% higher in the upper Ah horizon compared with the deeper ElBt (30–60 cm) layer. This is related to the impact of plant roots present in the upper topsoil layer and higher organic matter content, which are relevant factors for the formation of water stable aggregates. Liming and its combination with the application of manure had a positive influence on the formation of water stable aggregates in the deeper 30–60 cm soil layer as well. In the limed and the limed and manure-applied ElBt soil horizon, the content of water stable aggregates was 33.5 and 57.2% respectively higher than in the unlimed and unfertilized soil. This was associated with a slight increase in calcium ions and organic carbon resulting from the long-term application of crop and soil management practices. The highest content of water stable aggregates (59.4%) was established in the limed and manure-applied soil. Manure application alone did not have any impact on the formation of water stable aggregates in this soil layer.

6. Soil pH effect on crop productivity and weediness

Liming of acid soils, particularly periodical liming and FYM fertilization, is the most important amelioration means of Dystric Albeluvisols, which significantly changes their ecological state. Long-term research enables assessment of the effects of various agronomic practices on the productivity of various agricultural crops. Soil acidity limits the yield of many agricultural crops. A low amount of base cations, particularly toxicity of calcium and aluminum, affect root growth and plant water and nutrient uptake; crop yields most often decline due to acid soils [37]. In the field experiment, winter wheat was grown after perennial grasses. According to the experimental design, before wheat sowing, individual plots were fertilized with solid cattle manure at rates 40 t ha^{-1} and 60 t ha^{-1}. The phytometric indicators that form wheat productivity (ear length and number of grains per ear and 1000 grain weight) were significantly higher in the limed soil and this had a direct effect on grain yield (**Table 5**). In limed soil, winter wheat grain yield was 1.8 times higher and that of straw 1.3 times higher than in unlimed soil. The highest winter wheat grain yield, 3.45 t ha^{-1}, was produced in soil fertilized with 60 t ha^{-1} FYM.

Lupine performs best in acid soils (pH 4.5) and worst in neutral or alkaline soils. It is susceptible to too high concentration of calcium ions, particularly at the beginning of growth. Oats are moderately susceptible to soil acidity. They perform best at soil pH 5.5–6.0 and satisfactorily at pH 4.5. Lupine was grown after winter wheat. Both in unlimed and limed soil, the total number of emerged plants were very similar 156–157 ha^{-1}. The highest dry matter yields of the lupine-oats mixture (9.60-10.0 t ha^{-1}) were recorded in soil fertilized with 60 t ha^{-1} FYM and green manure (bkgd of FYM 40).

Like winter wheat, spring barley is also very susceptible to soil acidity, particularly to mobile aluminum. In limed soil, the values of yield-forming indicators (number of productive and unproductive stems, number of grains per ear, and 1000 grain weight) were significantly higher than in unlimed soil. In limed soil, barley grain yield was 2.2 times higher than in acid soil. The highest yields (3.93–4.00 t ha^{-1}) were produced in soil fertilized with 60 t ha^{-1} FYM and green manure (bkgd of FYM 40). Similar trends were identified for straw yield.

Research showed that in a naturally acid soil (pH 4.1–4.3), in the crop of winter wheat before harvesting, the number of weeds was higher by 79.8%, in lupine–oats mixture by 35.7%, in spring barley crop by 29.1% compared with limed soil (pH 5.8–6.0) (**Table 6**). Similar trends were established when analyzing the data of dry matter mass of weeds. Irrespective of the soil pH, the effect of different organic fertilizers manifested itself best for the second member of the crop rotation (lupine-oats mixture). The highest number of weeds (356.7 m^{-2}) and weed mass (122.7 g m^{-2}) were established in the treatment where the mixture was grown without organic fertilizers. The number and mass of weeds were on average 1.3 and 1.9 times lower in the

Treatment.	**Winter wheat yield t ha^{-1}**		**Lupine-oats mixture DM, t ha^{-1}**	**Spring barley, t ha^{-1}**	
	Grain 14% moisture	**Straw DM**		**Grain 14% moisture**	**Straw DM**
Soil pH, liming (factor A)					
Unlimed soil (pH_{KCl} 4.1–4.3)	1.90	2.57	8.82	1.99	1.72
Limed soil (pH_{KCl} 5.8–6.0)	3.51**	3.26**	9.25	4.34**	3.00**
Organic fertilizers (factor B)					
Without organic fertilizers	1.83	2.04	8.55	2.17	1.41
Green manure or plant residues	1.91	2.04	8.55	2.31	1.72
Farmyard manure 40 t ha^{-1}	3.04**	3.34**	8.81	3.62**	2.78**
Green manure (bkgd of FM 40)	3.16**	3.44**	10.0**	4.00**	2.52**
Farmyard manure 60 t ha^{-1}	3.45**	3.36**	8.70	3.93**	2.96**
Green manure (bkgd of FM 60)	2.86**	3.29**	9.60	3.00**	2.76**
Interaction of factors A × B					
	**	**	ns	**	**

ns, not significant; DM, dry matter.*Significant at $P \leq 0.05$.
**Significant at $P \leq 0.01$.

Table 5. The effect of soil acidity and organic fertilizers on crop productivity.

treatments where the mixture was grown in the soil applied with organic fertilizers. Fertilization with farmyard manure decreased the number and dry matter mass of weeds during the first 2 years after application. However, the use of green manure on the background of different rates of farmyard manure increased the number of weeds in the cereal crops of the rotation system. In the naturally acid soil, organic fertilization also reduced weed incidence in crops, especially in the lupine-oats mixture. The effect of organic fertilization was weaker in the winter wheat crop, where green manure was incorporated on the background of different rates of farmyard manure. Significantly higher number of weeds was established in these plots. However, reduction in the number of weeds was observed in the farmyard manure fertilization plots.

Fertilization with farmyard manure did not have any effect on the weed number in stands of spring barley. In all the experimental years, short-lived weeds predominated in the crops and accounted for 96.4% of the total weed number. Statistically significant correlations were determined between the agrochemical indicators of soil and the total number and mass of weeds in the rotation crops. Statistical analysis showed that in the first year of the crop rotation, with liming-induced reduction of soil acidity, the number of weed species declined ($r = -0.96^{**}$). In all experimental years, the total number of weeds significantly depended also on the mobile aluminum content in the soil: $r = 0.92^{**}$ in winter wheat crop, $r = 0.86^{**}$ in lupine-oats mixture, and $r = 0.84^{**}$ in spring barley crop.

Treatment	Winter wheat		Lupine-oats mixture		Spring barley	
	Number m^{-2}	Mass of DM g m^{-2}	Number m^{-2}	Mass of DM g m^{-2}	Number m^{-2}	Mass of DM g m^{-2}
Soil pH, liming (factor A)						
Unlimed soil (pH 4.1–4.3)	182.1	101.6	335.4	96.6	127.3	71.4
Limed soil (pH 5.8–6.0)	101.3**	32.0**	247.2**	57.9**	98.6	8.54*
Organic fertilizers (factor B)						
Without organic fertilizers	137.3	83.1	356.7	122.7	98.0	38.5
Green manure or plant residues	128.7	90.5	348.3	97.1	123.7	62.2
Farmyard manure 40 t ha^{-1}	92.7*	35.6**	256.7**	70.1*	112.0	28.9
Green manure (bkgd of FM 40)	177.7	42.1**	243.3**	45.3**	110.3	27.7
Farmyard manure 60 t ha^{-1}	116.3	41.4**	222.7**	63.7*	105.3	14.8*
Green manure (bkgd of FM 60)	197.7	108.0	320.2	64.5**	128.3	68.8
Interaction of factors A × B						
	ns	**	**	**	ns	**

ns, not significant; DM, dry matter.*Significant at $P \le 0.05$.

**Significant at $P \le 0.01$.

Table 6. The effect of soil acidity and organic fertilizers on the weed infestation in the crops of the rotation during the maturity stage.

The plowed layer (0–20 cm) of limed soil was significantly less contaminated with weed seeds compared with naturally acid soil.

The naturally acid soil without organic fertilization contained 7.5 times more weed seeds compared with limed soil. Irrespective of the soil pH, green manure increased soil contamination with weed seeds by 9.4%. However, fertilization with different rates of farmyard manure or incorporation of green manure on the background of different rates of farmyard manure resulted in 54.1–66.0% reduction in the weed seed bank in the soil. This might have been influenced by more active microbiological processes in the soil because of which part of weed seeds were more rapidly mineralized. In the total weed seed bank of not limed soils (pH 4.0–4.1), *Spergula arvensis* L. and or *Scleranthus annuus* L. accounted for 73.7%. In the total weed seed bank of limed soils (pH 6.4–6.8) nitrophilous weed *Chenopodium album* L accounted for 72.8%.

7. Conclusions

The findings of the long-term (more than half a century) liming and fertilizing experiments indicate that liming alone was less efficient for improvement of moraine loam soil Bathygleyic

Dystric Glossic Retisol acidity indicators than its combination with farmyard manure (FYM). The acidification of the soil was neutralized in the topsoil (Ah) and subsoil (ElB) up to 60 cm depth by a systematic soil liming with 1.0 rate every 7 years of powdered limestone in combination with the application of 60 t ha^{-1} of FYM every 3–4 years. The highest mobile P_2O_5 content 220 mg kg^{-1} was in the soil which had been limed and fertilized with 60 t ha^{-1} manure. Changes of nutrients caused by the long-term liming and manuring were established in the deeper horizons (ElB and ElBt) of soil profile as well. Liming is the most efficient practice used to reduce soil acidity, as it eliminates aluminum toxicity and increases calcium content. Under the effect of liming and FYM fertilization, a significant increase (1.9 and 1.2 times) occurred in the exchangeable Ca content in ElB and ElBt horizons, compared with the unlimed soil. Liming exerts an effect not only on exchangeable Ca accumulation in the soil profile but also promotes its leaching especially when combining liming, mineral and FYM fertilization. Liming and NPK fertilization as well as liming, and NPK and FYM fertilization of moraine loam resulted in slightly higher NO^{3-} concentrations in the infiltration water during crop vegetation season compared with other practices. Soil pH determines the activity of biological processes occurring in the soil (CO_2 emission, atmospheric nitrogen fixation intensity, organic matter mineralization rate, distribution of beneficial microorganisms). The content of SOC was 1.45% in the unlimed treatment, while in periodically limed soil at 2.0 liming rate every 3–4 years, it was approximately 0.18 percentage points lower. The highest amount of soil organic carbon (1.91%) was obtained in the limed soil applied with FYM. Soil liming significantly decreased the amount of fulvic acids. Soil amendment with organic fertilizers significantly increased the amount of humic acids, which indicates an increase in organic matter fixation and its stability in the soil. The highest content of water stable macroaggregates (59.35%) was determined in the limed and manure-applied soil. Manure application alone did not have any effect on the formation of water stable aggregates in the topsoil. In limed soil, winter wheat grain yield was 1.8 times and barley grain yield 2.2 times higher than in acid soil. Soil acidity had a significant influence on crop weediness during the stage of maturity. In limed soil, the weed number decreased by 31.1% and their mass by 65.5%, compared to unlimed soil.

Acknowledgements

The chapter presents research findings, obtained through the long-term research program "Productivity and sustainability of agricultural and forest soils" implemented by Lithuanian Research Centre for Agriculture and Forestry. Also the chapter presents research findings, obtained through the project "The effect of long-term management of resources of varying intensity on the soils of different genesis and other components of agro-ecosystems (SIT-9/2015)" funded by the Research Council of Lithuania.

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