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PM_{10} concentration reduction due to the wet scavenging in the Ciuc Basin, Romania

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Abstract. The PM_{10} concentration reduction caused by large-scale precipitation in the Ciuc Basin was studied under no-wind conditions. The PM_{10} concentration changing before, during, and after the rainfall was followed up from 2008 to 2019. After the rainfall episode, the PM_{10} concentrations were lower in the cold and warm periods with 2.8 $\mu g/m^{3,}$ and 2 $\mu g/m^3$ respectively. The highest PM_{10} concentration reduction was detected in the cold season, by the moderate and light rain intensity, after 6 hrs of continuous rain (35.61%, 32.46%), and the average PM_{10} concentration reduction in the cold and warm periods was 22.3% and 16.1% respectively.

Keywords: particulate matter, atmospheric purification, rainfall, meteorological condition

1. Introduction

Particulate matter (PM_{10}) with aero-diameter less than 10 µm harms human health, causing various respiratory and cardiovascular diseases and premature death around the world [1]–[3]. The precipitation washout mechanisms perform the primary atmospheric purification. The unfavourable meteorological parameters, such as inversion and fog, have a negative influence on air pollution concentration [4], [5]. On the other hand, the favourable meteorological conditions via dilution and elimination have a reduction effect [6]. The atmospheric purification may happen through dry and wet scavenging [7]. Wet scavenging can take place in two different ways: in the cloud and below cloud [8]–[10]. The effectiveness of washout is strongly related to the seasonal PM_{10} variation between the seasons: relatively high PM_{10} concentration in the cold period and a significantly lower level in the warm period [11]–[13].

In the background of the PM_{10}, washout effects are responsible for several mechanisms such as Brownian diffusion, thermophoresis, diffusion, inertia, and electric washing [4], [8], [10], [14]. The lifting condensation level variation also plays an essential role in particulate matter variation. Many studies show the effect of precipitation washout by comparing the PM_{10} concentration with the precipitation and non-precipitation periods [9], [15], [16]. The air pollutant (PM_{10}) reduction efficiency by precipitation scavenging depends on the precipitation quantity and duration [8].

This research paper presents the PM_{10} concentration reduction efficiency in the Ciuc Basin caused by rainfall washout under windless conditions, taking into account rainfall intensity, duration, and meteorological parameters.

2. Materials and methods

2.1. Meteorological data and PM_{10} sampling procedure

The study was conducted in the Ciuc Basin area and covered 11 years between 2008 and 2019. The sampling site is an inter-Carpathian, closed-type depression, located in the central part of the Eastern Carpathians, where the fog and inversion phenomena are very frequently present mainly in the cold period [17]–[19]. The hourly PM_{10} and meteorological dates, such as air temperature, relative humidity, precipitation quantity, and wind speed, were provided by the National Environmental Protection Agency Harghita. The monitoring station is situated in Jigodin, with the following coordinates: latitude: 46.33 °N, longitude: 25.81 °E, and altitude: 697 m.

In order to determine the purification effect of precipitation, the hourly PM_{10} concentration was followed up and compared in the case of rainfall and no precipitation period. For an in-depth assessment to determine the atmospheric purification by wet scavenging, different conditions have been set: 1. no-wind condition (< 1 m/s), 2. three rain intensity levels (light: 0.2–0.4 mm h^{-1}, moderate: 0.4–3.9 mm h^{-1}, and heavy rains: > 3.9 mm h^{-1}), and 3. rain duration from 1 to 6 hours were analysed. The analysis was carried out separately for the cold (Oct–Mar) and warm period (Apr–Sep).

2.2 Procedure for determining the removal efficiency coefficient ΔC

The effectiveness of PM_{10} scavenging by precipitation was calculated based on *Equation 1*, where the percentage change (ΔC) was obtained from the concentration variation before (C_0) and after (C_t) episodes of rain.

$$\Delta C = \frac{C_t - C_0}{C_0} * 100 \quad (1)$$

Based on air temperature and relative humidity, the lifting condensation level (LCL) was calculated. Regarding the lifting condensation level (LCL), it was calculated according to *Equation 2*:

$$LCL = 20 + \frac{T}{5}(100 - RH), \quad (2)$$

where: *LCL* – lifting condensation level, *T* – air temperature (ºC), and *RH* – relative humidity (%).

3. Results and discussions

3.1. Statistical description and Spearman's correlation

Summary statistics data are presented in *Table 1* including data regarding the selected air pollutant (PM_{10}), meteorological parameters, and the precipitation, examined during the 11-year experiment. Rainfall intensity (light, moderate, heavy) and duration (1 to 6 hours) have an essential role in the PM_{10} concentration reduction from the troposphere. The analysis of these parameters showed that the main form of wet deposition was carried out by rainfall with low intensity (0.2–0.4 mm h^{-1}) in cold and warm periods (68.82%, 55.92%). The lowest occurrence was observed for rainfall with high intensity (0.4%, 3.4%). During the cold period of observation, the average PM_{10} concentration was 1.32 times higher than the annually acceptable limit (20 μg/m^3). Due to the different emission sources and the unfavourable meteorological conditions, a significant difference was found between the cold and warm period PM_{10} concentration, which was 26.42 μg/m^3 and 10.97 μg/m^3 respectively. The LCL has an important effect on the PM_{10} concentration evolution – almost two-fold differences were found between the average cold and the warm season LCL highs (344 m, 626 m).

Table 1. PM_{10} and meteorological parameters characterization

Precipitation period	Number of rain episode		PM_{10}	T (°C)	RH (%)	LCL (m)	Ws (m s^{-1})
Cold season (Oct–Mar)	L (6176) M (2762) H (36)	**Avg.**	26.42	0.47	83.13	344	0.71
		Med	16.23	0.56	86	269	0.3
		Min	0.10	-27.6	15	0	0
		Max	251.82	28.37	100	1,900	50
Warm season (Apr–Sep)	L (4407) M 3199) H (274)	**Avg.**	10.97	15.28	73.71	626	1.28
		Med	9	15.1	77	524	0.5
		Min	0.1	5.01	12	0	0
		Max	102.1	35.75	100	2,273	50

Note: T – temperature, RH – relative humidity, LCL – Lifting Condensation Level, Ws – wind speed, Avg. – average, Med. – median, Min – minimum, Max – maximum, L – light 0.2–0.4 mm h^{-1}, M – moderate 0.4–3.9 mm h^{-1}, H – heavy rains > 3.9 mm h^{-1}

During large-scale rain events in the cold and warm seasons, the average air temperature was around 0.47 °C and 15.25 °C respectively. The cold season is characterized by higher relative humidity and lower wind speed than the warm season with 83.13%, 73.71% and 0.71 m s^{-1}, 1.28 m s^{-1} respectively.

3.2. PM_{10} concentration with and without precipitation

Concentrations of PM_{10} were lower in the case of rainfalls than in the non-precipitation period, and a noticeable difference in average PM_{10} concentrations was observed in the cold and warm period: 2.8 µg/m³ and 2 µg/m³ respectively (*Figure 1*).

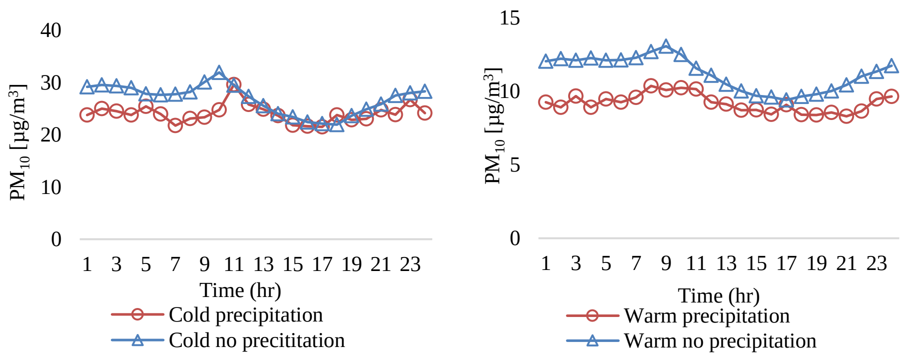
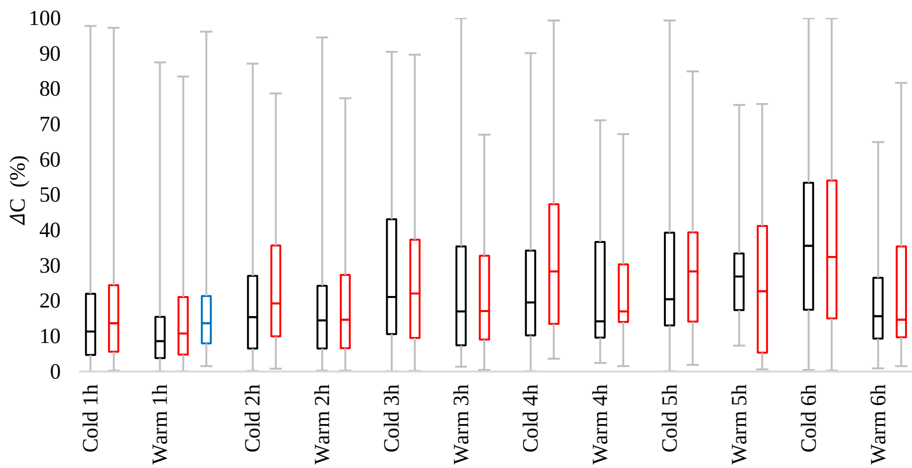


Figure 1. PM_{10} hourly variations under conditions of precipitation and non-precipitation: in the cold and in the warm period

The pattern of hourly PM_{10} concentration variation under conditions of precipitation and non-precipitation was quite similar to each other. Quantitatively, the reduction effect of precipitation scavenging in the cold period was higher than in the warm period. Still, the percentage reduction was 11.69% and 22.06%, resp., thanks to the unequal PM_{10} concentration during the cold and warm season. The hourly PM_{10} concentration in the warm period increased due to a relatively more substantial direct effect of vehicle emissions despite the rainfall.

3.3 Effectiveness of PM_{10} scavenging

Precipitation cases with a duration of one hour in the cold and warm season had the highest frequency with 48.56 % and 76.11%, resp., and a duration of 2 hours was next with 11.51% and 9.72% resp. The PM_{10} concentration reduction due to rain is presented in *Figure 2*. The graphical results indicate that the removal efficiency is growing with the rainfall intensity and duration in the cold period. This increasing trend in the warm period was valid for a rain duration of 1 to 3 hours.



Note: the box plot colour symbolizes rain intensity: black – light (L), 0.2–0.4 mmh^{-1}; red – moderate (M), 0.5–3.9 mmh^{-1}; blue – heavy (H), > 4 mmh^{-1}.

Figure 2. Effectiveness of PM_{10} scavenging in the function of the duration and intensity of large-scale precipitation

The constant values of the ratios ΔC6h to ΔC1h (1.6 and 1.4) and ΔC3h to ΔC1h (1.3 and 1.2) are observed for the cold and warm seasons respectively. The average PM_{10} concentration reduction in the cold and warm periods was

22.3% and 16.1% respectively. In all studied cases, the highest PM_{10} concentration reduction was detected in the cold season in the case of low and moderate rain intensity, after 6 hrs of continuous rain (35.61%, 32.46%). Following the observation, it became evident that the PM_{10} concentration reduction from the atmosphere in the case of the light (< 1 mm) rainfall was smaller thanks to the fact that the impact of pollution exceeded the washout effect. In the cold season, the PM_{10} concentration reduction by wet scavenging from the air was 11.58% higher for the case of heavy rains than for the light ones. In the warm period, this reduction was lower, with 9.09%.

Using the Pearson correlation, the statistical data confirms the significant relationship between PM_{10} concentration reduction and rainfall duration, with a considerable degree of relation ($r = 0.93$). The correlation coefficient was higher than the significance level ($P < 0.05$, $r = 0.7$) in the case of moderate and low rainfall: 0.98 and 0.88 respectively. For the Pearson correlation coefficient calculation in the warm period, the rainfall duration between 1 to 3 hours was taken into consideration only when the continuous removal PM_{10} effect was detected. The Pearson correlation coefficient was higher than the significance level ($P < 0.05$, $r = 0.87$), with 0.97 and 0.99 in the case of low and moderate precipitation resp.

4. Conclusions

During the cold period of observation (2008–2019), the average PM_{10} concentration in the Ciuc Basin was 1.32 times higher than the annually acceptable limit (20 $\mu g/m^3$). The dominant rainfall was the precipitation with low intensity, and the most frequent duration for rainfalls was 1 hour. Due to the different emission sources and the meteorological conditions, a significant difference was found between the PM_{10} concentration measured in the cold and warm periods. The LCL had an essential effect on the PM_{10} concentration evolution; almost two-fold differences were found between the average cold and the warm season. Quantitatively, the reduction effect of precipitation scavenging in the cold period was higher than in the warm period.

In all cases studied, the highest PM_{10} concentration reduction was detected in the cold season in the case of the low and moderate rain intensity, after 6 hrs of continues rain. In the cold season, the efficiency of PM_{10} scavenging by rainfall shows a continuously increasing trend based on the rain duration from 1 to 6 hours. This increasing trend in the warm period was true for rain durations of 1 to 3 hours. The Pearson correlation based on the statistical data confirms the significant relationship between PM_{10} concentration reduction and rainfall duration, with a considerable degree of relation.

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Comparative analysis of relevant climate change, landscape and regional development strategies regarding the areas pertaining to Debrecen (Hajdú-Bihar County)

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Abstract. Today, the countries of the world have to face several global challenges with regard to the plans they have developed together. The protection of the natural values of our country and their sustainable use is receiving more and more attention in today's society. In order to achieve the above, a change of perspective in social strategy built on knowledge and professional training is inevitable. An environmental strategy paradigm shift emphasizing the protection of biodiversity, resources, and landscape cultivation is also necessary. The need for documents providing a basis for the paradigm shift is indubitable; however, more efforts are needed to induce fundamental changes by plans detailed in the documents.

The aim of this study is to review the current environmental protection initiatives in Hajdú-Bihar County and to assess the degree to which the relevant plans are harmonized and that the objectives outlined in the documents overlap.

Keywords: climate change, strategy, biodiversity, sustainable development

1. Introduction

Developed countries enjoy the benefits of globalization on a daily basis; however, they are less effective in terms of the measures taken to combat its negative consequences. Nevertheless, it is undeniable that the unsustainable use of resources, the increase of biological diversity, and climate change lead to irreversible changes affecting even our everyday lives. According to *Climate Change 2014: Synthesis Report*, one of the major causes of climate change is the anthropogenic emission of greenhouse gases (burning of fossil fuels and industrial processes). This leads to the increase of surface-level mean temperature, the acidification of oceans, the change of global water cycles, the faster melting of ice caps, and the increase of global sea level [4]. Several reports and research studies agree that in order to adapt to the above issues and slow down these processes international cooperation and sustainable economy should be achieved. An economy is considered sustainable when it does use its natural resources but only to a degree that does not have excessive impacts on the carrying capacity and tolerability of the environment [3]. The United Nations Conference on Environment and Development (UNCED) held in Rio de Janeiro in 1992 made the first steps towards this goal, yielding the documents which provide the foundations for today's climate change negotiations. These are the United Nations Framework Convention on Climate Change, the Convention on Biological Diversity (CBD), and the Rio Declaration [5]. This is complemented by the Kyoto Protocol signed in 1997 and effective as of 2005 [6]. Among further climate protection events, the following are also worth mentioning: The World Summit on Sustainable Development (WSSD), which summarized the results achieved during the last ten years passed since UNCED [7]; 10 years later, in 2012, a global summit was held again in Rio de Janeiro: The United Nations Conference on Sustainable Development (UNCSD) [8]. In all three conferences as well as in the preparatory negotiations organized during the interim periods, the representatives of the participant countries agreed that the mitigation of factors causing global climate change is necessary, especially in the form of common obligations. From the above, notable goals are the decrease of greenhouse gases and the compilation of national strategies. In the period between 1992 and 2012 as well as afterwards, the decrease of greenhouse gases was a highlighted issue in almost every conference [9]. However, the participant countries were still not able to complete the objectives determined 27 years ago. The measures regarding the compilation of national strategies are still in development today. For the most part, these strategies serve surveying purposes, and they are rarely implemented successfully. Therefore, despite the many strategies, concrete results reflecting a proactive approach are hard to find. Even though the protection and sustainable use of the natural values of Hungary is becoming more and more important for our society, the contradictions between the basic documents fostering the change in perspective suggest that the presentation of the baseline situation is not the same either at the national or at the local level; priorities and objectives are constantly changing, and there are differences in the methods and tools used for the implementation.

2. Materials and methods

In our study, we attempt to highlight the contradictions found in the following Hungarian strategic documents and development programmes as well as to review the implementation of national objectives in the urban development plans of Hajdú-Bihar County and Debrecen [10–16]:

- National Development 2020 (NF2020),
- National Development 2030 – National Development and Regional Development Plan (NF2030),
- National Climate Change Strategy (NÉS 2008–2025),
- Second National Climate Change Strategy 2014–2025 (NÉS II),
- National Landscape Strategy (NTS),
- Hajdú-Bihar County Regional Development Programme 2014–2020,
- Debrecen MJV Urban Development Concept and Integrated Urban Development Strategy 2014–2020.

3. Results and discussions

A. Demographic processes

One of the main causes of the global issues is the rapid increase of population; however, in Hungary, the population is decreasing and the fundamental problem is emigration. One of the top priorities of the National Development 2020 (NF2020) [10] is to arrest and reverse the negative demographic processes. However, this objective is missing from the National Development 2030 [11] as if it acknowledged the fact that the population of the country is decreasing at a constant rate. The population of Hajdú-Bihar County has been decreasing by approximately 1,000 inhabitants annually since the turn of the millennium – today, the number of inhabitants of the county is close to that of the year 1975. The population of Debrecen had been increasing until 2011, when it reached 210,000; since then, it has been slowly decreasing, and it was only 202,000 in 2017 (KSH) [17].

The constant and even permanent draining effect of the capital, of the western regions and western countries can be detected across the county. According to the National Landscape Strategy (NTS) [14], internal migration affects mainly young career entrants, which leads to the aging of the population in the areas impacted by the emigration processes. Hajdú-Bihar County is not one of the most rapidly aging counties, showing a trend which is even below the national average; e.g. the number of people older than the age of 65 per 100 children is increasing at only a moderate rate according to the data of the Hungarian Central Statistics Office regarding 2009 [17].

B. The urban development plan of Debrecen

The settlement and regional structure of Hungary started to change, as a result of which cities play a more significant role, including Debrecen as a major city in the Great Hungarian Plain since it is one of the most economically developed settlements of the so-called outer city ring (Miskolc–Nyíregyháza–Debrecen–Békéscsaba–Szeged–Pécs–Kaposvár–Nagykanizsa–Zalaegerszeg–Szombathely–Sopron–Győr) (according to NF2020). Even though it is a regional centre, it cannot counteract the dominant effect of the capital. However, the city is responsible for providing opportunities for innovation and transmitting development to other Hungarian cities and regions across the border.

Debrecen is a settlement of great economic potential – it is an economic and technological core area because it can build upon more take-off points such as food industry, biomass utilization industries, chemical engineering, or the pharmaceutical industry. The developments made in the field of chemical industry provide more economic advantages. Their regional distribution is focused since they are associated with universities and research centres; therefore, Debrecen has a major advantage in this regard as well. The development of environmental technologies could also provide significant potential for the city. In the MJV Urban Development Concept of Debrecen, we can find more horizontal principles: According to the principle of “Liveable, sustainable housing and institutional environment for the citizens of Debrecen”, the key areas of sustainability are the brown-field and green-field investments, building rehabilitation projects, and the touristic use of thermal water. According to the principle of “Improving life quality as a response to the challenges of the aging society”, it is necessary to provide healthcare prevention, retirement insurance opportunities and to increase the available recreation services. Furthermore, the principle “Integration of innovation perspective into the economic, societal, and environmental processes” highlights the significant role of innovation and knowledge; therefore, the use of environmentally-friendly technologies can be a key element to the development of this area [16]. The parts of NF2020 and 2030, where the needs and responsibilities of the counties and the capital are discussed, match entirely, and the contents of other sections of the strategies are in accordance as well. However, the green-field industry development approach mentioned in the MJV Urban Development Concept of Debrecen is no more included in NF2030. This is directly contradicted by the statement made in the summer of 2018, according to which the new green-field investment of BMW will be implemented in Debrecen, which can be accompanied with the establishment of a new freeway junction and a new railway section.

C. Opportunities for restructuring energy production

Development strategies include the improvement of environmental quality by focusing on renewable energy sources and the implementation of developments in a way that takes landscape protection aspects into account. Natural resources are becoming scarcer and scarcer and more vulnerable, while the energy demands of Europe continue to increase. That is the reason why the most necessary measures to be taken will increase the degree of energy conservation and energy efficiency by relying more heavily on renewable energy sources. The reform of the Common Agricultural Policy of the EU is partly about achieving a sustainable management of natural resources. Even though the geothermic properties of Hungary are exceptional even on the international level (the Tiszántúl region is one the areas where sun, wind, and geothermic energy use are recommended), their degree of utilization is far from what could be achieved from a technological standpoint. It should be noted that, contrary to the information stated in NF2020, the average wind speed of the region is even lower than the already limited national wind energy potential. In Debrecen and the neighbouring settlements as well as throughout the country, the degree of solar energy utilization is increasing (e.g. with the use of solar cells), but Hungary is still extremely dependent on the import of solar energy carriers. According to the Hajdú-Bihar County Urban Development Programme (2014–2020), the energy efficiency and the ratio of renewable energy should be increased by 20% and the emission of greenhouse gases should be decreased with 20% until 2020 to meet the objectives outlined in the EU2020 strategy. According to the programme, the dependence of Hungary on energy import could be reduced to zero with sustainable energy use, the establishment of zero-energy buildings, and the use of energy plants [15]. This dependency is highlighted by the First National Climate Change Strategy (NÉS), according to which approximately 80% of the domestic energy use is based on fossil fuels, and it is almost entirely imported energy [12]. The increase of energy demands covered through import energy is an important challenge according to NF2020 as well as NF2030. Based on the above, the most important objectives include the increase of energy efficiency and the increase of the ratio of renewable sources. Even though during the 1990s the energy use and output significantly decreased as a consequence of the structural change occurred in the energy sector, no major changes can be expected on the midterm according to NÉS, whereas the goal would be the prevention of industrial developments with large energy demands and greenhouse gas emission. With regard to renewable energy use, in 2001, Hungary committed to double the ratio of renewable energy by 2010 (in 2010, the ratio of renewable energy sources was 8.7% in the domestic energy supply). Even though the country met this objective in 2005, it was done mostly by burning forests, which cannot be considered a long-term solution. Efforts to achieve these goals go beyond national borders. In terms of the energy efficiency, especially the use of biogas has been the focus of many research programmes (I. Fazekas et al. 2013, Gy. Szabó et al. 2014, Gy. Szabó et al. 2014) within the framework of the cooperation programme between Hungary and Romania [23, 24, 25]. Based on a decision associated with the European Commission, a "renewable heat" directive had to be drawn up, which stated that a support system should be developed for providing the foundations for renewable heating and cooling. In January 2017, METÁR was launched, which is a type of support system for power stations to generate electricity from renewable energy sources [18]. According to NÉS, it is an EU requirement to increase the proportion of biofuel use by 10%. This ratio objective can be met by 2020 through domestic production. It is surprising that these requirements and obligations are not included in any of the national development concepts. The reason for this may be that the national energy policy is committed to increase the capacity of the Paks nuclear plant.

D. Agriculture as a key sector

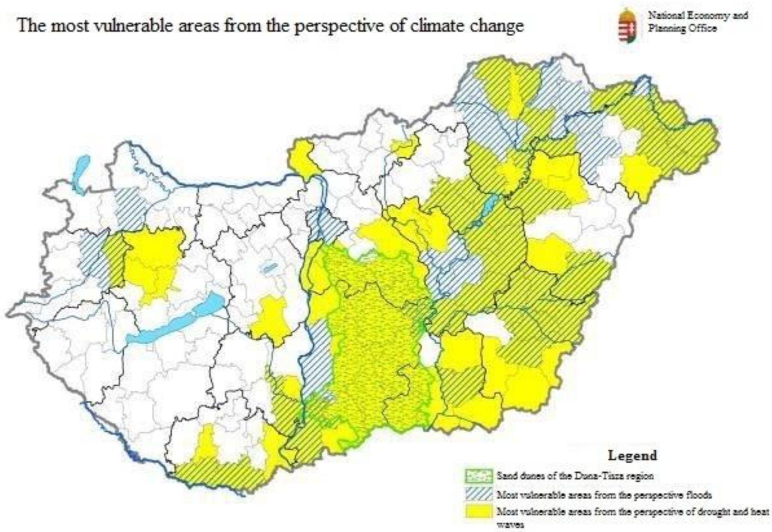
Considering that the county is situated in the Great Hungarian Plain, the most highlighted field in strategies regarding Hajdú-Bihar County was agriculture. Since this sector showed the greatest GDP increase, the area may have significant economic potential. However, the degree of development is unclear – according to National Development 2020, the added value of the sector increased by 10% in 2011, whereas the same value is 24% in National Development 2030. With the development of irrigation and using breeds that are better suited to tolerate extreme weather conditions, it should be a sector which not only increases the economic power of the country but adapts to the climate change – at least according to the development plan. However, it raises the question of how sustainable the development of irrigation is if the climatic water balance of the Great Hungarian Plains is already negative. The connection between climate change and the changes occurring in the agricultural sector was already included in the first NÉS. It states that agriculture is especially vulnerable to extreme weather changes. Extreme water balance conditions (drought, floods, and inland water) pose significant economic risks. According to the strategy, agriculture could be the most vulnerable sector to climate change if the land use type is not chosen in accordance with the conditions of the land. According to the second NÉS, the main problem is that the average annual precipitation would decrease, and the amount of precipitation would be redistributed – during winter and summer months, significantly more and less precipitation can be expected respectively. Nevertheless, heavy storms will be more frequent, thereby increasing the risk of floods, and Hungarian rivers may dry to half of their regular levels during summer seasons within decades. Furthermore, according to the report, the level of groundwater can also decrease due to lack of supply and the presence of surface water [13].

According to the Hajdú-Bihar County Regional Development Programme, the county is poor in surface water and rich in underground water. From the perspective of climate change, the county is especially vulnerable, and the ratio of regions exposed to the risk of drought is continuously increasing. The most significant issues are the management of rainwater in the areas of settlements, the elimination of inland water from lands, the establishment of irrigation systems, the provision of healthy drinking water, and the diversion and management of communal sewage water. According to the programme, the area of lands which can be irrigated has increased by 55% since 2000. The solution could be the complex water management that would include agricultural irrigation, inland water elimination, and storage as well as its use for recreational purposes. With regard to the above, it was almost two decades ago that the CIVAQUA programme was developed, which would transport water from River Tisza to the area of Debrecen via the Eastern Main Canal. The CIVAQUA programme would provide the appropriate water supply for the Tócó stream, solving the problem of managing the water supply of the Erdőspuszta lakes and the capacity expansion of the reservoir of Látókép. However, the elements of the CIVAQUA development programme conceived in the early 2000s are yet to be realized. These are planned to be completed via a new project (a multi-purpose water management system of Hajdúhátság). The goal is to develop the irrigation potential of the agricultural areas situated west of Debrecen, for which the Eastern Main Canal would provide the water supply. Another goal is to establish the reservoir of the Valley of Ágod as a solution for the inland water diversion and retention issues of the area. The project is aimed to be completed in 2019 [19].

Agricultural professions have a low prestige; therefore, it is advisable to prevent the shortage of professionals in demand in accordance with the objectives determined for the sector. This is further aided by the noteworthy attempt of the University of Debrecen to offer the first agricultural and water management engineer MSc training programme of the country from 2018. The curriculum includes modules of traditional and precision irrigation technologies, water management and quality assurance tasks, the management of oxbow lakes and wetland conservation, and the students will also learn the necessary skills to perform tasks related to hydrology, hydraulics, geoinformatics, remote sensing, and data collection and modelling [20].

Vulnerability

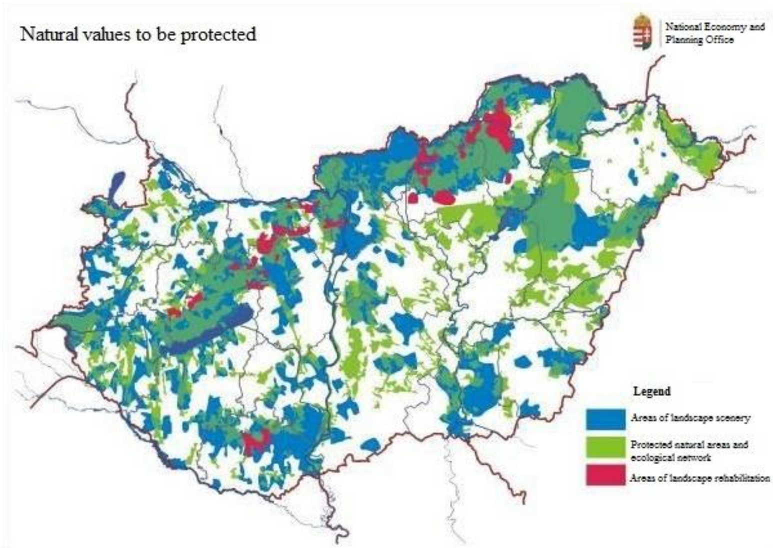
The effects of climate change show a great variability in Hungary – the degree of vulnerability varies from area to area. According to the first NÉS, the entire Carpathian Basin can be considered a vulnerable area. The second NÉS, however, differentiates between the climate change vulnerability of agricultural plant cultivation, forest management, and natural habitats, but neither of these correspond with the figure included in the NF2020 and NF2030 concepts (*Fig. 1*).



Source: National Development 2020 [10]

Figure 1. The most vulnerable areas from the perspective of climate change

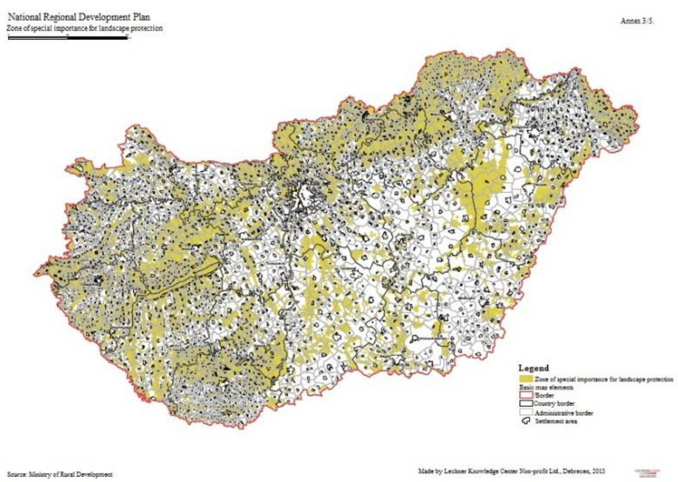
It is problematic that the above figure rates climate vulnerability based on administrative borders and regions, but an essentially natural geographical issue cannot be presented in the framework of administrative units. For example, the extent of vulnerability is obviously equal on the entire area of the Hajdúhát loess territory, but according to *Figure 1* its northern and southern parts were classified into different groups. Reports based on natural landscape borders have also been created, but these apply to landscape scenery protection (*Fig. 2*).



Source: National Development 2020 [10]

Figure 2. Natural values to be protected

The contents of *Figure 1* are also hard to reconcile with the map applying the conservation of natural values published in NF2020 (*Fig. 3*).



Source: National Regional Development Plan 2013 [22]

Figure 3. Zone of special importance for landscape protection

Furthermore, NF2020 states that the most vulnerable region is the part of the country situated south from the Nagykanizsa–Budapest–Tokaj line, which is in contradiction with the information presented in *Figure 1*. According to *Figure 1*, the least vulnerable areas can be found in this county.

E. The role of vegetation coverage

Beyond the predominant continental effects, the weather of the country is also affected by oceanic and Mediterranean impacts. For the most part, this climatic transition zone is the cause of the mosaic nature of soils, precipitation patterns, and natural vegetation coverage. The biogeographic zonality characteristic of Eastern Europe ends in the Carpathian Basin, where it gradually changes to the speckled pattern which can be seen in the western part of the continent. It results in a great variability, which is one of the major natural geographical characteristics of the Carpathian Basin and the reason why the Pannonian Biogeographical Region is recognized as a separate type. Now, when global climate changes pose a significant risk to the existence of several natural habitats, the mosaic characteristic is a beneficial property because the different degree of climate vulnerability of each habitat type allows the gradual transformation of the regional pattern (NTS).

The NTS explains in detail that the changes in urban lifestyle habits as well as the accompanying changes and needs lead to an ever denser infrastructure network, the decrease of biodiversity, and the fragmentation of natural habitats. The need for larger living spaces, the increase in the number of services, and the use of cars all contributed to the development of city peripherals, increased fragmentation, and the change of landscape features. Based on the data of the Hungarian Central Statistics Office, urban areas made up 22% of the area of the country in the period of 2013–2014, which is comparable to the combined ratio of Natura 2000 and forest areas.

With regard to the mitigation of the negative consequences of climate change, forest coverage plays a key role since it absorbs 12–15% of the carbon emission, among others. The forest coverage ratio of Hungary (21%) is lower than the EU average, even though according to the National Development 2020 as well as the National Development 2030 the extent of forest areas is constantly increasing. According to the National Landscape Strategy, the forest coverage of Hajdú-Bihar County (with the exception of its northeastern part) is lower than 10%, the increase of which is neither expected nor necessary considering Hortobágy and the exceptional land properties of Hajdúhát. The nature conservation priorities in connection with the Hortobágy National Park can only partly apply to forest areas [21]. Forestation and green area management in the county should be focused on urban environments because these are the areas where direct life conditions can be improved most significantly. This special situation of the county is not detailed either in NF2020 or in NF2030. Forestation is considered a general method for improvement even though it is obvious that the forestation ratio of the county is low now and will stay low in the future.

F. Landscape strategy

As opposed to other strategies and concepts, the NTS includes specific goals and tasks in connection with the government, the local government, NGOs, and research organizations. A compilation of the landscape reports and documents specifying landscapes is required by the European Commission. The NTS meets this requirement by stating that its comprehensive goal is responsible landscape use based on landscape properties, and in order to reach these goals it also contains horizontal principles such as the protection of natural resources and cultural heritage, economical land use, and adaptation to climate change. One of its priorities is the enhancement of landscape identity, social participation and its integration into education. Further objectives include the improvement of data of records, complex landscape research, reassessment of land use, the climate-friendly development of settlement models that take value conservation into account, the reassessment of the National Curriculum for Basic Education, and the organization of training programmes. The fact that a complex landscape evaluation method which would appropriately reflect the interests of sectors from forest management to tourism is still under development makes it more difficult to implement the goals of the landscape strategy.

G. The financial framework of developments

The most significant support for the implementation of developments comes from the European Union in connection with the operative programmes (EEOP, CCOP, EDIOP, HFOP). According to the 2020 and the 2030 concepts, the current financial framework applies to the periods of 2007–2013 and 2007–2014 respectively. This difference is important because the amount of the support is 24.9 billion euros, and it matters exactly how many years it is available for. It is not reassuring either that, e.g., the Hajdú-Bihar County climate strategy created in February 2018 hardly mentions the financial aspects of the implementation.

4. Conclusions

As we could see above, many ambitious strategies have been created to facilitate changes. However, in many cases, there are significant differences between the contents of the National Development 2020, 2030, and other strategies. We find that the data applying to the same year differ in the two development plans, or they are simply not in accordance with the landscape strategy and the climate change strategies. Among the projects supported by the European Union, the most important are the Hajdú-Bihar County climate strategy accepted in February 2018 and the still ongoing national programme with the identification number KEHOP-4.3.0-15-2016-00001 (strategic assessments supporting the long-term conservation of natural values of community interest as well as the national implementation of the EU Biodiversity Strategy for 2020), which can facilitate the further involvement of the society, and thereby we can make a step towards the solution of a very complex natural problem.

In the case of Hungarian strategies, it would be important to demonstrate how well they correspond with the plans of the European Union and to what extent they meet the requirements of the EU. The changing objectives of the European Union make it difficult to develop mutually supportive strategies. These strategies can only fulfil their purpose if the real motivations behind their creation are the battle against climate change and the need for development. It is, of course, a very delicate subject to discuss these guidelines in a region which is lagging behind, where the current commitments are not in accordance with the EU objectives.

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Nodulation and biological nitrogen fixation in soybean (*Glycine max* L.) as influenced by phosphorus fertilization and arbuscular mycorrhizal inoculation

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Abstract. Arbuscular mycorrhizal fungi (AMF) can be used to promote the productivity of legumes on phosphorus- (P) deficient soils. The present study investigates the inoculation effects of three AMF species (*Funneliformis mosseae*, *Rhizophagus intraradices,* and *Claroideoglomus etunicatum*) and the control (uninoculated) on nitrogen fixation efficiency and growth performance of tropical soybean cultivar (TGx1448-2E) under varying P fertilizer rates (0, 20, and 40 kg P_2O_5 ha^{-1}) in a derived savannah of Nigeria. The results showed that shoot and root dry matter, number of nodules, relative ureide abundance (RUA), nitrogen derived from atmosphere (Ndfa), total N fixed, shoot P uptake, grain, and biomass yield significantly increased with AMF inoculation, with better performance observed in plants inoculated with *Rhizophagus intraradices* and *Funneliformis mosseae* compared to *Claroideoglomus etunicatum*. Similarly, the soybean growth variables, P uptake, and nitrogen fixation activities increased with increasing P application rates. Conversely, AMF root colonization significantly reduced with increasing P rate. Interaction of AMF inoculation and P rates significantly influenced soybean growth and nitrogen fixation. *R. intraradices* inoculation with 20 kg P_2O_5 ha^{-1} resulted in the highest amount of RUA, Ndfa, N fixed, and grain yield. It could be concluded from this study that *R. intraradices* with moderate P rate could be used to enhance nodulation, nitrogen fixation, and soybean yield in P-deficient soils.

Keywords: biological nitrogen fixation, mycorrhizal fungi isolates, phosphorus uptake, ureide assay, soybean productivity

Introduction

Legumes (*Fabaceae*) are important crops in agriculture with abundant levels of proteins and oils for both human and livestock consumption. They do great service to the sustainability of most agricultural systems. Their symbiotic relationship with soil rhizobia helps them in the fixation of atmospheric dinitrogen (N_2), thus reducing the intensive application of inorganic nitrogen (N) fertilizers on agricultural soils while maintaining soil fertility for sustainable crop production. Soybean (*Glycine max* L. Merrill) is an important oil-seed legume cultivated in many parts of the world, including Nigeria [1]. However, low phosphorus (P) availability is the major factor limiting legume productivity in most tropical soils [2]. The nitrogen fixation process in legumes is highly sensitive to adverse environmental conditions, P deficiency [3]. This has become crucial because the way N_2 fixation in soybean is affected by P deficiency is not fully explored under field conditions. Therefore, a better understanding of the responses of N_2-fixation in soybean in P-deficient soils is urgently needed for promoting soybean production towards achieving food security, particularly in sub-Saharan Africa.

Phosphorus plays an important role in the regulation of many metabolic processes in plants, including photosynthesis and N_2 fixation [4, 5]. Nevertheless, P deficiency has negative implications on nodule functioning in legumes [6, 7] due to the high P demand in N_2-fixating nodules for optimal functioning and cell energy metabolism [3, 8]. In addition, the amount of carbon delivered to the nodules and the absorption of ureides in the nodules are reduced under P deficiency [9, 10]. The report of [11] showed that P application positively influenced N_2 fixation in legumes with increased N content. Hence, an adequate P availability in the nodules is essential for N_2 fixation in P-deficient soils.

The intensive use of inorganic P fertilizers has been widely promoted in most agro-ecosystems to maintain a sufficient level of P for crop productivity [12]. The recovery of P application is reported to be very low (10–30% of the fertilizer P applied) [13]. In addition, most farmers in sub-Saharan Africa are resource-limited and may not be able to afford the high P fertilizer cost. Moreover, excessive use of P fertilizer may lead to environmental pollution due to the eutrophication of underground waters and the contamination of soils with heavy metals. There is also report of the potential scarcity of P fertilizers due to the rapid depletion of phosphate rocks. In the context of sustainable agriculture and global P crisis faced by farmers, arbuscular mycorrhizal fungi (AMF) are essential components of sustainable soil-plant systems, forming symbiotic relationships with most plants, including legumes [14]. AMF stimulate the growth of the host plants through increase in soil inorganic nutrients' uptake, particularly P [14, 15]. With reduction in the application of chemical P fertilizers, they could play an important role in meeting the food requirements of an increasing human population [16, 17]. In legumes, AMF has been reported to promote growth and yield performance in soils with low P availability [18, 19, 20]. With increased number of nodules and size, AMF have positive effect on nodulation [21]. The co-inoculation of AMF and rhizobia is reported to promote N_2 fixation in legumes [22]. Nevertheless, the symbiotic contribution of different AMF isolates on N_2 fixation in legumes has not been fully explored, particularly in a derived savannah of Nigeria with low soil phosphorus availability. The potential of AMF in promoting P uptake in host plants under low P availability could be explored within the context of enhancing nodulation and biological nitrogen fixation in soybean grown on P-deficient soils in this agro-ecology.

The present study will help in understanding how AMF inoculation could promote the symbiotic nitrogen-fixing ability of a promiscuous soybean cultivar (TGx 1448-2E) on P-deficient soils. Thus, this study hypothesized that AMF would enhance the nodulation, nitrogen fixation, and growth performance of soybean on P-deficient soils. Hence, this study aimed to investigate the effects of AMF inoculation with three AMF (*Funneliformis mosseae*, *Rhizophagus intraradices*, and *Claroideoglomus etunicatum*) isolates on the nodulation, nitrogen fixation, and yield performance of soybean plants under different soil P availabilities.

Materials and methods

Study site

The experiment was carried out at the experimental field of the Federal University of Agriculture Research Farm, Abeokuta, Southwest Nigeria (latitude 7° 15' N, longitude 3° 28' E) in the 2017 cropping season. The initial soil properties were determined using standard methods (*Table 1*). A bulk soil sample was collected before planting to determine the physico-chemical properties of the soil using standard protocols. The soil properties determined were soil pH (1:1 soil: water) using glass electrode pH meter [23], organic carbon using the Walkley–Black method as modified by [24], total nitrogen using the micro-Kjeldahl distillation method [25], available phosphorus using Bray-1 [26] and determined colorimetrically using the method of [27]. The exchangeable bases were extracted with 1N ammonium acetate (IN NH_4 OA_C), buffered at pH of 7, and soil particle size distribution was determined using the hydrometer method [28]. The initial AMF spore count was determined using the wet sieving method [29].

Experimental treatments and design

The study consisted of four levels of AMF inoculation (non-AMF (control), *Funneliformis mosseae*, *Rhizophagus intraradices*, and *Claroideoglomus etunicatum*) and three P fertilizer rates (0, 20, and 40 kg P_2O_5 ha^{-1}) laid in a split-plot design with three replications. The plot size was 4 m × 4 m.

Source of experimental materials, sowing, and AMF inoculation

Pure AMF inocula (*F. mosseae, R. intraradices,* and *C. etunicatum*) were obtained from the International Institute of Tropical Agriculture, Ibadan, Nigeria. The inocula were multiplied in sterilized sand, using single-plant culture of maize (*Zea mays* L.) for four months consisting of spores, hyphae, and colonized root pieces. Soybean seeds (TGx 1448-2E) were obtained from the Institute of Agricultural Research and Training, Ibadan, Nigeria.

The field was prepared by ploughing followed by harrowing. The plots were marked out with ropes and pegs and then labelled. The seeds were sown at a depth of 2–5 cm and a spacing of 50 cm × 10 cm on 20 July 2017. AMF inoculum (10 g) was applied to the planting hole before sowing. Phosphorus was supplied through single superphosphate at the time of sowing.

Data collection

Dry matter accumulation and nodulation

Ten plants were randomly harvested from the net plot at the early pod filling stage and separated into leaf, stem + petiole, root, and nodules. The samples were oven-dried at 70 ºC for 72 hrs to determine the dry weights using a precision measuring scale. The number of nodules was counted, and the average number of nodules per plant was determined.

Estimation of relative ureide abundance and biological nitrogen fixation

The N_2 fixation in the soybean cultivars was measured during the early pod-filling stage using the xylem ureide assay method [30]. Blended subsamples of stem + petiole (0.5 g) were used to extract the xylem solutes in boiling water, and the ureide and nitrate concentration were measured according to the procedures of [31]) and [32]. The blended plant samples were digested in concentrated hot acid (H_2SO_4), and the total N content was determined colorimetrically using automated analysis. The relative ureide-N of the sample was calculated using the equation:

$$\text{Relative ureide} - \text{N}\ (\%) = \frac{4\ \times\ \text{ureide}}{[(4\ \times\ \text{ureide}) + \text{nitrate}]} \times 100$$

The standard curve relating the proportion of N derived from N_2 fixation (Ndfa%) to RU-N for soybean during pod filling was obtained using the equation below:

$$x = 10.7 + 0.50P + 0.0034P^2,$$

where P is N derived from atmosphere (% Ndfa), and x is the ureide-N.
The shoot N (kg ha^{-1}) was calculated using the equation below:

Shoot N (kg ha^{-1}) = [shoot N concentration (%) × shoot dry matter (kg/ha)]

Total N was calculated by multiplying with a factor of 1.5 [33].

Crop N (kg ha^{-1}) = (Shoot N × 1.5)

The amount of N_2 fixed was calculated using the equation below:

Amount of N_2 fixed (kg ha^{-1}) = (%Ndfa × Crop N)

Estimation of AMF root colonization

Root colonization by AMF was measured according to [34]. The preserved root samples were cleared in hot KOH solution (10% w/v, at 90 ºC) for 1 hour and stained with trypan blue lacto-glycerol (1:1:1:0.5g) at 90 ºC for 30 minutes. The AMF colonization (RLC) was determined as the percentage of root length colonized by AMF was calculated [20].

$$RLC = \frac{Number\ of\ colonized\ roots}{Total\ number\ of\ roots} \times 100$$

Grain yield

At harvest maturity, the plants were harvested from the plots, threshed, and air-dried to estimate the grain yield per hectare.

Statistical analysis

The data collected were subjected to two-way analysis of variance (ANOVA) with Genstat Release 12.1 (Copyright 2009, VSN International Ltd). Duncan’s multiple range test was used to separate treatment means at 5% probability level. Data expressed as percentages and relative to counts were arcsine-square-root and log (× + 1) transformed.

Table 1. Soil physical and chemical properties of the experimental site

Soil Property	**Value**
Texture	Sandy loam
Sand (g kg^{-1})	707.5
Silt (g kg^{-1})	127.5
Clay (g kg^{-1})	165.0
pH (H_2O)	5.70
Organic Matter (%)	1.78
Nitrogen (%)	0.09
Available Phosphorus (mg kg^{-1})	6.13
Potassium (cmol kg^{-1})	0.61
Calcium (cmol kg^{-1})	6.68
Magnesium (cmol kg^{-1})	1.47
Sodium (cmol kg^{-1})	0.29
Total Exchangeable Acidity (cmol kg^{-1})	0.11
Cation Exchange Capacity (cmol kg^{-1})	9.17

Results

Dry matter accumulation

Phosphorus application and AMF inoculation had significant effect on the shoot and root dry matter of the soybean (*Table 2*). The shoot and root dry matter increased with P fertilizer application compared to the control (no P fertilizer added); however, no significant differences were recorded between the application of 20 and 40 kg P_2O_5 ha^{-1} (*Table 3*). AMF inoculation significantly increased the shoot and root dry matter over non-AMF treatment (*Table 3*). The increase in shoot and root dry matter induced by *F. mosseae* (20.6 and 16.9%) and *R. intraradices* (23.9 and 15%) over non-AMF treatment was significantly greater than that obtained with *C. etunicatum* (9.3 and 5.2%). There was a significant interaction effect of P application and AMF inoculation on the shoot and root dry matter of the soybean plant (*Table 2*). Soybean inoculated with *R. intraradices* with 20 kg P_2O_5 ha^{-1} fertilizer rate significantly had the highest shoot and root dry matter, followed by inoculation with *F. mosseae* at 20 kg P_2O_5 ha^{-1} application rate and then 40 kg P_2O_5 ha^{-1} fertilizer application alone (figures 1a and b). With high P fertilizer application (40 kg P_2O_5 ha^{-1}), the shoot and root dry matter decreased, but no significant difference was observed between the AMF isolates and non-AMF treatments (figures 1a and b).

Nodulation

The results showed that P fertilizer application rates and AMF inoculation had significant effect on the number of nodules of soybean but not on the nodule dry weight (*Table 2*). The number of nodules increased with the application of 20 and 40 kg P_2O_5 ha^{-1} rates (58.6 and 60.2% respectively), but no significant difference was observed between the two rates (*Table 3*). Soybean inoculated with *R. intraradices* significantly had the highest number of nodules compared to other AMF isolates and non-AMF treatment (*Table 3*). A significant interaction between P fertilizer application rates and AMF inoculation was observed for the number of nodules of soybean (*Table 2*). The lowest number of nodules was recorded in non-AMF soybean plants and no P fertilizer application, while the highest number of nodules was recorded in plants inoculated with *R. intraradices* with 20 kg P_2O_5 ha^{-1} rate (*Figure 2).*

Table 2. Mean squares for the effect of P fertilizer application (P) and mycorrhizal inoculation (M) and their interactions (P × M) on dry matter accumulation and nodulation in soybean

	df	Shoot dry weight (g $plant^{-1}$)	Root dry weight (g $plant^{-1}$)	Number of nodules ($plant^{-1}$)	Nodule dry weight (g $plant^{-1}$)
Reps	1	11.8	0.016	102.1	0.029
P rates (P)	2	20.1**	0.33**	674.6**	0.061^{ns}
Error a	2	0.09	0.005	7.72	0.014
AMF (M)	3	10.2***	0.12***	190.9***	0.027^{ns}
P × M	6	7.32**	0.091***	96.8***	0.019^{ns}
Error b	9	0.09	0.003	2.84	0.011
Total	23				

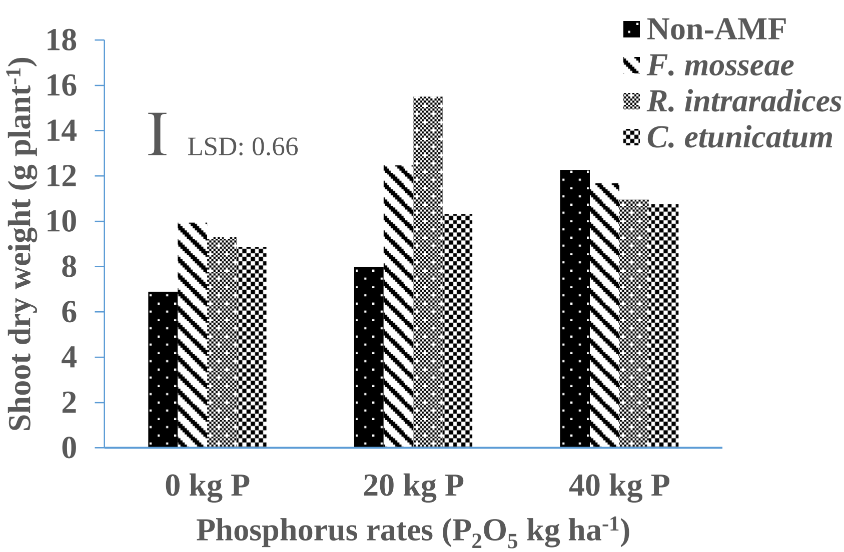
Notes: *, **, *** indicate significance at 0.05, 0.01, and 0.001 probability level respectively; ns indicates not significant.

Table 3. Effects of P fertilizer application (P) and mycorrhizal inoculation (M) on dry matter accumulation and nodulation in soybean

	Shoot dry weight (g plant^{-1})	Root dry weight (g plant^{-1})	Number of nodules (plant^{-1})	Nodule dry weight (g plant^{-1})
P rates (P)				
0 kg ha^{-1}	8.75	1.39	10.9	0.19
20 kg ha^{-1}	11.6	1.73	26.3	0.35
40 kg ha^{-1}	11.4	1.76	27.4	0.32
Lsd ($p < 0.05$)	0.67	0.15	5.97	ns
AMF inoculation				
Non-AMF	9.05	1.47	15.0	0.22
F. mosseae	11.4	1.77	24.6	0.37
R. intraradices	11.9	1.73	27.6	0.31
C. etunicatum	9.98	1.55	18.9	0.24
Lsd ($p < 0.05$)	0.39	0.07	2.20	ns

Note: ns indicates non-significant differences at p value < 0.05.

a.



b.

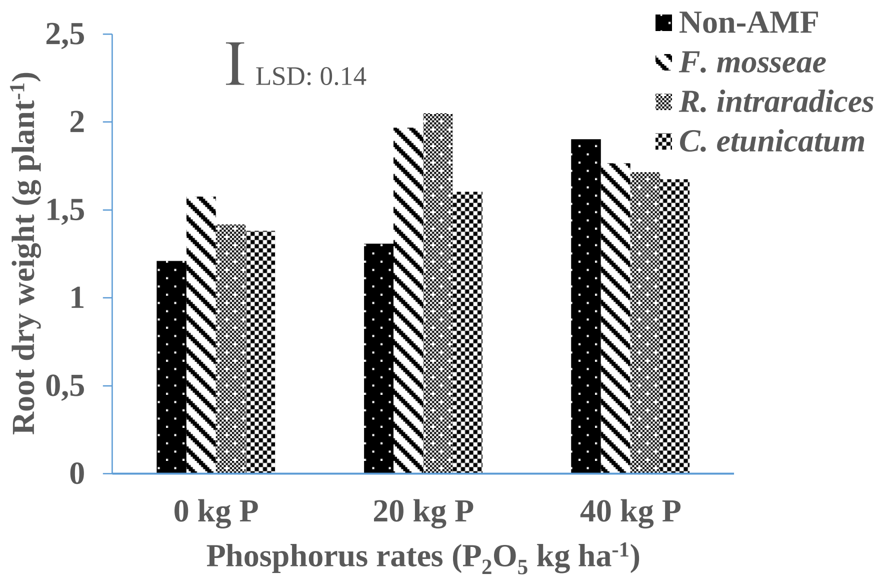


Figure 1. Interactive effect of phosphorus rates and AMF inoculation on (a) shoot dry weight and (b) root dry weight of soybean. Bar denotes Lsd value at 5% probability level.

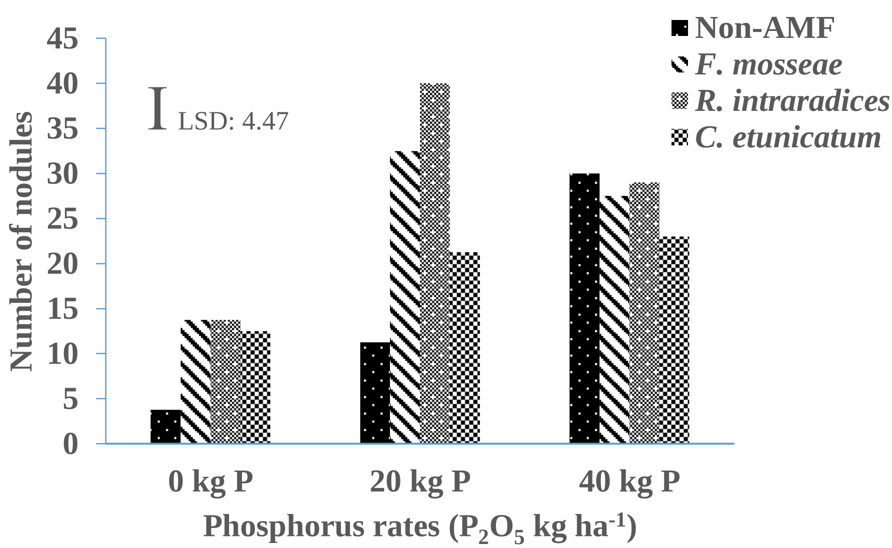


Figure 2. Interactive effect of phosphorus rates and AMF inoculation on the number of nodules of soybean. Bar denotes Lsd value at 5% probability level.

Nitrogen fixation

The P fertilizer application had significant effect on the relative abundance of ureide-N and the amount of nitrogen fixed by the soybean plant but not on concentrations of nitrate-N (NO_3-N) and ureide-N and the proportion of N derived from atmosphere (Ndfa) (*Table 4*). The relative ureide-N and the amount of N fixed increased with increase in P rates (*Table 5*). However, there was no significant difference between the application of 20 and 40 kg P_2O_5 ha^{-1} rates (*Table 5*). The concentration of nitrate-N was recorded to decrease with increase in P rates, while ureide-N and Ndfa increased with increase in P rates; however, no significant difference was observed among the P rates (*Table 5*).

The concentrations of nitrate-N (NO_3-N) and ureide-N, the relative abundance of ureide-N, the proportion of N derived from atmosphere (Ndfa), and the amount of N fixed in the soybean plant were significantly influenced by AMF inoculation (*Table 4*). Significant differences were observed among the AMF isolates and the non-AMF treatments regarding these parameters (*Table 5*). The *Funneliformis mosseae* and *R. intraradices* inoculation effects were significantly higher than in the case of *C. etunicatum*. Soybean inoculated with *F. mosseae* had the highest concentrations of nitrate-N and ureide-N in the soybean, while those inoculated with *R. intraradices* had the highest relative ureide-N, Ndfa, and the amount of N fixed in soybean (*Table 5*).

Table 4. Mean squares for the effect of P fertilizer application (P) and mycorrhizal inoculation (M) and their interactions (P × M) on the concentration of nitrate, ureide, relative ureide-N, the proportion of N derived from atmosphere, and the amount of N fixed in soybean

	Df	Nitrate N (µmol)	Ureide-N (µmol)	Relative ureide-N (%)	Ndfa (%)	Amount of N fixed (kg ha^{-1})
Reps	1	0.002	2.91	5.33	5.12	30.01
P rates (P)	2	0.029^{ns}	1.07^{ns}	441.3^{ns}	789.9^{ns}	560.3**
Error a	2	0.006	7.26	70.9	110.6	3.80
AMF (M)	3	0.006***	1.03**	211.1**	432.6**	900.1***
P × M	6	0.013**	1.24^{ns}	88.2*	170.5*	253.3**
Error b	9	0.002	8.51	26.0	44.1	30.5
Total	23					

Notes: *, **, ***indicates significant at 0.05, 0.01, and 0.001 probability level respectively; ns indicates not significant.

Table 5. Effects of P fertilizer application (P) and mycorrhizal inoculation (M) on the concentration of nitrate, ureide, relative ureide-N, the proportion of N derived from atmosphere, and the amount of N fixed in soybean

	Nitrate-N (µmol)	Ureide-N (µmol)	Relative ureide-N (%)	Ndfa (%)	Amount of N fixed (kg ha^{-1})
P rates (P)					
0 kg ha^{-1}	0.32	0.028	27.3	27.4	18.7
20 kg ha^{-1}	0.26	0.033	35.3	38.1	32.2
40 kg ha^{-1}	0.20	0.036	42.2	47.3	34.0
Lsd ($p < 0.05$)	ns	Ns	18.1	ns	4.19
AMF inoculation					
Non-AMF	0.37	0.029	26.9	25.8	12.6
F. mosseae	0.24	0.037	38.9	43.2	35.7
R. intraradices	0.22	0.035	39.9	44.2	40.0
C. etunicatum	0.23	0.029	34.1	37.1	24.8
Lsd ($p < 0.05$)	0.07	0.004	6.66	8.67	7.21

Note: ns indicates non-significant differences at p value < 0.05.

There were interactive effects of P fertilizer rates and AMF inoculation on the concentration of nitrate-N, relative ureide-N, Ndfa, and nitrogen fixed in the soybean plants (*Table 4*). The concentration of nitrate-N was observed to significantly reduce with AMF inoculation under zero P (0 kg P_2O_5 ha^{-1}) and low P fertilizer (20 kg P_2O_5 ha^{-1}) application, but no significant difference was observed at high P rate (*Figure 3*). The highest concentration of nitrate-N was observed in uninoculated soybean plants under zero P application (0.49 µmol) and 20 kg P_2O_5 ha^{-1} (0.44 µmol) (*Figure 3*).

The relative ureide-N of AMF-inoculated soybean was observed to significantly increase under zero P and low P application rates compared to non-AMF soybean plants; however, no difference was observed at high P rate (*Figure 4*). Soybean plants inoculated with *F. mosseae* had the highest relative ureide-N at zero P rate, while those inoculated with *R. intraradices* had the highest relative ureide-N at 20 kg P_2O_5 ha^{-1} and 40 kg P_2O_5 ha^{-1} application rates (*Figure 4*).

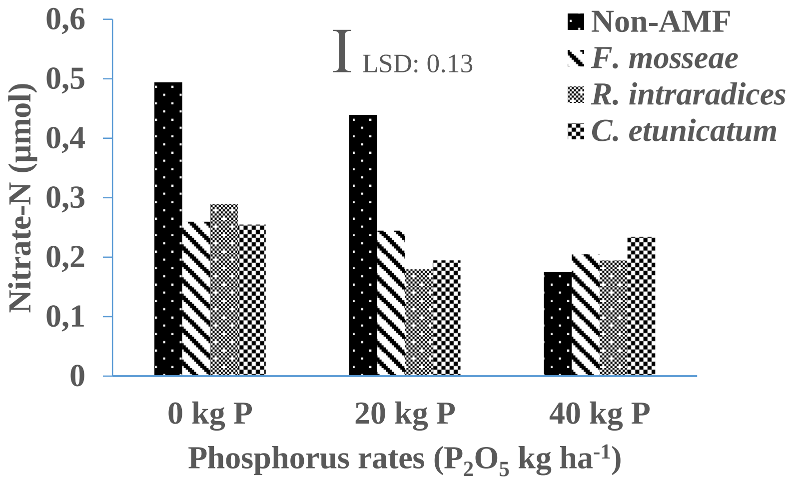


Figure 3. Interactive effect of phosphorus rates and AMF inoculation on nitrate-N of soybean. Bar denotes Lsd value at 5% probability level.

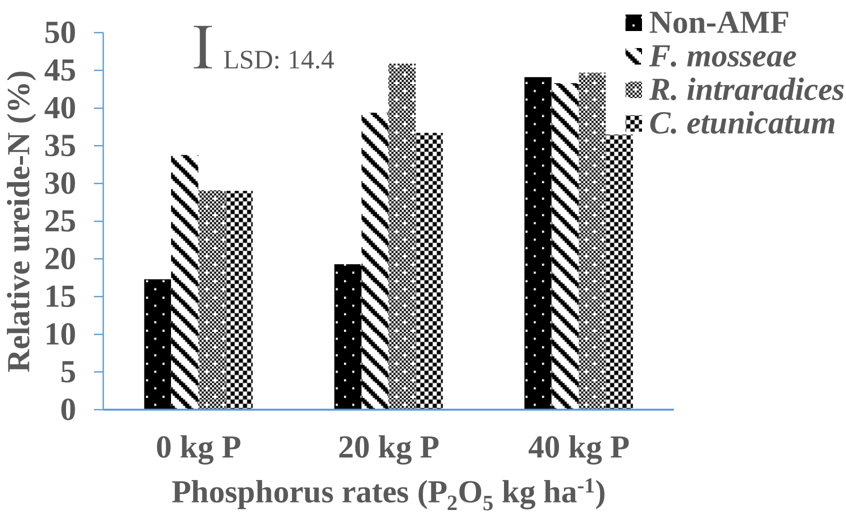


Figure 4. Interactive effect of phosphorus rates and AMF inoculation on relative ureide-N of soybean. Bar denotes Lsd value at 5% probability level.

The Ndfa was recorded to increase with the inoculation of the three AMF isolates under zero P and low P application rates compared to non-AMF soybean plants, while under high P rate no significant difference was observed among the AMF and non-AMF treatments (*Figure 5*). The highest Ndfa was observed in soybean inoculated with *R. intraradices* fertilized with 20 kg P_2O_5 ha^{-1}.

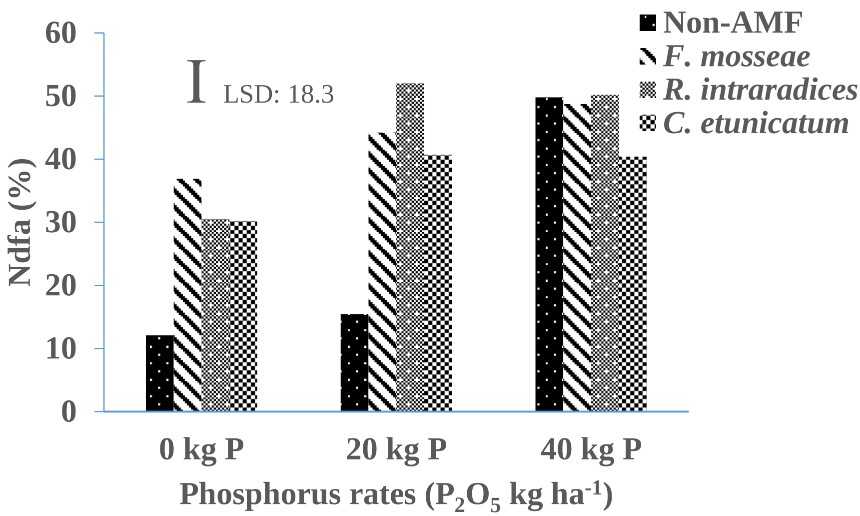


Figure 5. Interactive effect of phosphorus rates and AMF inoculation on N derived from atmosphere (% Ndfa) of soybean. Bar denotes Lsd value at 5% probability level.

Soybean plant inoculated with *R. intraradices* fertilized with 20 kg P_2O_5 ha^{-1} had the highest amount of N fixed (59.4 kg ha^{-1}), while the lowest (2.4 and 3.8 kg ha^{-1}) was recorded in zero P and low P fertilizer application (*Figure 6*).

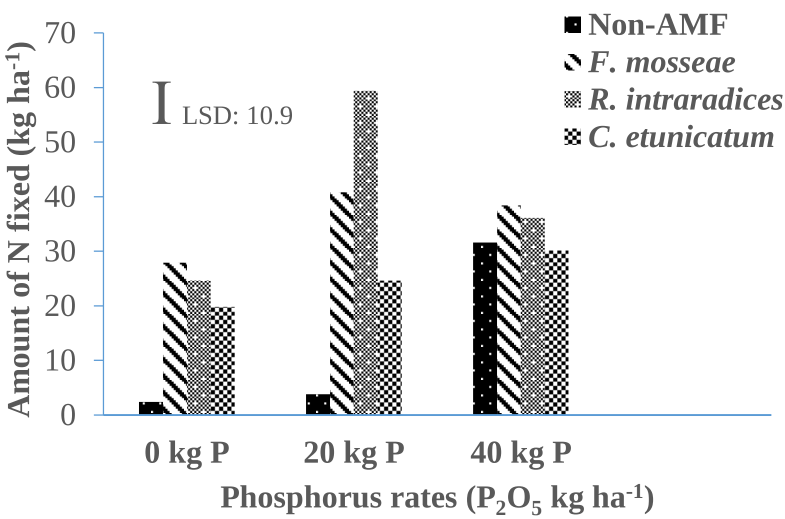


Figure 6. Interactive effect of phosphorus rates and AMF inoculation on the amount of N fixed by soybean. Bar denotes Lsd value at 5% probability level.

AMF root colonization

AMF root colonization was significantly affected by P fertilizer rates, AMF inoculation, and the interactive effect between the treatments (*Table 6*). There was a decrease in AMF root colonization with increase in P rates. The application of 40 kg P_2O_5 ha^{-1} rate significantly reduced the AMF root colonization by 55.1% compared to 20 kg P_2O_5 ha^{-1} (10%) relative to the zero P fertilizer rate; however, no significant difference was observed in the AMF root colonization of the soybean between zero P and 20 kg P_2O_5 ha^{-1} fertilizer rate (*Table 7*). Significant difference was observed among the AMF isolates and non-AMF treatments on the root colonization of the soybean (*Table 6*). The lowest root colonization (15.3%) was observed in non-AMF treatment, while soybean inoculated with *R. intraradices* had the highest root colonization (62.7%), followed by *F. mosseae* (58.7%) and *C. etunicatum* (48%) (*Table 7*).

Table 6. Mean squares for the effect of P fertilizer application (P) and mycorrhizal inoculation (M) and their interactions (P × M) on AMF root colonization, shoot N and P content, and grain and biomass yield of soybean

	Df	AMF root colonization (%)	Shoot N uptake (kg ha^{-1})	Shoot P uptake (kg ha^{-1})	Grain yield (kg ha^{-1})	Biomass yield (kg ha^{-1})
Reps	1	294.0	141.1	206.9	135219	106719
P rates (P)	2	2392.7*	179.8^{ns}	799.8*	2769766*	3421473*
Error a	2	114.0	128.1	17.6	73360	125881
AMF (M)	3	2765.1***	1525.5***	107.7***	998672***	1994264***
P × M	6	165.1***	290.4^{ns}	48.2**	495792***	687066***
Error b	9	14.0	109.0	8.08	14303	63831
Total	23					

Notes: *, **, ***indicates significant at 0.05, 0.01, and 0.001 probability level, respectively; ns indicates not significant.

Regardless of the AMF treatments, the soybean root colonization was reduced at 40 kg P_2O_5 ha^{-1} fertilizer rate (*Figure 7*).

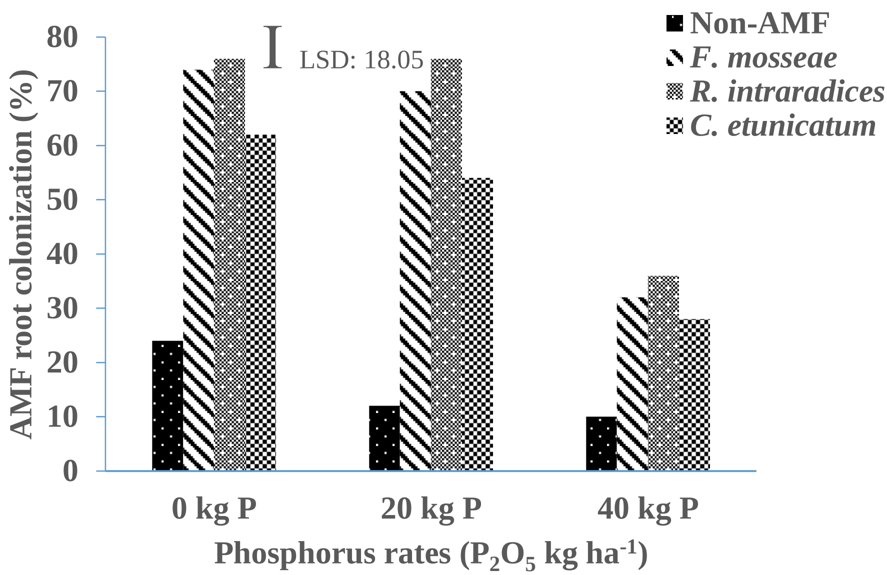


Figure 7. Interactive effect of phosphorus rates and AMF inoculation on AMF root colonization of soybean. Bar denotes Lsd value at 5% probability level.

Shoot N and P content

The soybean shoot N content was significantly influenced by AMF inoculation (*Table 6*). Significant difference was observed among the AMF isolates (*Table 7*). Inoculation with *R. intraradices*, *F. mosseae*, and *C. etunicatum* increased the soybean shoot N content by 59.7, 56.4, and 46.7%, respectively, compared to non-AMF treatment. The shoot P content was significantly influenced by P fertilizer rates, AMF inoculation, and the interactive effect between the treatments (*Table 6*). Shoot P content increased with increase in P rates. Application of 20 and 40 kg P_2O_5 ha^{-1} rates increased shoot P content by 33.2 and 57.6%, respectively, compared to zero P fertilizer rate (*Table 7*). Inoculation with *R. intraradices*, *F. mosseae*, and *C. etunicatum* increased the soybean shoot P content by 32.7, 35.4, and 27.9%, respectively, compared to the non-AMF treatment, but no significant difference was observed among the AMF isolates (*Table 7*). With zero P and 20 kg P_2O_5 ha^{-1} fertilizer rates, the three AMF isolates increased the shoot P content of the soybean compared to the non-AMF treatment, while no difference was observed among the AMF treatments at 40 kg P_2O_5 ha^{-1} fertilizer rate (*Figure 8*).

Table 7. Effects of P fertilizer application (P) and mycorrhizal inoculation (M) on AMF root colonization, shoot N and P content, and grain and biomass yield of soybean

	AMF root colonization (%)	Shoot N uptake (kg ha^{-1})	Shoot P uptake (kg ha^{-1})	Grain yield (kg ha^{-1})	Biomass yield (kg ha^{-1})
P rates (P)					
0 kg ha^{-1}	59.0	40.8	14.5	851	1899
20 kg ha^{-1}	53.0	48.8	21.7	1698	2779
40 kg ha^{-1}	26.5	49.4	34.2	1982	3177
Lsd ($p < 0.05$)	22.9	Ns	9.03	582.7	763.3
AMF inoculation					
Non-AMF	15.3	24.2	17.3	991	1800
F. mosseae	58.7	55.5	25.7	1858	2974
R. intraradices	62.7	60.0	26.8	1813	3067
C. etunicatum	48.0	45.4	24.0	1782	2632
Lsd ($p < 0.05$)	4.89	13.6	3.71	156.2	330.0

Note: ns indicates non-significant differences at p value < 0.05.

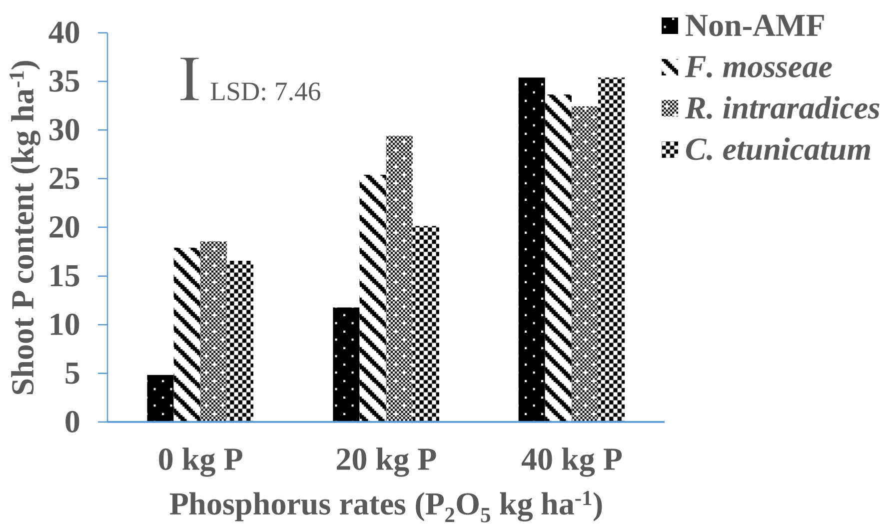


Figure 8. Interactive effect of phosphorus rates and AMF inoculation on the shoot phosphorus content of soybean. Bar denotes Lsd value at 5% probability level.

Grain and biomass yields

The results showed that the grain and biomass yield of the soybean were significantly influenced by P rates and AMF inoculation (*Table 6*). The grain and biomass yields increased with P rates (*Table 7*). Application of 40 kg P_2O_5 ha^{-1} rate increased the grain and biomass yields by 57.1 and 40.2%, respectively, followed by 20 kg P_2O_5 ha^{-1} rate (49.9 and 31.7%) compared to zero P fertilizer rate. Inoculation with *R. intraradices*, *F. mosseae,* and *C. etunicatum* increased the soybean grain yield by 46.7, 45.3, and 44.4%, respectively, compared to the non-AMF treatment, but no significant difference was observed among the AMF isolates (*Table 7*). A similar pattern was observed with the biomass yield (*Table 7*). Significant interactions between P fertilizer rates and AMF inoculation were observed for grain and biomass yield (*Table 6*). The highest grain yield was obtained in soybean inoculated with *R. intraradices* under 20 kg P_2O_5 ha^{-1} application rate, followed by *F. mosseae* under the same P treatment, while the lowest grain yield was recorded in the control plot as shown in *Figure 9*. For the biomass yield, soybean inoculated with *R. intraradices* and *F. mosseae* under 20 kg P_2O_5 ha^{-1} rate application gave the highest biomass yields, with the lowest one observed in the control plot (*Figure 10*).

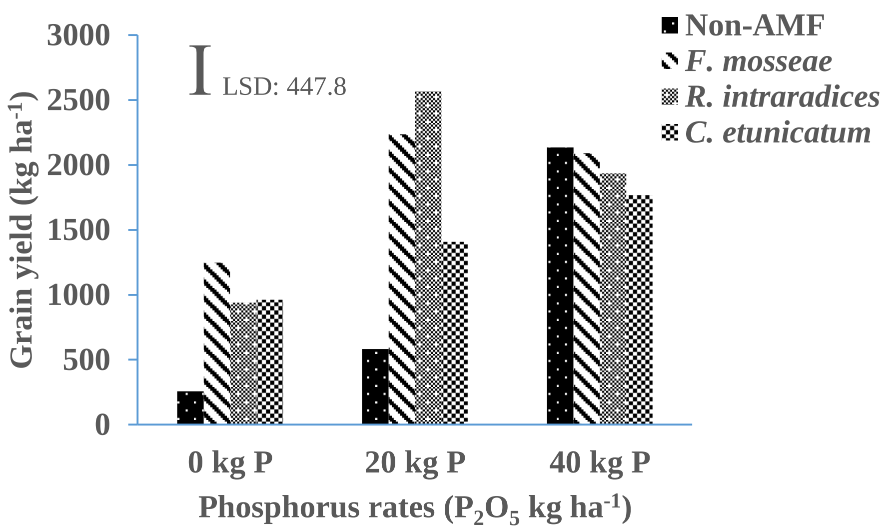


Figure 9. Interactive effect of phosphorus rates and AMF inoculation on the grain yield of soybean. Bar denotes Lsd value at 5% probability level.

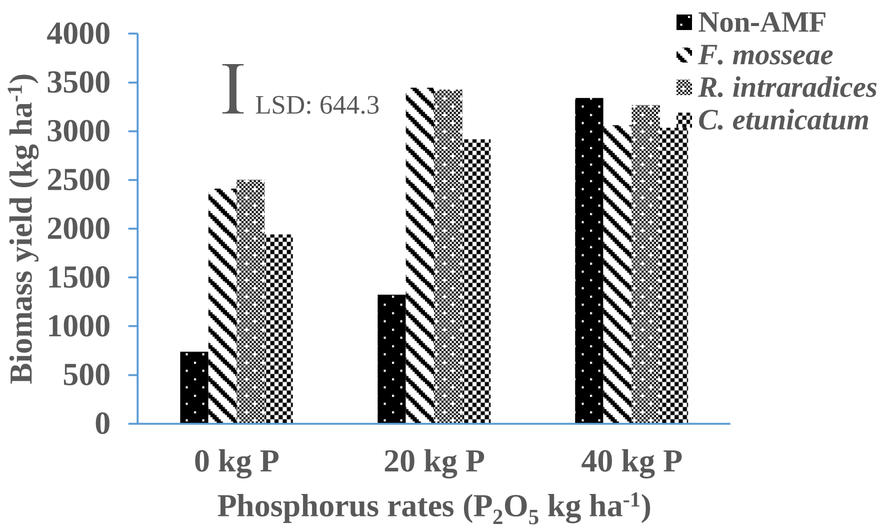


Figure 10. Interactive effect of phosphorus rates and AMF inoculation on the biomass yield of soybean. Bar denotes Lsd value at 5% probability level.

Discussions

The growth and yield performance of soybean improved with P fertilization. The increased shoot dry matter, number of nodules, and seed yield of the soybean are attributed to the increased P supply to the shoot P, which is essential for many metabolic processes such as photosynthesis in the plants. P fertilization in legumes has been reported to increase P content in the nodules and shoot, with increased dry matter accumulation and grain yield [35]. The poor nodulation in terms of number of nodules and the nodule weight of the soybean with low available P could be attributed to the inability of the plant to partition sufficient P for nodule development [36]. This confirmed the reports of [37] that P deficiency reduced the number of nodules and weight in legumes. The relative ureide-N and the amount of N fixed in the soybean plants were improved with P fertilization compared to P-deficient plants in this present study. This is attributed to the increased concentration of ureides and reduced nitrate-N with P fertilization levels. P deficiency has been reported to reduce ureide translocation in the xylem sap, thus reducing nitrogen fixation potential [38]. The increased relative ureides-N with increasing P fertilization resulted in high N_2 fixation observed in this study. Low P availability is reported to reduce nitrate reductase activity in roots, which restricts nitrate transport from roots to shoots in P-deficient soybean plants [39, 40]. Low P availability has been reported to reduce the supply of C from photosynthesis to the nodules [41]. The report of [42] showed that nodule construction cost and growth respiration of different legume plants increased with P deficiency. Furthermore, this study demonstrated increased shoot N content with increased P fertilization levels. This corroborates the evidence from [11], who reported increased N_2 fixation with P fertilization. Based on the above evidence, the low grain and biomass yield of the soybean could be attributed to the reduced dry matter accumulation and nitrogen fixation under P deficiency conditions.

The present study showed the increased root colonization of the soybean in all the AMF isolates used. This suggests the adaptability and higher competitiveness of AMF isolates with the indigenous mycorrhizal population and the soybean cultivar. The positive effects of AMF inoculation on the dry matter and grain yield of the soybean observed in this study corroborate several other reports under field conditions [43, 20]. Several reports showed that AMF promoted plant growth through increased P uptake [44, 45], which also confirmed the result of this study with high shoot P uptake in AMF-inoculated plants compared to the non-AMF control plants. The main benefit of AMF symbiosis with plants is the enhancement of P by the extra-radicular hyphae [14]. The increased N_2 fixation in the soybean plants with AMF inoculation in this study could be attributed to the improved P uptake in mycorrhizal inoculated plants, thus enhancing the transport of ureide-N in the xylem.

The AMF isolates used for this study (*F. mosseae*, *R. intraradices*, and *C. etunicatum*) are generalist AMF widely distributed in most P-deficient soils and can colonize a wide variety of host plants, including legumes [46]. The root colonization of soybean plants by AMF isolates was reduced at high P fertilization levels. High soil P availability has been reported to reduce AMF root colonization and hyphae development [14, 47]. In contrast, the application of P has been reported to promote mycorrhizal development in P-deficient soils [48]. This confirmed the result observed upon the application of 20 kg P_2O_5 ha^{-1} in this study. Among the three AMF tested, *R. intraradices* and *F. mosseae* had the best effect on plant performance in terms of phosphorus uptake, dry matter accumulation, nodulation, nitrogen fixation, and the yield performance of the soybean. The increased growth performance and nitrogen fixation in soybean inoculated with *R. intraradices* and *F. mosseae* could be attributed to their high ability in shoot P uptake [49, 50, 51].

Conclusions

The findings demonstrated the potential of field inoculation with compatible and effective arbuscular mycorrhizal isolates in increasing phosphorus, nodulation, nitrogen fixation, and the grain yield of soybean. Among the three AMF isolates, *Rhizophagus intraradices* and *Funneliformis mosseae* gave higher nodulation and nitrogen fixation of the soybean. Therefore, to ensure higher soybean productivity and sustainable agriculture with reduced high input of chemical P fertilizer through nitrogen fixation, the inoculation of soybean with *Rhizophagus intraradices* or *Funneliformis mosseae* (combined with 20 kg P_2O_5 ha^{-1}) could be recommended, thus ameliorating the negative effect of P deficiency on soybean productivity in the derived savannah of Nigeria. This study was limited to a single soybean variety and a year's field study in one location. It is recommended that further field studies should be undertaken in multi-location sites and different seasons, which will be a significant step towards the stable use of mycorrhizal fungi inoculation for promoting legume productivity in most agricultural soils of Nigeria.

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Seedling and adult plant resistance to *Pyricularia oryzae* in Ethiopian rice cultivars

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Abstract. Two separate experiments were done for seedling and adult resistance in rice varieties against blast. Each experiment consists of 20 varieties and is evaluated under artificial inoculation with blast. The result of the study confirmed that NERICA varieties have shown low disease infection at the seedling stage whereas the varieties Chewaka and Edget have shown adult plant resistance. Severe yield reduction and highly diseased grain were obtained from Superica-1, which is highly susceptible at adult plant stage. In contrast, the maximum grain yield was obtained from the Chewaka and Edget varieties, these having a high level of adult resistance. Therefore, Chewaka and Edget are promising candidates for utilization in yield and blast resistance in rice improvement.

Keywords: adult, blast, seedling, susceptible, resistance, rice

Introduction

Rice (*Oryza sativa*) is one of the most stable foods providing half of the daily calories for the world's population, including African countries [1, 3]. It is the most important source of carbohydrates, vitamins, minerals, and protein for the developing countries [4–7]. The crop has steadily increased in demand and growing area over the last two decades; thus, it becomes evident that it is a strategic commodity in the food security planning policies of many developing countries [8].

In Ethiopia, rice is one of the new target commodities, whose promotion is emphasized and that has played a significant role in the food security of the country [9]. The country has an extensive and suitable ecology for rice production along with the possibility of growing in water-logged areas, where other crops cannot grow. However, the national average yield of rice does not go beyond 2.8 t ha, which is 37.8% below the world average yield [10,11]. There are many constraints affecting rice productivity in Ethiopia. Actual productivity and yield stability are highly influenced by biotic factors, including broad and grass weeds and a number of pathogens.

Rice blast is the most devastating pathogen, which causes a significant reduction in grain yield and seed quality [12]. Infection of the blast can occur in any developmental stage of rice, symptoms of the pathogen being mainly found on leave, node, neck, and panicle. Early blast infection can kill seedlings, while and infection occurring at young plant stages results in leaf blast, neck blast, and panicle blast. The blast causes about 10–20% of yield loss in moderately susceptible varieties, but in severe cases the loss may go up to 80% [13].

To overcome yield loss due to blast pathogen, making use of host resistance and chemicals are the main strategies applied worldwide, including Ethiopia. The application of fungicides is one of the control measures aimed at minimizing yield loss due to blast disease [14, 15]. However, in developing countries, farmers could not use chemicals prior to the occurrence of the disease. The use of a chemical substance is neither practical nor environmentally friendly [16, 17]. The development of cultivars resistant to blast is considered to be the most effective strategy for protecting rice; this is the cheapest and most effective way of controlling rice blast in the fields of resource-poor farmers [18, 19]. Unfortunately, achieving effective and long-lasting blast resistance is restricted because of breakdowns in resistance due to the occurrence of virulent races [20, 21]. Therefore, considerable effort has been made in developing and identifying blast-resistant varieties with the aim of making low-cost blast management procedures available to farmers.

Materials and methods

Inoculum preparation

Isolates were collected from an infected rice variety and then cut into small pieces, containing both the infected and the healthy tissue. Sterilization was done for cut isolates using hypochlorite solution and tap water to eradicate contamination. The cut portions were relocated to oatmeal agar medium and incubated at 25 °C in a moist chamber for 10 days. Ten days after incubation, the surfaces of the isolated colonies were rubbed gently with a paintbrush. The isolated colonies were exposed under fluorescent light for three to four days to reduce sporulation time. The surface of the Petri-dish containing blast isolates was rubbed again with the paintbrush to prepare the conidial suspensions. The conidial suspensions were filtered through filter paper and adjusted to 105 conidia/ml concentration.

Evaluation of seedling resistance

Twenty rice cultivars were individually planted in the seedling plastic pots containing forest soil and were placed in the lath house. The fertilizer was applied at the rate of 3 gram/pot in the form of Nitrogen at planting, and 2 grams were applied 5 days after inoculation. When the plants had fully expanded primary leaves, they were inoculated with leaf blast adjusted at the conidial suspension of 10^5 conidia/ml concentration. The inoculated plants were placed in shade until the growth of the lesion started. Reactions of the inoculated plants were evaluated by the visual observation of the type and severity of the lesions present on leaves and in accordance with the diagrammatic scale described by Urashima and Kato [22]. On each plastic pot, the leaf of each five grown seedlings was evaluated.

Resistance for leaf blast at adult stage

Twenty rice varieties were grown for the assessment of leaf and panicle blast resistance under field conditions. The investigations were carried out at the main station of Bako Agriculture Research Center, Ethiopia, during the main seasons of the 2016–2018 period. Randomized complete block design with three replications was used for the experiments. The fertilizers were applied at the rate of 100 kg/ha in the form of phosphorus, and 100 $kgha^{-1}$ Nitrogen were applied: half at planting and the other half 5 days after inoculation. A seed of each genotype was sown in a plot size of 2.5 m in length, with 6 rows and 20 cm apart. A seed rate of 20 g per plot was used for each variety. A susceptible variety, Superica-1, was used as an infector row for both experiments. Inoculation was done at maximum tillering stage for leaf blast evaluation, and heading for panicle blast was adjusted at the conidial suspension of 10^5 conidia/ml concentration. Disease assessment started from the occurrence of the disease and continued for six observations within seven-day intervals for both leaf and panicle blast. Ten plants were randomly selected from each plot and tagged. Disease rating was done for both pathogens on the tagged plant, based on the standard evaluation system for rice [23].

The area under the disease progress curve (AUDPC) is a better indicator of disease expression over time and is used to determine the levels of resistance of rice varieties to blast in field, leaf, and panicle. Blast severity data were converted to areas under disease progress curves (AUDPC) according to the formula described by Shaner and Finney [24–26].

$$\mathrm{AUDPC} = \sum_{i=1}^{n-1} 0.5(y_{i+1} + y_i)(t_{i+1} - t_i),$$

where a = total number of observation days, t_i = day *I* (time) expressed as number of days after sowing, $t_{(i+1)} - t_i$ = time (days) between two disease observation dates, and y_i = blast severity at i^{th} observation.

Data analysis

Data were subjected to statistical analysis using SAS statistical software (version 9.3) to determine the level of significant difference between varieties. The mean separation was done using LSD (0.05) to facilitate the comparison of all pairs of treatment means. The simple linear association between variables like panicle severity, leaf severity, percentage of deteriorated/infected grain, adult plant severity, and seedling stage severity was determined for 20 rice test materials.

Results

Evaluation of resistance at seedling stage

Based on seedling evaluation, the varieties tested against *Magnaporthe oryzae* were grouped into three categories based on a 0–5 scale as described by Mackill and Bonman [27], i.e. resistant, moderately resistant, and susceptible. The resistant group consists of 6 varieties, which are: Nerica-3, Nerica-12, Nerica-14, Nerica-15, Nerica-18, and Eram-194; this group was found with few types of lesions on some plants. The second group is that of moderate resistance and consists of eleven varieties as follows: Getachew, FOFIFA-3737, FOFIFA-3730, Andassa, Tana, Hidassie, Suparica-1, Nerica-4, Nerica-13, Chewaka, and Edget. The third group (susceptible) consists of three varieties: Kokit, FOFIFA-4129, and X-Jigna. This group consists of large-sized and a high number of lesions and is characterized by the rapid reproduction of the pathogen (*Table 1*).

Table 1. Type of reaction and infection of rice varieties at seedling stage against *Pyricularia oryzae* in glass house

Varieties	Type of infection	Type of reaction	Varieties	Type of infection	Type of reaction
Suparica-1	3	MR	Nerica-14	2	R
Edget	3	MR	Andassa	3	MR
Nerica-3	2	R	Nerica-12	3	R
Nerica-4	3	MR	Getachew	3	MR
Nerica-15	2	R	Nerica-13	3	MR
Nerica-18	2	R	FOFIFA-4129	4	S
Tana	3	MR	Eram-194	2	R
Hidassie	3	MR	FOFIFA-3737	3	MR
X-Jegna	4	S	Chewaka	3	MR
FOFIFA-3730	3	MR	Kokit	4	S

Notes: infection types were measured based on a scale of 0.0 to 5.0 (Mackill and Bonman, 1992)
R = resistant, MR = moderately resistant, and S = susceptible

Evaluation of resistance for leaf and panicle at adult stage

Analysis of variance for final leaf and panicle blast severity score (FSC) showed a significant difference in the studied rice cultivars (*Table 2*). The mean comparison of leaf blast severity (LBS) showed that the Edget and Chewaka varieties had the lowest values with 1.8 and 2.0 rating score, respectively, while, Nerica-3(5), Nerica-4(5), Nerica-14 (5), and Nerica-15(5) had the highest leaf blast severity score (*Table 2*). Mean comparison of panicle blast severity (PBS) showed that Edget and Chewaka yielded low severity scores with 3.0 and 3.4 score, respectively, while the highest ($\geq$ 7) severity scores were obtained for Nerica-14, Nerica-15, Nerica-4, Nerica-3, and Fofifa-3037 (*Table 2*).

The ANOVA obtained from the estimated areas under disease progress curves (AUDPC) for both leaf and panicle blast significantly varied in the studied rice varieties (*Table 2*). The highest estimated AUDPC value for leaf blast was recorded for Nerica-14 (533.3), followed by Nerica-15 (523.0) and Superica-1 (520.0), while the lowest values were obtained for Edget (266.6) and Chewaka (380.0). Similarly, the maximum mean value of AUDPC from panicle blast was obtained for Nerica-14 (1861.9) and Superica-1 (1823.8), whereas the lowest values were obtained for Edget (895.2) and Chewaka (955.2) (*Table 2*).

The analysis of variance for grain yield showed significant ($P < 0.05$) differences among the varieties (*Table 2*). The highest grain yield was obtained for Chewaka (4213.3 kg ha^{-1}), followed by the Edget (2625.3 kg ha^{-1}) variety, both varieties showing high levels of adult resistance to both leaf and panicle blast. In contrast, low yield was obtained for Nerica-14 (202.7 kg ha^{-1}), followed by Superica-1-1 (606.0 kg ha^{-1}), which were susceptible to both pathogens and highly susceptible to panicle blast (*Table 2*).

Table 2. Infection type (leaf blast), panicle blast severity, area under the disease progress curve (AUDPC), leaf and panicle blast, and grain yield for rice varieties in field conditions

Varieties	Area under progress curve (AUDPC)		FLBS (1–5 scale)	Host response to leaf blast	FPBS (1–9 scale)	Host response to panicle blast	Grain yield (Kg/ha)
	leaf blast	panicle blast					
Nerica-14	533.3a	1861.9a	5.0a	S	7.8a	S	202.7fe
Superica-1	520.0a	1823.8ba	4.9ba	S	6.6bac	S	660.0dfe
Xchegna	516.3a	1819.0ba	4.7ba	S	7.0ba	S	1172.0dfe
Nerica-15	523.0a	1795.2ba	5.0a	S	7.5a	S	272.0fe
Nerica-3	506.6ba	1790.5ba	5.0a	S	7.3ba	S	1233.3dfe
Andassa	514.3a	1781.0ba	4.6bac	S	7.0ba	S	1792.0dc
Tana	500.0ba	1780.9ba	4.5bac	S	6.6bac	S	1441.3dce
FOFIFA-3037	505.6ba	1776.2ba	4.8ba	S	7.2ba	S	1316.0dfe
Nerica-4	521.0a	1771.4ba	5.0a	S	7.4ba	S	682.7dfe
Nerica-12	506.6ba	1757.2ba	4.8ba	S	6.6bac	S	827.3dfe
FOFIFA-3737	500.0ba	1757.1ba	4.9ba	S	6.3bac	S	931dfe
Getachew	5003.0ba	1733.3ba	4.6bac	S	6.3bac	S	920.3dfe
Hiddasie	502.0ba	1704.8bac	3.6bdc	MR	5.0bc	MR	1000.0dfe
Nerica-18	473.3bac	1604.8bac	4.3bac	S	6.0bac	S	102.7f
Nerica-13	466.6bac	1585.7bac	4.7bac	S	5.0bc	MR	340.0fe
Koki	440.0bac	1571.4bac	3.8bdc	MR	4.4bc	MR	1753.3dc
Eram-194	446.6bac	1571.4bac	4.7bac	S	6.2bac	S	1108.0dfe
FOFIFA-4129	440.0bac	1557.1bac	4.0bdc	R	6.5bac	S	801.3dfe
Chewaka	380.0dc	955.2d	2.0d	R	3.4d	R	4213.0a
Edget	266.6d	895.2d	1.8d	R	3.0d	R	2625.3bc
Lsd	118	361	1.08		1.6		1279.5
CV	15.1	13.2	14.3		12.2		15
F-test	0.01	0.001	0.01		0.004		0.002

Notes: Means followed by the same letter within the column are not significantly different at 5% probability level. LSD = least significant difference, CV = coefficient of variation, P = probability, ** = highly significant ($p < 0.01$), * = significant ($p < 0$ .05), ns = non-significant, FLBS = final leaf blast severity, FPBS = final panicle blast severity, R = resistant, MR = moderately resistant, and S = susceptible.

Rice grain deterioration due to panicle blast

Results of the study revealed that the grains of Chewaka and Edget were less deteriorated compared with Nerica and other tested varieties (*Fig. 1*). Significantly lowest percentage of infected/deteriorated grains was obtained for Chewaka (20.4%) and Edget (21%), whereas the highest percentage of infected grains was recorded for NERICA varieties (*Fig. 1*).

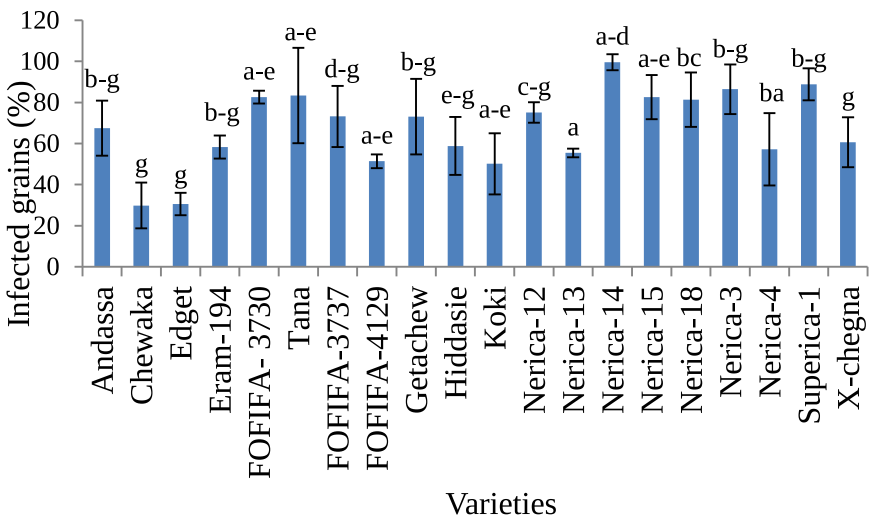

Figure 1. Percentage of infected grains for 20 rice varieties due to panicle blast

Analysis of correlation coefficient

The simple linear association between variables, such as severity scale due to leaf blast at seedling and adult plant stage, area under disease progress curve for leaf and panicle blast, or percentage of deteriorated grain due to panicle blast, were determined for 20 rice cultivars (figures 2–5). The percentage of affected (diseased) grains was significant and was found in positive correlation with the final severity score of panicle blast ($r = 0.64$, $P \leq 0.05$) (*Figure 2*). The final severity score of leaf blast had a negative and non-significant association with severity score at seedling stage ($r = -0.38$, $P \leq 0.05$) (*Figure 3*). Panicle severities were negative and highly significantly correlated with diseased grain ($r = -0.77$, $P < 0.01$). The estimated AUDPC from leaf blast had a positive and highly significant correlation with AUDPC calculated for panicle blast ($r = 0.908$, $P \leq 0.01$) (*Figure 5*).

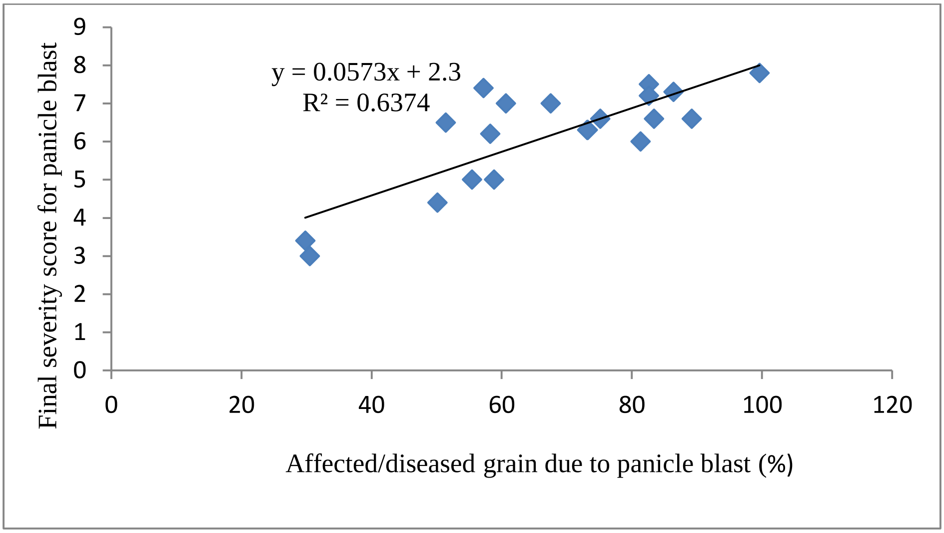

Figure 2. Correlation between the percentage of diseased/infected grain and panicle blast for 20 rice varieties (significant by student's t-test at 5% probability)

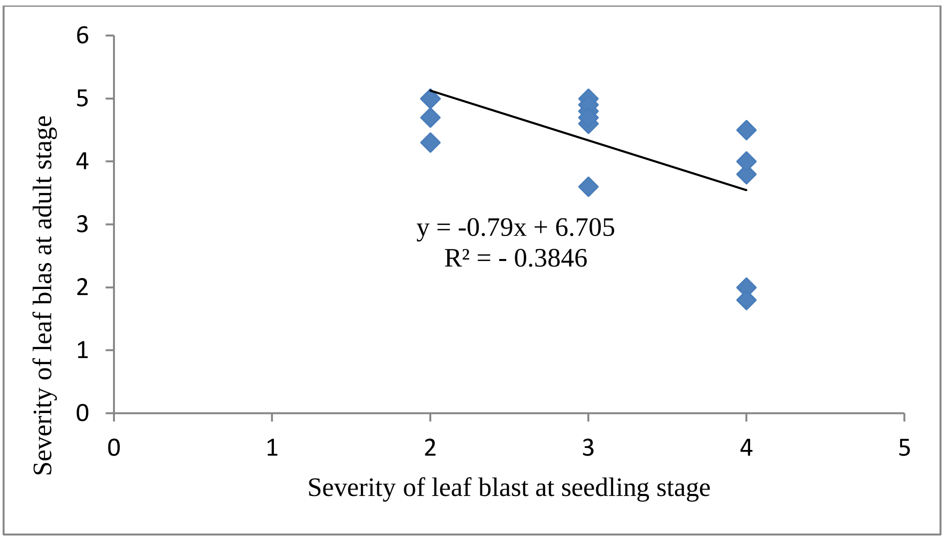

Figure 3. Correlation between blast infection at seedling and adult plant stage for 20 rice varieties (non-significant by student's t-test at 5% probability)

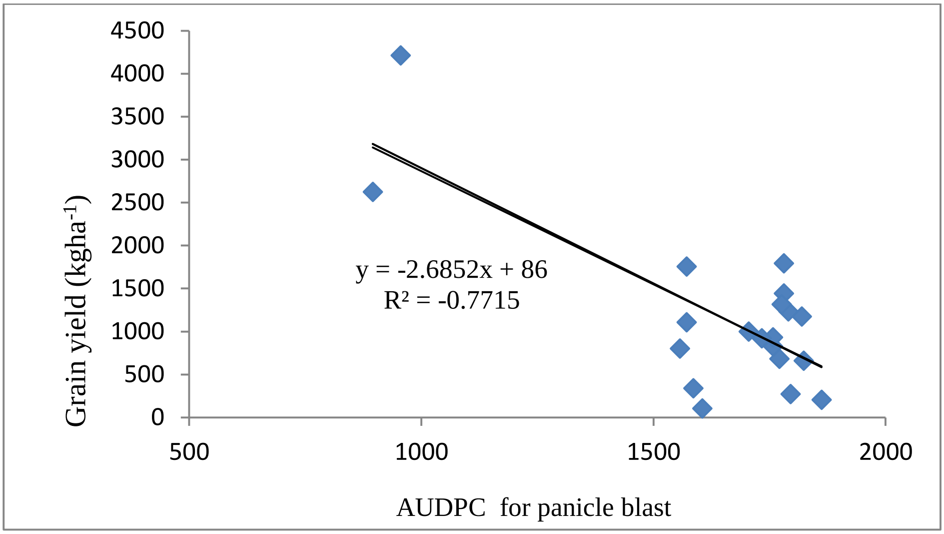

Figure 4. Correlation between AUDPC and grain yield for 20 Rice varieties (significant by student's t-test at 5% probability)

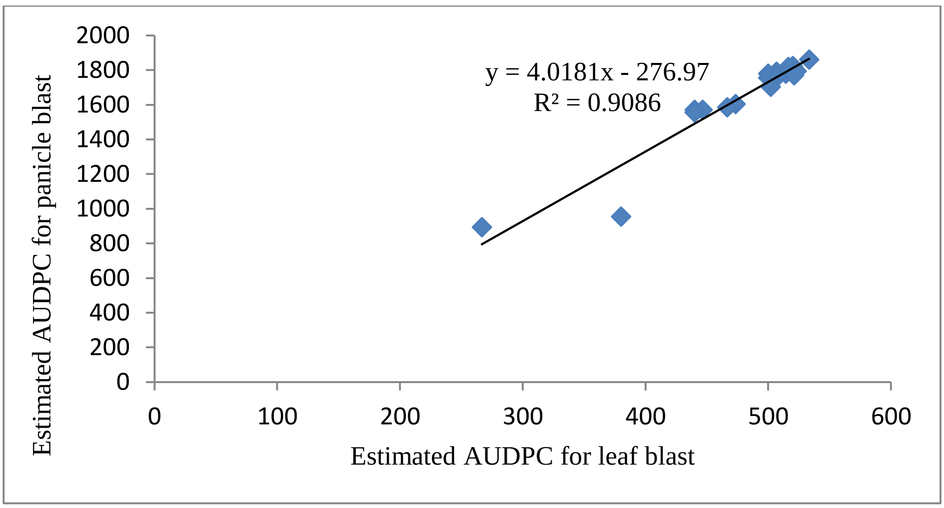

Figure 5. Correlation between AUDPC and panicle blast for 20 rice varieties (significant by student's t-test at 5% probability)

Discussion

Evaluation results of seedling resistance in rice varieties to *Pyricularia oryzae* revealed that NERICA-3, NERICA-12, NERICA-14, NERICA-15, NERICA-18, and Eram-194 showed low infection types (ITs “1”–“2”) based on a 0–5 scale as described by Mackill and Bonman [27]. These varieties yielded small numbers of sporulation lesions and reduced spore reproduction, suggesting that they may have complete resistance genes against blast. Complete resistance minimizes the entrance of the pathogen through developing anti-pathogen by the plant and reduces spore reproduction and sporulation lesions [28–29]. This interaction is under a gene-for-gene control [30]. Getachew, FOFIFA 3737, FOFIFA 3730, Andassa, Tana, Hidassie, Superica-1, Nerica-4, Nerica-13, Chewaka, and Edget varieties showed moderate infection types, while Kokit, FOFIFA 4129, and X-Jigna showed high infection types (susceptible reaction) (ITs “4”–“5”) against blast at seedling stage. Thus, these varieties showed low levels of resistance at seedling stage.

The field evaluation for adult plants indicated that NERICA 3, NERICA 4, NERICA 12, NERICA 14, NERICA 15, and NERICA 18 had low levels of resistance to leaf and panicle blast, while Chewaka and Edget had the lowest infection rate and AUDPC value for both types of blast, which showed a high level of resistance to leaf and panicle blast. This indicates an increase in blast severity corresponding with an increased area under the disease progress curve. The Chewaka and Edget varieties showed a slow development of lesions, low disease pressure, and fewer as well as smaller blast lesions, which reduce the extent of pathogen reproduction in the compatible interaction. These suggestions are in agreement with Bonman [31] and Elsa [32], who reported the nature of partial resistance, the compatible lesion types but also the low level of disease pressure and fewer and smaller blast lesions than with fully susceptible cultivars and that spore production may be reduced.

These findings confirmed that Chewaka and Edget had moderate blast infection at seedling stage compared with lower ones (2.8–3.0) at adult stage. Some contend that seedling resistance is effective at all growth stages, while some others argue that resistance is effective at adult or seedling growth stages only [33]. Results of linear relationship imply that rice blast infection at seedling stage is negative and is non-significantly correlated with adult stage ($r = -0.38$, $P < 0.05$). According to Qi [34], increase in resistance with increase in growth stage might be due to: morphological and physiological differences in leaf tissues, latent period-prolonging genes may not be expressed in seedling stage but are better expressed in adult plant stage. Koizumi et al. [35] reported similar results.

Results of the study indicated differences in yield reduction among rice cultivars that were dependent on leaf and panicle blast development. The highest yield reduction occurred in the variety Superica-1, which is highly susceptible to both leaf and panicle blast, while Chewaka and Edget had high levels of adult resistance to both leaf and panicle blast. Similarly, Sim et al. [36] and Charles [37] reported that severe yield reduction was caused by severe leaf and panicle blast, including neck rot, resulting in complete yield loss. Rice blast disease severity for panicle was found positive and highly significantly correlated with grain yield reduction. This indicates that panicle blast disease severity was directly related to grain yield losses. The finding that an increase in leaf or panicle blast disease severity corresponds with an increase in grain yield losses has also been reported by Shim et al. [38] and Charles [37].

Severe infection of rice panicle by *Pyricularia oryzae* has also been considered a major cause of reducing the grain quality of rice by reducing the percentage of ripe spikelets and the percentage of fully mature grains [12, 36, 38]. This suggestion supports the current studies that grains of the varieties susceptible to panicle blast, such as Nerica-14, Nerica-13, Nerica-15, and Nerica-18, were highly affected due to panicle blast. The lowest percentage of diseased grain was obtained for Chewaka and Edget. The relationship between panicle blast disease severity and the percentage of diseased/affected grains indicated that an increase in disease severity resulted in a simultaneous increase in the diseased/affected grain. Panicle severities were positive and significantly correlated with diseased grains.

Conclusions

In conclusion, the rice varieties Chewaka and Edget were recorded to have superior grain yield, had effects on the reduction of disease development rate and the lower number and size of blast spots on the leaves/panicle, and minimized grain deterioration. Therefore, these two varieties are promising solutions, which could be utilized in yield and blast resistance breeding programmes in western Ethiopia.

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Genetic variation in common bean (*Phaseolus vulgaris* L.) using seed protein markers

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Abstract. The genetic diversity of common bean accessions were assessed using seed storage protein markers. At regional level, accessions from the two major growing regions showed the highest level of gene diversity (H = 0.322, I = 0.485, and H = 0.312, I = 0.473), which can be exploited for the future improvement of the crop. Based on phaseolin, the major storage protein in common bean, the majority of the accessions (86%) were grouped under Mesoamerican gene pool. Seed proteins were also used to differentiate various Phaseolus species, indicating the usefulness of seed storage proteins in species identification in this genus.

Keywords: diversity, phaseolin, SDS-PAGE, seed protein

1. Introduction

Common bean (*Phaseolus vulgaris* L.) is among the cultivated bean types belonging to the genus Phaseolus. Due to long storage life, good nutritional properties, and requirements of easy storage and preparation, common bean has high value in the developing world [16]. It contains proteins (15%), starch (80%), and fats (2%); it provides folic acid, dietary fibre, and complex carbohydrates; some parts of the plant also have a therapeutic value [23].

Common bean is one of the major export commodities and a cash crop for small-scale farmers in Ethiopia, and it supplies about 60% of the total export of pulses [25]. Produced by about 3.38 million smallholders, it covered 21.6% of the total pulse growing area in the 2015/16 cropping season. The regions of

Oromia, Southern Nations Nationalities and Peoples' Region (SNNP), and Amhara are the major growing areas in the country [7]. Pests and diseases, lack of access to improved germplasms, and unreliable climatic conditions are among the constraints encountered by smallholder farmers that result in the low agricultural productivity of common bean [4].

Studying the diversity in common bean is useful to generate information that can be used in crop improvement and genebank management. Genetic diversity in common bean has been carried out using morphological markers [6], isozymes, seed proteins [3, 24], and various types of molecular markers [1, 2, 10, 18].

Seed storage protein fractions are mixtures of components which show polymorphism both within and among genotypes of the same species [22]. Bean seeds contain 20% to 25% proteins, dominated by the storage protein phaseolin [4], which determines both the quantity and nutritional quality of proteins in bean seeds [5, 12]. Polymorphism, environmental stability, and biochemical complexity characteristics enable phaseolin to be an informative marker [11]. Genetic diversity of common bean germplasms from Ethiopia has been carried out using inter-simple sequence repeat (ISSR) markers [8] and simple sequence repeat (SSR) markers [10]. To the knowledge of the authors, seed storage protein and phaseolin types have not been utilized to study Ethiopian Phaseolus collections. Hence, the present study was undertaken to analyse the suitability of seed storage protein for diversity assessment in Phaseolus collections of Ethiopia. It also examines the potential of seed storage proteins in discriminating different Phaseolus species and identifies phaseolin types observed in the analysed accessions.

2. Materials and methods

A total of 50 common bean accessions obtained from the Ethiopian Biodiversity Institute were used for this study (*Table 1*). Three accessions (240523, 235507, and 235506) conserved as *P. acutifolius*, one accession each conserved as *P. lunatus* (211481) and *P. sativus* (241742) were also included in the analysis. Fifteen seeds per accession were ground to fine powder with mortar and pestle. Seed proteins were extracted using 0.002M borate buffer. Protein profiling of extracted samples was analysed using sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE) in 10% polyacrylamide gel [13]. Electrophoresis was carried out at constant voltage (100 V). At the end of the run, gels were stained with staining solution (40% ethanol, 10% acetic acid) containing 0.1% (w/v) Coommassie brilliant blue R-250 for overnight. Destaining of gels was carried out using 40% ethanol and 10% acetic acid.

The presence (1) or absence (0) of every band was scored in reference to a standard protein ladder (NEB P7712S). The resulting binary data matrix for the 50 accessions (750 individuals) was used to perform the data analysis. GenAlEx version 6.5 [19] was used to calculate the percentage of polymorphic bands, heterozygosity, and diversity index. The relationship among the analysed samples was revealed by cluster analyses from the binary data using NTSYS v 2.1 [21]. To complement the information on clustering, principal coordinate analysis (PCoA) was conducted using GenAlEx software [19].

3. Results and discussions

Genetic diversity

On the basis of the relative mobility of seed proteins on the gel, a total of 41 reproducible bands were detected. The size of the protein bands detected ranged from 11 to 210 kDa. The Phaseolus accessions under study showed variations in total number of bands, which ranged from 14 to 33. The percentage of polymorphic bands ranged from 2.44% (accession 5) to 58.54% (accession 8) and averaged 27.05%, which revealed a wide and diverse genetic base in common bean accessions collected from different regions. High percentages of polymorphic loci were observed in Ac8, which was collected from the Oromia region (Mirab Harerge zone) and Ac41 from the Benishangul-Gumuz region (Metekel zone) with a value of 58.54% and 51.22% respectively (*Table 1*). The least band polymorphism was found in Ac5 (2.44%) collected from SNNP (Sidama zone). The highest gene diversity was shown by Ac8 (H = 0.213, I = 0.318), and the least gene diversity was observed in Ac5 (H = 0.002, I = 0.004). In general, at accession level, the diversity estimates show low level of variation within each accession (mean I = 0.135, mean H = 0.090). Using SSR markers, previous studies reported higher level of diversity among accessions collected from different regions of Ethiopia [1, 10], showing the limited potential of seed protein markers in revealing variation within each accession.

Table 1. Summary of parameters for genetic diversity assessment in common bean accessions: Accession code (code); Accession number (Ac No.); Region code (RC); percentage of polymorphic bands (PB), Heterozygosity (H), and diversity index (I)

Code	Ac No.	RC	PB	H	I	Code	Ac No.	RC	PB	H	I
Ac1	241756	1	17.07	0.058	0.087	Ac26	211338	2	7.32	0.020	0.031
Ac2	241742	1	26.83	0.103	0.150	Ac27	214664	1	4.88	0.014	0.020
Ac3	241737	1	17.07	0.063	0.094	Ac28	244805	1	7.32	0.033	0.047
Ac4	241738	1	41.46	0.175	0.253	Ac29	207935	3	24.39	0.084	0.126
Ac5	241733	1	2.44	0.002	0.004	Ac30	211277	1	26.83	0.064	0.103
Ac6	241734	1	17.07	0.049	0.075	Ac31	211282	1	21.95	0.057	0.089
Ac7	237080	2	9.76	0.048	0.067	Ac32	211276	1	24.39	0.077	0.118
Ac8	241134	2	58.54	0.213	0.318	Ac33	211278	1	12.20	0.049	0.072
Ac9	237993	1	17.07	0.073	0.105	Ac34	211376	2	24.39	0.085	0.128
Ac10	241736	1	41.46	0.107	0.167	Ac35	211266	5	31.71	0.090	0.140
Ac11	241748	1	17.07	0.048	0.075	Ac36	208639	2	31.71	0.091	0.142
Ac12	212861	2	26.83	0.089	0.134	Ac37	211322	2	26.83	0.092	0.138
Ac13	241739	1	31.71	0.102	0.155	Ac38	208637	2	43.90	0.138	0.210
Ac14	237079	2	24.39	0.084	0.126	Ac39	212860	2	19.51	0.059	0.090
Ac15	230044	2	36.59	0.147	0.214	Ac40	211313	2	26.83	0.084	0.128
Ac16	215048	2	31.71	0.101	0.153	Ac41	211344	3	51.22	0.150	0.230
Ac17	207943	2	41.46	0.140	0.211	Ac42	208645	2	17.07	0.053	0.082
Ac18	207940	3	36.59	0.086	0.138	Ac43	208705	2	39.02	0.114	0.174
Ac19	222872	4	48.78	0.142	0.219	Ac44	211340	2	26.83	0.091	0.137
Ac20	228911	2	34.15	0.105	0.159	Ac45	207933	3	26.83	0.091	0.135
Ac21	214665	1	21.95	0.082	0.119	Ac46	228912	2	31.71	0.104	0.156
Ac22	208638	2	29.27	0.109	0.160	Ac47	201293	2	24.39	0.094	0.137
Ac23	211386	5	36.59	0.125	0.184	Ac48	211345	3	21.95	0.071	0.107
Ac24	211378	2	39.02	0.136	0.204	Ac49	211265	5	17.07	0.067	0.098
Ac25	211355	3	36.59	0.140	0.205	Ac50	235697	2	21.95	0.088	0.128
								Mean	**27.05**	**0.090**	**0.135**

Note: 1 – SNNP, 2 – Oromia, 3 – Benishangul – Gumuz, 4 – Gambella, 5 – Amhara

Diversity estimates obtained by grouping populations based on their geographic origin (region) revealed that accessions from Oromia and SNNP regions showed the highest percentage of polymorphic bands with a value of 97.56% each, followed by Benishangul-Gumuz (87.80%), Amhara (73.17%), and the least one was from Gambela region (48.78%). The highest diversity estimates were shown in Oromia (H = 0.322, I = 0.485) and SNNP (H = 0.312, I = 0.473) regions. Accessions from Oromia and SNNP were highly diverse in all the variability measures.

This agrees with the findings of Fisseha et al. [10], who reported high diversity on common bean accessions collected from these regions using SSR markers.

Similarly high diversity on common bean accessions collected from these regions was reported using SSR markers [10]. Hence, these two regions – which are also the most important regions in terms of common bean production in Ethiopia [25] – could be the most important regions for identifying important genes for the breeding and improvement of common bean.

On the basis of pair-wise genetic similarity matrix, the most distantly related accessions were Ac20 and Ac47, which were collected from the Oromia region, Illubabbor and Mirab Harerge zones, respectively, with a value of 0.751, indicating a wide range among the analysed accessions (data not shown). The most closely related accessions were Ac14 and Ac29 (GD = 0.00), which were collected from the Oromia and Benishangul-Gumuz regions respectively. The analysis of genetic distance among accessions pooled by region of collection revealed that the most distantly related accessions were the ones from SNNP and Gambela with a value of 0.085, and the least one was between Oromia and SNNP with a value of 0.012. Genetic distance is an important parameter for germplasm improvement, allowing the exploitation of distantly related populations, which may result in vigorous varieties that combine the traits of the distantly related parents.

Grouping of accessions based on similarity can be seen in *Figure 1*. The analysed accessions can be divided into three main clusters with accession 20 from the Oromia region, Illubabor zone shown as an outlier at coefficient of about 0.56. Accession 14 from Oromia and 29 from the Benishangul-Gumuz regions were the most similar ones (genetic similarity (GS) = 1.0), followed by accession 42 and 46 both from the Oromia region (GS = 0.86). Group I and II can be further divided into subclusters.

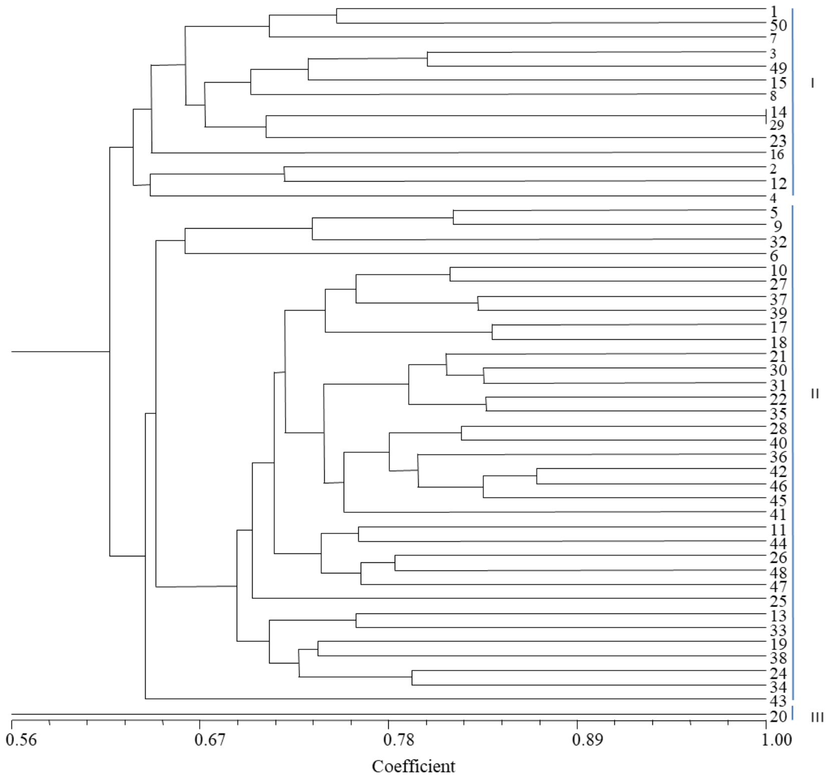

Figure 1. Dendrogram-compiled simple matching coefficient similarity coefficient showing the grouping of the analysed common bean accessions based on seed protein profile

The association among the analysed genotypes examined by principal coordinate analysis (PCoA) showed that the first three axes explained a cumulative variation of 60.03%. Individuals from an accession tend to group together; however, accessions from the same regions or zones did not show a specific grouping pattern (*Fig. 2*). In both PCoA and cluster analysis, accessions from different collection regions were grouped together, which may imply the exchange of plant materials and the possibility of gene dispersal by seeds in different common-bean-growing regions and zones of Ethiopia.

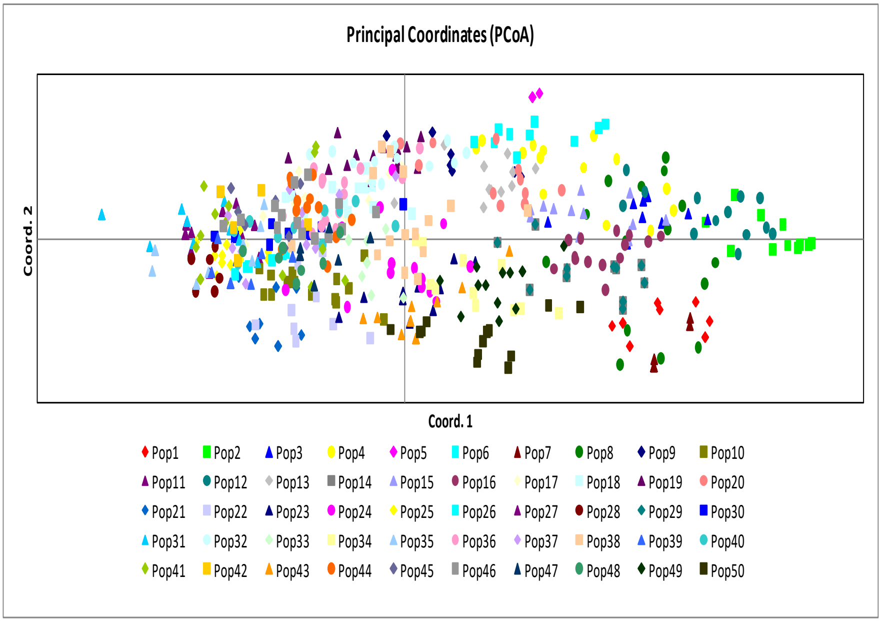

Figure 2. Principal coordinate analysis of 50 common bean accessions analysed by seed storage proteins

Diversity in phaseolin patterns

The protein profiles were also used to discriminate the existing variability by phaseolin patterns. Studies on phaseolin type determination allow researchers to understand the range of dispersal of bean genotypes from their original location to their secondary location [15]. Phaseolin protein has a narrow range of molecular weight (42–53 kDa), as reported by different researchers. The molecular weight for Phaseolin zone in the present study ranged from 42 kDa to 49 kDa (*Fig. 3*), which agrees with the findings reported by Madakbas et al. [15] and Tomlekova et al. [24]. This zone is used to determine the grouping of the accessions among the two centres of origins – Mesoamerican and Andean –, which are considered to be the primary centres of origin for common bean [11, 12].

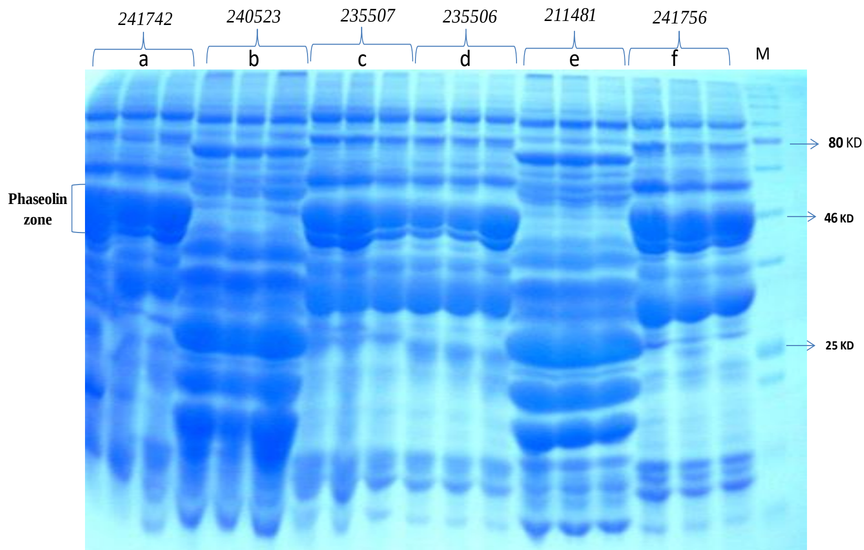

Figure 3. Similarity/difference among phaseolus species using seed protein profile: P. vulgaris (a, c, d), P. lunatus (b, e), f-control (P. vulgaris), M-molecular weight marker

There were three variants on this zone: three bands between 42 kDa and 49 kDa in the Andean phaseolin type (*Fig. 4*, samples c, d, e) and two bands in the Mesoamerican phaseolin type. The majority of the accessions analysed in this study were grouped under the Mesoamerican gene pool (86%) since they contained Mesoamerican phaseolin type, and seven accessions (Ac16, Ac17, Ac18, Ac32, Ac35, Ac36, and Ac44) contained samples that showed the Andean phaseolin type. According to Asfaw et al. [1], both the Mesoamerican and Andean gene pools are present in Ethiopia, with a higher frequency for the Mesoamerican type. Among East African countries, Andean genotypes are dominant in Kenya [1], while in Uganda the Andean and Mesoamerican genotypes occur with similar frequency [17]. The existence of both gene pools and the low proportion of Andean type common beans in Ethiopia may be due to the original introductions, subsequent imports of novel germplasm from various sources, the low level of adaptation of Andean types to the ecological conditions in Ethiopia, consumer preferences, and the occurrence of biotic and abiotic stresses which did not favour the Andean types [1, 27].

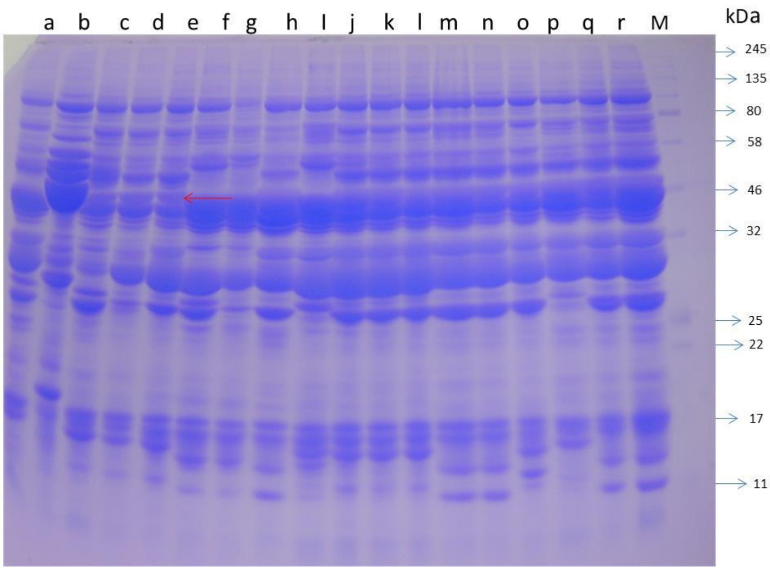

Figure 4. SDS-PAGE of total seed protein extracts of P. vulgaris samples.
Samples c, d, and e show T-phaseolin patterns (indicated by arrow).
M – protein weight marker

Assessment of the application of seed proteins for species identification in the phaseolus collection showed different banding patterns among the different species (*Fig. 3*). Three accessions were conserved as *P. sativus* and *P. acutifolius*. However, their banding pattern was similar to that of *P. vulgaris*. One accession conserved as *P. acutifolius* showed similar seed protein banding pattern with *P. lunatus*. These results were also supported by seed morphology and the examination of banding patterns using ISSR markers. Studies based on the electrophoretic analysis of seed proteins have been used to discriminate species and cultivars in other legumes as well. Seed protein profiles of different Lathyrus species showed unique electrophoresis patterns [9], and different electrophoretic seed albumin patterns were observed in different Lathyrus species [20]. In *Lupinus albus*, glutelins and glycoproteins were successfully used for cultivar identification [26], indicating the possible use of seed storage proteins in species identification in phaseolus and other legume species.

4. Conclusions

This study has examined genetic diversity in common bean using seed storage proteins. These markers show a low level of diversity within accessions, but a substantially higher level of diversity was observed among regions, which can be exploited for genetic improvement and further germplasm collection of the crop. Species identification using only plant morphology could result in misclassification of species. The study has also demonstrated that seed storage proteins, which are relatively inexpensive markers, can be employed in the identification of phaseolus species and genebank management.

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The analytical assessment of the weaknesses of the agriculture of Szabolcs-Szatmár-Bereg County based on empirical research results

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Abstract. In our empirical research, we examined the agriculture of Szabolcs-Szatmár-Bereg County. We surveyed the situation of agriculture through farmers using certain criteria to rank the situation within the framework of a SWOT analysis. The responses received from farmers in the county were quantified and evaluated on the basis of what these farmers have considered to be true. The interdisciplinary study of agriculture is a timely and important task. It can be enforced on the basis of systemic contexts studying not only food production but also environmental issues, preservation of rural habitats, employment, and regional development.

Keywords: system approach, situation assessment, interdisciplinary study, agriculture, regionalism

1. Introduction

Agriculture, as a product-producing and value-creating activity, plays a special but increasingly diminishing role in the structure of the Hungarian economy and thus in its overall performance. Based on its fundamental potential, it influences its processes and trends and defines both food production and environmental protection.

In this systematic approach, the most important question, if not the only one, of agriculture and humanity is our ability to preserve ecological and economic diversity, the natural quantity and diversity of our natural resources, and the richness of the flora and fauna [1]. In an environment under increasing pressure, such as a resource crisis, population explosion, ensuring healthy food production, energy demand, a proper way of life, and job security in rural areas, the technological and technical advances of agriculture are the basis of population growth [2]. The population growth rate has slowed down recently, but given the anticipated figures, new systemic solutions must be found to ensure sufficient water, food, and energy supply, while all solutions must be based on environmental protection and preserving biodiversity [3].

The utilized agricultural area of Hungary has decreased due to urbanization, land use due to the development of various elements of the infrastructure, and the transformed economy and ownership structure. As a result, during the last half a century, Hungary has lost almost 1.6 million hectares of agricultural land. Compared to the total area of Hungary, this represents 17.2%, and compared to all areas used for agriculture in Hungary this amounts to 28%. This decrease can be observed in the Northern Plains and Szabolcs-Szatmár-Bereg County as well. Despite the decreasing agricultural land, more and more food is needed for the growing population. In the crop structure configuration, farmers need to consider both external and internal factors as well as they need to determine their current situation and their goals to be achieved. The rapidly changing economic and natural environment, technological knowledge transfer and demographic change call into question many permanent economic and social paradigms [4].

According to the SWOT framework, the factors can be grouped into four categories to provide a basis for achieving specific goals. The analysis uses the method of strategic evaluation. We considered it necessary to consult the farmers participating in the survey on this issue. Taking account of the four criteria, through their own businesses, their advantages and disadvantages over their competitors are revealed. Within the external environment, opportunities are positive, and threats have a negative impact on the business [5]. SWOT analysis helps analyse the situation of businesses. We can identify Strengths, Weaknesses, Opportunities, and Threats. The development of the method can be linked to Stanford University. When classifying the criteria, we asked the farmers themselves to decide, to the best of their ability, which of the external and internal features of their agricultural enterprise were to be classified as weaknesses. The following attributes had the greatest proportion: land prices and land rents, changes in the selling price of cereals, available processing capacity, available irrigation capacity, changes in production costs, producer sales, animal nutrition and animal husbandry, and producer association and cooperation. These are all negative signs of the county. These factors are judged either to be negative or to have an inadequate degree or even none.

The environmental impact of agriculture is the most significant human activity [6]. The characteristic feature of Hungarian agriculture is that it is the most significant natural resource available based on its abilities and capabilities. It is determined by the totality of such conditions and system approach [7]. Agriculture, as a product-producing and value-creating activity, plays a special but increasingly diminishing role in the structure of the Hungarian economy and thus in its overall performance. Agriculture and regional development can be examined in a system approach [8, 9]. Factors considered as traditional strengths need to be continually examined, most notably climate, as environmental changes have a significant impact on food production volumes and safe production and supply.

Climate change affects rainfall and the amount of irrigation water available, the species and quantity of plant and animal pests and has a major influence on the yield of the most important agricultural measurement factor [10]. Economy and production plans are rendered nearly impossible by temperature fluctuations and the increasing occurrence of unplanned extreme events [11, 12]. This is confirmed by the observation that annual rainfall has decreased from 640 millimetres to 560 millimetres, and its distribution over time is unpredictable [13, 14]. The type of irrigation is primarily determined by the soil, the temporal and spatial distribution of the hydrological water shortage, the cultivation technology, the asset and workforce potential of business organizations, and the quality and quantity of surface water or groundwater available [15, 16]. They form the same body of water [17]. Water management has become a key domestic and global strategic issue, now more important than ever in human history [18].

Mapping, evaluating, and correctly interpreting competitive advantages at the level of individual and national economies is one of the most important tasks of agricultural policy. This is the basis of commercial success [19]. The structure of the formulation and implementation of the agricultural strategy must change. Instead of traditional thinking, faster and more flexible forms should be preferred. Mostly strategies based on improvisation, action plans, and visions are viable. They can keep up with the rapidly changing market situation [20]. There is an important fundamental interest in competitiveness that favours the introduction of strategies that search for the direction and application of sustainable development in the field of environment and nature [21, 22, 23].

Most of Szabolcs-Szatmár-Bereg County, which provided the framework for the study, is lagging behind [24, 25], is a peripheral area [26, 27, 28] missing an economic and social centre [29, 30]. Our research, however, is a niche, one that offers novel insights into a previously less researched topic. Our empirical results can also contribute to the preparation of a larger (nationally representative) research.

2. Materials and methods

In our paper, we examined the agriculture of Szabolcs-Szatmár-Bereg County through the opinions and experiences of farmers. Research has been carried out among maize and winter wheat cultivation organizations as these are the two most important terrestrial cereals. The basic population of the survey is the agricultural entrepreneurial world of the county, data on which was obtained through sixty agricultural advisers of the National Chamber of Agriculture in Szabolcs-Szatmár-Bereg County. They were contacted electronically with the help of the NAK County Board, closed-ended questionnaire using Google Forms (CAPI – Computer-Aided Personal Interview). The questionnaire was sent electronically to the agricultural advisers, and responses were also sent back electronically. Correlation coefficient analysis was performed to detect relationships. Farmers were able to answer the questions with a multiple-choice numerical value on a 5-point Likert scale (a measurement scale between two extremes) with 1 – the least and 5 – very textual explanations. It was a good solution to choose five grades for the well-known and well-managed value range because everyone could relate the value of individual grades to the value of school grades. In addition, the SWOT analysis method was used in the questionnaire for the values of the answers to the questions. The focus of the questions was to get to know the farms that produce arable crops.

The agricultural advisers helped to fill out the questionnaire for each farmer. Each farmer who was producing arable crops was randomly selected from the clients of the agricultural adviser (400-450 clients per adviser) in a way that a representative subjective knowledge may be discovered. By this method, we can also discover the objective data linked to crop production. We asked for consensual answers beyond those based on non-quantifiable data. It takes the greatest effort to reach a common decision: time, attention, flexibility; but in this case the data may reflect the views of a larger community. In reaching an agreement, it may be difficult to involve a sufficiently wide range of participants and stakeholders in the decision. This is why the knowledge of agricultural advisers is important. Thus, the data are based on a convincingly large number of databases. The knowledge of agricultural advisers is extensive and unmatched (8,000 clients per year). Due to the occurrence of randomness, the research is considered representative.

The research was conducted between 12 December 2018 and 12 September 2019. Sampler: 60 agricultural advisers. Sample size: 8,000 persons.

Sampling: the questionnaire was sent to all agricultural advisers with a return rate of 98.3%, i.e. 59/60. We used the following methods to increase the level of response we receive: motivational letter of invitation through the board, telephone, and personal questions and answers. The results can be generalized to the whole county because this sampling group is responsible for liaison, information transfer, and any assistance related to their farming activities. Filling in the questionnaires can be affected by the following factors: Internet access (it was provided by agricultural advisers), interest, and commitment to the topic. These factors are taken into account when interpreting our results. During data processing and compression, I used simple and weighted arithmetic mean calculations as well as distribution ratio calculations. The closeness of the relationship between the quality criteria was examined by cross-table analysis and calculated by Tschuprow's T measure of association.

3. Results and discussions

The figures in the study show the data and ratings of the participants as well as their views on a particular issue. From the data in the tables, which show the results of the whole research, in this publication we evaluate the answers based on the results of the SWOT analysis and reveal possible correlations. The tables display percentages.

Participants included almost four times as many men (47 persons, 80%) as women (12 persons, 20%). The county's agriculture as a whole is typically dominated by men.

By age distribution: 11 persons (19%) below 40 years of age and 48 persons (81%) above 40 years of age. The average age of participants is 49.06 years of age. Average deviation of farmers' age from the average – 24 and + 16 years.

The respondents were divided into four groups according to their education. Higher education: 10 persons (17%), secondary education: 39 persons (66%), primary education: 7 persons (12%), and no formal education: 3 persons (5%). In terms of formal agricultural education: 53 persons (90%) have some sort of formal education, and 6 persons (10 %) do not have formal agricultural education.

There are 13 districts in Szabolcs-Szatmár-Bereg County. We have received data from 9 districts. *Figure 1* shows the percentage of participants from the different districts. The largest number of participants is from the Nyíregyháza district with 13 persons. This district has the largest population within the county as well as the county seat. The second largest samples came from the Nagykálló district. This is located right next to Nyíregyháza (10 persons). The third and fourth largest providers of participants were Csenger (8 persons) and Fehérgyarmat (7 persons). These two districts are located at the east side of the county. Szabolcs-Szatmár-Bereg County is a special agricultural county: the weight of agriculture is more than twice the national average [2, 20].

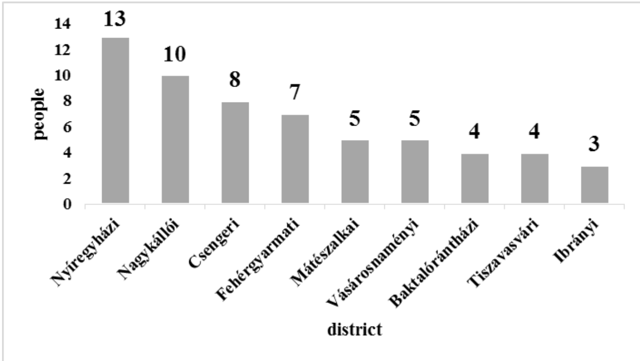

Figure 1. The geographical location of farmers, by districts

The size of arable land is given by the participants (*Figure 2*). The area of the farmers ranges from 1 to 470 ha. They farm an average of 40.16 ha, the standard deviation of the area being 72.7 ha. Self-owned arable lands range from 0 to 380 ha, of which on average 31.48 ha are privately owned, with a standard deviation of 57.6 ha. The area under other cultivation is between 0 and 200 ha, with an average of 9.07 ha and with a standard deviation of 28.3 ha. The largest area is the Tiszavasvári (700 ha) and the smallest is the Baktalórántházi district (42.12 ha).

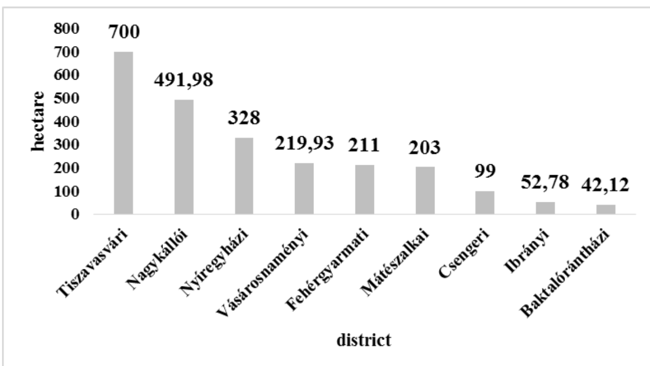

Figure 2. Size and district of farmers' arable lands

The arable area given by the survey participants is the typical Golden Crown (GC) in the particular districts. The average Golden Crown value of the participating farmers is 16.24 GC. The best value is in the Tiszavasvári area (*Figure 3*). There is a significant difference between the districts in this value between the cultivated fields. The average Golden Crown value of the agricultural areas of the region is 16.26 GC. Szabolcs-Szatmár is around 12.31 GC in Bereg County, 17.08 GC in Hajdú Bihar, and 19.41 GC in Jász-Nagykun-Szolnok County. The country average is 18,15 GC [31].

The typical Golden Crown value of arable land given by the survey participants ranges from 4 to 29 GC in each district. The average Golden Crown value for farms is 16.24 GC, its standard deviation being 5.2 GC. The typical Golden Crown value of arable land given by the survey participants ranges from 4 to 29 GC in each district. The average Golden Crown value for farms is 16.24 GC, its standard deviation being 5.2 GC. The Tiszavasvári area has the best average value, while the Baktalórántházi has the worst. The average Golden Crown value of the farms is above the county's average by 3.93 GC. It is almost at the regional level, just below by 0.02 GC. The average GC value of the farms participating in the survey is significantly worse than the average of Jász-Nagykun-Szolnok County by -3.17 GC. This is below the average value of Hajdú-Bihar County by -0.84 GC.

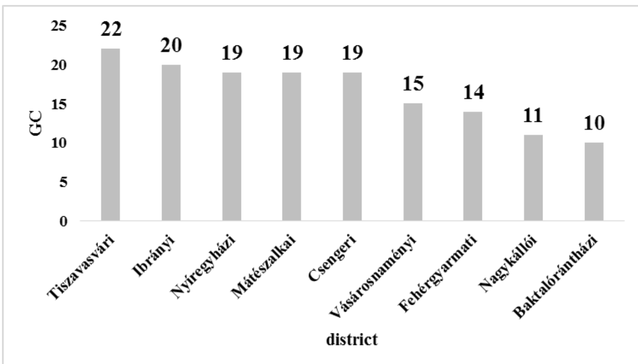

Figure 3. Average Golden Crown value per farmer per district

With the help of the SWOT analysis, we assessed the external and internal environment of the county's agriculture and identified the specific weaknesses in the presence of which farmers should produce. The SWOT analysis by county farmers is based on Strength, Weaknesses, Opportunities, and Threats acronyms (*Table 1*).

County farmers do not regard the weather and natural conditions of the county as weaknesses. However, it is a significant discovery that the factors of production attributed to weakness point out both abilities and skills. Based on this, it can be said that in a good natural environment (external environment) agriculture with low potential (internal environment) is conducted in the county. The close values of the opportunity, with the exception of land prices and production costs, show that farmers see some room for improvement in this respect. The results on production costs highlight that farmers pay less attention to cost administration and record keeping when they plan their budget.

Table 1. "Weaknesses" of county agriculture

Factors/Characteristics	**Strengths (%)**	**Weaknesses (%)**	**Opportunities (%)**	**Threats (%)**
Land prices and land rents	1.7	54.2	25.4	18.6
Changes in the selling price of cereals	5.1	54.2	23.7	16.9
Available processing capacity	6.8	47.5	42.4	3.4
Available irrigation capacity	5.1	50.8	40.7	3.4
Changes in production costs	5.1	35.6	23.7	35.6
Producer sales	6.8	44.1	40.7	8.5
Producer association and cooperation	3.4	47.5	47.5	1.7
Animal nutrition and animal husbandry	11.9	42.4	42.4	3.4

When formulating typical questions, we were guided by the principle of being able to form a broad view of the area in which they should perform their task. We provided an opportunity to evaluate both agricultural activities that can be interpreted closely, and by examining the complex economic, sociological, agricultural, legal, and environmental spheres of their enterprise, from a systemic point of view, we can get answers about their own interpretation and evaluation aspects. When evaluating Producer Association, we can see that they are not taking advantage of the cost-cutting and revenue-raising effects of joint purchasing and selling. This is reassured by listing Producer Sales as a weakness. Considering the fact that Producer Association is a weak and an opportunity factor in the county, it is understandable that Producer Sales is weak, and it is vulnerable to the processing industry. Examining this item along with the purchase price, it shows that they cannot negotiate a good price from the processing companies. Selling to end users directly is very rare. The vulnerability of county farmers is high due to the production of raw materials.

Factors broken down by districts and tabulated according to the four categories show that there is a difference in the opinion of farmers by geographical location. Most of the points in the assessment of weaknesses came from the Nagykálló district. According to the results, this district experiences the weaknesses of the county's agriculture the most (*Table 2*).

The role of common interest groups and co-operation is not nearly as significant as we would expect it to be, given its importance. Farmers in the Csenger district feel it is one of the biggest issues. They do not want or are not able to make use of the opportunity of joint action to sell at better prices and conditions. The task of common interest groups is to assist their members in purchasing and selling. The agriculture of the county is characterized almost every year by an old problem, which is related to the sale. The processing industry puts farmers in a difficult position. The processing capacity was also labelled as a weakness by the farmers, mainly by the farmers of Nagykálló district. Market production of goods is by no means an advocacy task, and it is not a political question. There is a need to negotiate jointly on sales, processing, and purchasing. An adequate financing is also easier to achieve if the sale is resolved.

Table 2. Result of the SWOT analysis, broken down by districts: weaknesses

Factors/Characteristics	Baktalórántházi; 4 people; 7%	Csengeri; 8 people; 14%	Fehérgyarmati; 7 people; 12%	Ibrányi; 3 people; 5%	Mátészalkai; 5 people; 8%	Nagykállói; 10 people; 17%	Nyíregyházi; 13 people; 22%	Tiszavasvári; 4 people; 8%	Vásárosnaményi; 5 people; 8%	Total amount
Producer sales	0	4	3	3	3	6	2	2	3	26
Available processing capacity	0	4	4	3	3	8	3	1	2	28
Land prices and rents	2	7	3	2	3	7	4	2	2	32
Change in production costs	0	5	2	1	1	3	4	3	2	21
Producer association and cooperation	0	6	5	3	3	3	3	2	3	28
Changes in the selling price of cereals	0	6	5	3	4	5	3	3	3	32
Available irrigation capacity	0	5	5	2	3	7	4	3	1	30
Degree of animal feeding and animal husbandry	0	1	4	3	2	4	7	2	2	25

According to the association results of the SWOT analysis, the most important is the geographical location in the feature-to-factor relationship. The second factor that influenced responses was the age of respondents. Respondents' gender was the third factor in this division, where we could establish a certain relationship. There was a significant relationship between respondents' age and the livestock status.

We were able to show a weak but clear influence on agricultural education. The highest education level has a weak correlation with the characteristics of the relatives' evaluation (*Table 3*) [32].

Table 3. SWOT analysis, the Value of Association Coefficients (T) (where "T" indicates the closeness of the relationship between the factor and the answers to the scientific questions)

Factors/Characteristics	Age	Gender	Education	Agricultural education	District
Land prices and land rents	0.076	0.233	0.142	0.119	0.191
Changes in the selling price of cereals	0.096	0.116	0.106	0.137	0.224
Available processing capacity	0.160	0.199	0.167	0.214	0.304
Available irrigation capacity	0.226	0.059	0.178	0.243	0.224
Changes in production costs	0.285	0.053	0.142	0.155	0.236
Producer sales	0.169	0.259	0.164	0.176	0.249
Producer association and cooperation	0.274	0.040	0.133	0.214	0.231
Animal nutrition and animal husbandry	0.407	0.056	0.121	0.209	0.242

4. Conclusions

It is characteristic of the results of our investigations that factors classified into the group of weaknesses are traditionally related to sales difficulties. Factors influencing production need to be examined continuously as environmental changes have a significant impact on the volume of food production as well as on safe production and supply. Evaluating and analysing the results of the micro-region and the opinions of the farmers provides the statistical values of the larger units. It has always been a difficult task to identify, assess, and take into account the strengths, weaknesses, opportunities, and threats of farming. However, data are needed to analyse the state of the sector: mapping its capabilities, reducing its handicaps, or making use of its competitive advantage. Another challenge that needs to be addressed is that economic calculations take due account of the present and future value of ecology [16, 33]. Planning has already taken into account the organizational strengths and weaknesses, has identified environmental opportunities and threats, and is more likely to achieve goals than in cases where the capabilities of their internal and external environments have not been assessed. In summary, one of the common problems is that the expected difficulties are not calculated during the strategy preparation period, and thus no solutions to mitigate difficulties are developed.

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The influence of slope aspect on soil moisture

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Abstract. In this research, we investigated the variability of soil moisture on two slopes of opposite aspect (a northern slope and a southern slope) but with the same soil type. To identify the spatial disposal of the soil type on both slopes, we georeferenced the paper-based soil map of Sfântu Gheorghe, using the QGIS platform. In order to use the correct slope aspect, we used a numerical model of the terrain (relief). The research plot was soil sampled at the depth of 10 cm on two different dates: on 7 November 2019 and on 3 March 2020, using a Buerkle soil sampler.

Gravimetric method was used to determine the soil moisture values that proved to be the most accurate for our purpose.

The soil moisture values, obtained in weight percentage, were assigned to the coordinates of the sampling points, and soil moisture maps were generated in QGIS for both slopes and for both sampling dates. These maps gave us the opportunity to evaluate the variability in time of the soil moisture distribution on the sample plots.

The water holding capacity of the soils is mostly influenced by their organic C content. So, the total organic carbon content of the soil from the sampling plots was measured using the Tyurin method.

Keywords: georeference, same soil types, grid soil sampling, organic carbon content

1. Introduction

The slope aspect is an abiotic cultivation factor, element of the relief, as one of the main pedogenetic factors. Slopes having different aspects have a wide range of microclimatic properties. The development of these different endowments is determined by the different values of the insolation. The quantity of the incident solar radiation influences the pedogenetic processes, the structure of the soils, and their texture. The regime of the temperature in the soil has an indisputable effect on the total edaphon, which contributes to the decomposition of the organic matter and to the formation of humus. All these parameters influence the seasonal water management of the soils and the changes in their moisture content [1]. Practising an efficient crop production in today's climatic and economic environment is only possible by the thorough knowledge of the endowments of the production plot [2].

Increasingly advanced GIS and satellite positioning tools are expanding the possibilities for investigating the influence of the slope aspect on different properties of the soils.

By georeferencing the paper-based soils map of Sfântu-Gheorghe, we had the opportunity to do our research on the same soil type (faeoziom), while the grid-point sampling method allowed us to compare more efficiently the samples from multiple sampling occasions and to better understand the results.

2. Materials and methods

Georeferencing the soil map

In order to find out whether there is the same soil type on the northern and southern slopes in the surroundings of Sfântu-Gheorghe, it was necessary to georeference the paper-based soil map, issued by the Office of Pedological and Agrochemical Studies (O.S.P.A.) in 1999, as the map does not show the topography and elevation values.

The soil map was geoferenced using the image-to-image method (*Fig. 1*), based on a georeferenced orthophoto. For the operation, we have selected points with known coordinates in the target coordinate system, and these could also been identified on the raster [3].

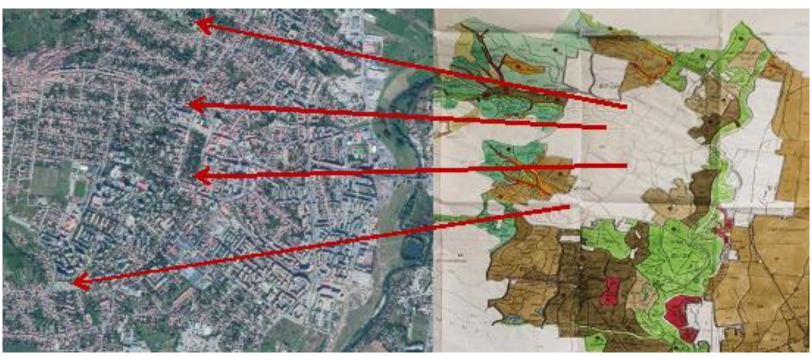


Figure 1. Georeferencing image to image

After georeferencing, the soil map became comparable with a topographic map. We overlaid the two maps and were able to identify a northern slope and a southern slope having the same soil type.

Verification of slope aspect by generating a slope aspect map

To prove that the two slopes we have selected truly dispose of the northern and the southern aspect, we generated a slope aspect map in QGIS [4]. To generate this map, we used a digital elevation model in GeoTIFF format (*Fig. 2*) with a resolution of 25 m. This means that a pixel contains the average elevation data of a 25 x 25 m plot [5].

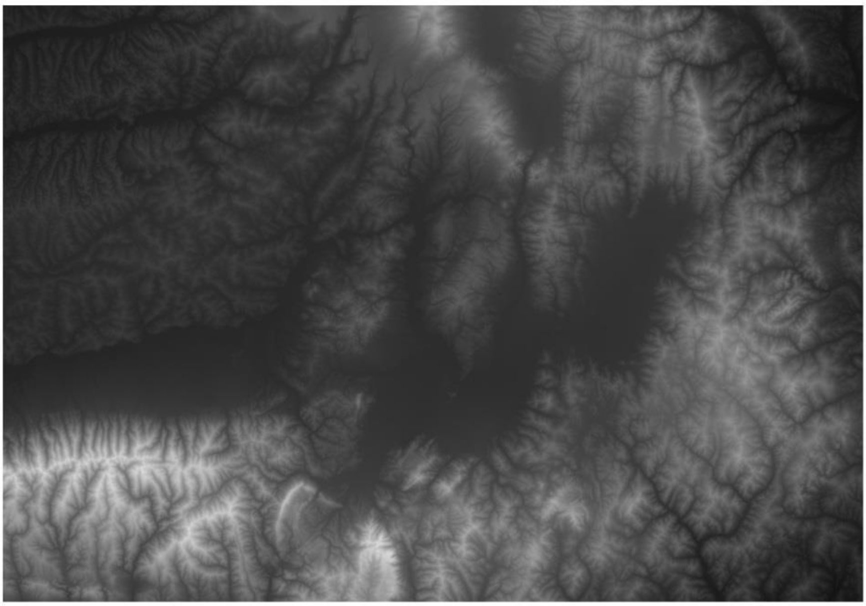
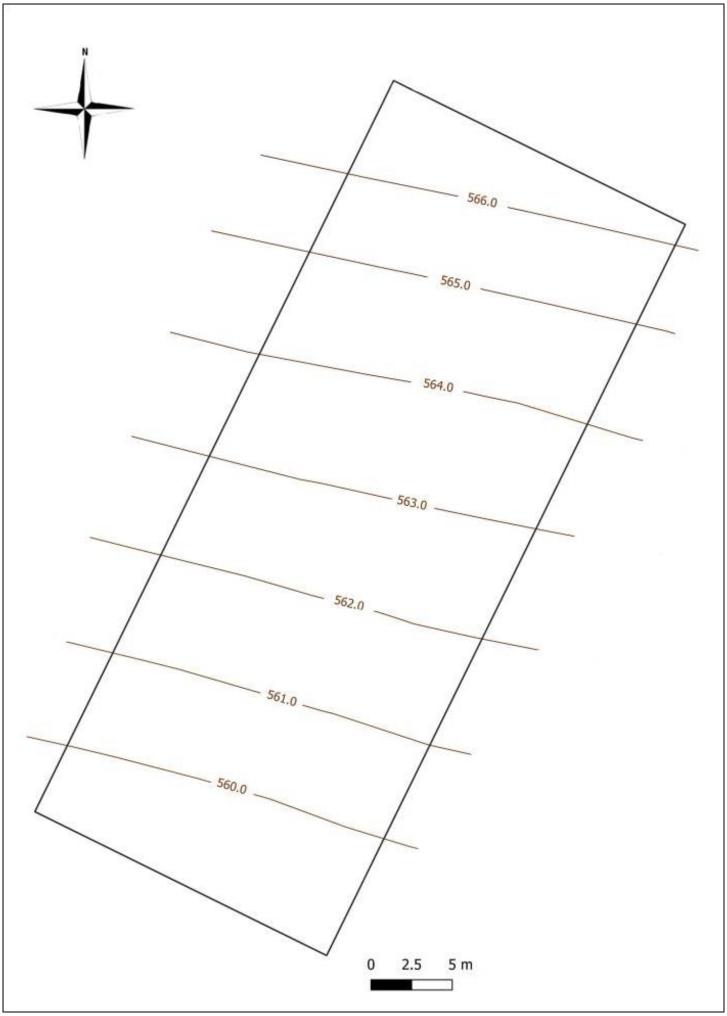
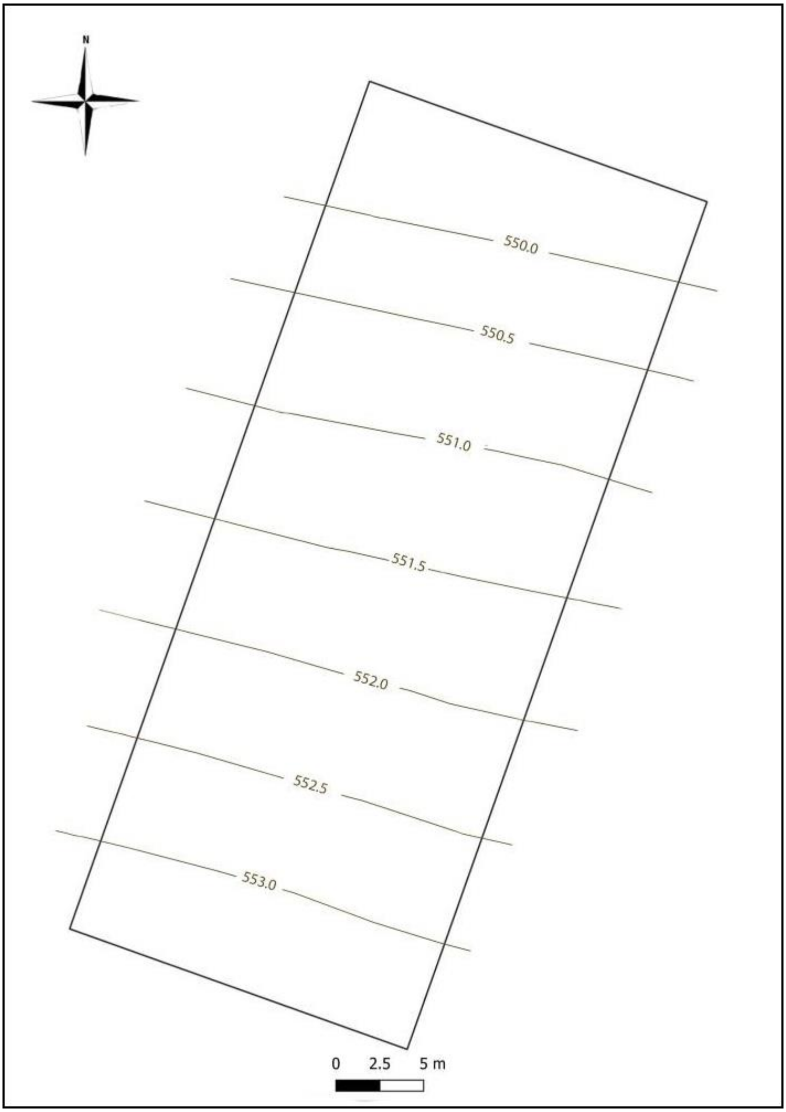


Figure 2. Digital elevation model

Before selecting the sampling plots on the slopes, topographic measurements were made as part of a preliminary field survey. These measurements were made with a Horizon Kronos C3 RTK (Real-Time Kinematic). With these measurements, we recorded the X and Y coordinates of the points in Stereo 70 projection and their elevation. The corrections were provided by the ROMPOS system [6]. Data were processed in QGIS, and an elevation curve map could be generated for the two plots/part of the two slopes (*Map 1* and *Map 2*).



Map 1. The elevation curve map of the southern slope



Map 2. The elevation curve map of the northern slope

Editing the sampling grid in QGIS

Two sampling plots were established, one on the northern slope and one on the southern slope. Essentially, this means two sampling grids of 1,000 sq. m edited in QGIS (*Fig. 2*), having the longer side downwards the slope.

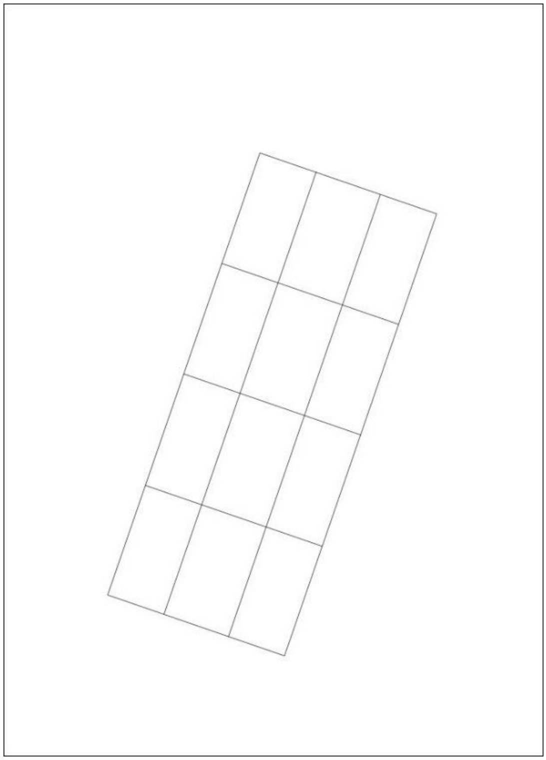


Figure 3. Sampling grid

Sampling

The soil samples were collected on two dates: on 7 November 1999 and on 3 March 2020.

The soil samples were collected at the grid points; so, we needed to set these points as topographical ones on the sampling plot. The operation was made with the same Kronos Horizon RTK (Real-Time Kinematic), controlled by Carlson SurvCE 6.0 software. The sampling was made with a Buerkle soil sampler from the upper 10 cm soil layer. From the same point, we took samples both for soil moisture and total organic C content determinations.

Generating the map of the soil moisture distribution of the sampling plots

The soil moisture of the samples was determined with the gravimetric method [7]. These values were ordered in a table with the coordinates and elevations of the sampling grid points. We obtained a table having 4 values for each point, and it was introduced in QGIS in CSV format [8] (*Table 1*).

Table 1. The coordinates of the southern slope sampling points with moisture

Sampling points	Northing	Easting	Soil moisture %
S1	484616.237	559573.968	28.4
S2	484619.398	559567.41	25
S3	484608.14	559561.987	27.3
S4	484604.973	559568.551	22.2
S5	484593.613	559563.088	24.7
S6	484596.78	559556.516	22

Table 2. The coordinates of the northern slope sampling points with moisture

Sampling points	Northing	Easting	Soil moisture %
N1	487477.848	560973.336	21.5
N2	487480.223	560966.439	20.5
N3	487492.146	560970.541	22.3
N4	487489.77	560977.431	21.6
N5	487501.591	560981.491	19.8
N6	487503.962	560974.607	22.5

Determination of the total organic C content of the soil using the Tyurin method

Total organic C is one of the matters that can absorb a great amount of water. So, we considered that its content can influence the quantity of water restrained by the soil [9].

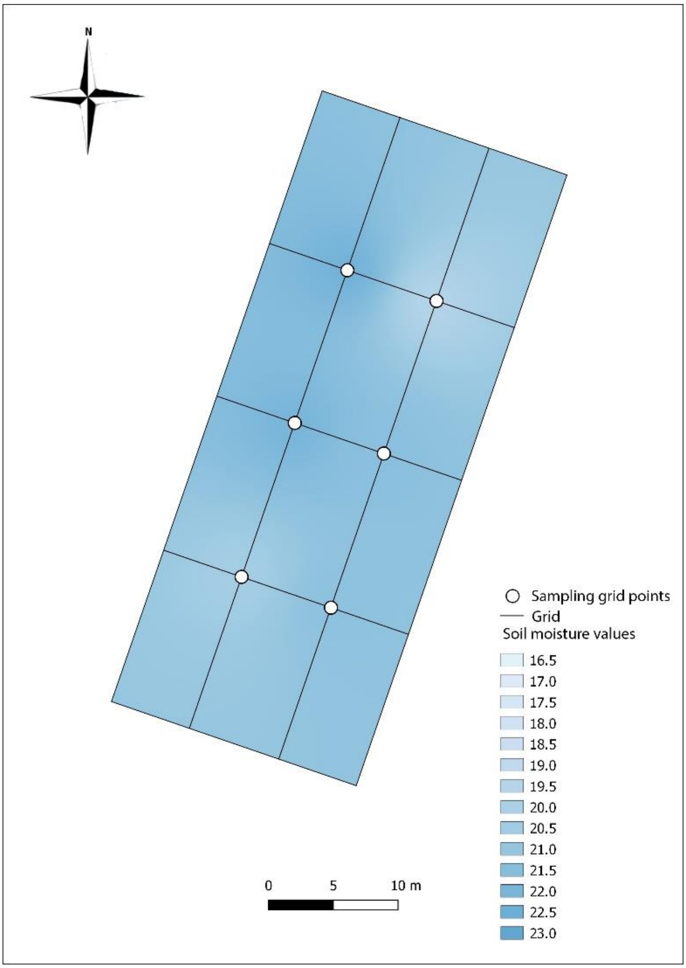
The soil samples were prepared by sieving through a 2 mm sieve, and any visible organic residues were removed. We measured 0.1–0.5 g of soil according to their colour and put them in tubes. We added 10 ml of dichromate solution acting as an oxidizing agent, covered the tubes with glass funnels, heated them in a water bath to the boiling point, and boiled the samples slowly for 5 minutes. After that, the samples were titrated with 0.2 normal Mohr salt; essentially, we re-titrated the dichromate which was not used during the oxidation process [9], [10].

3. Results and discussions

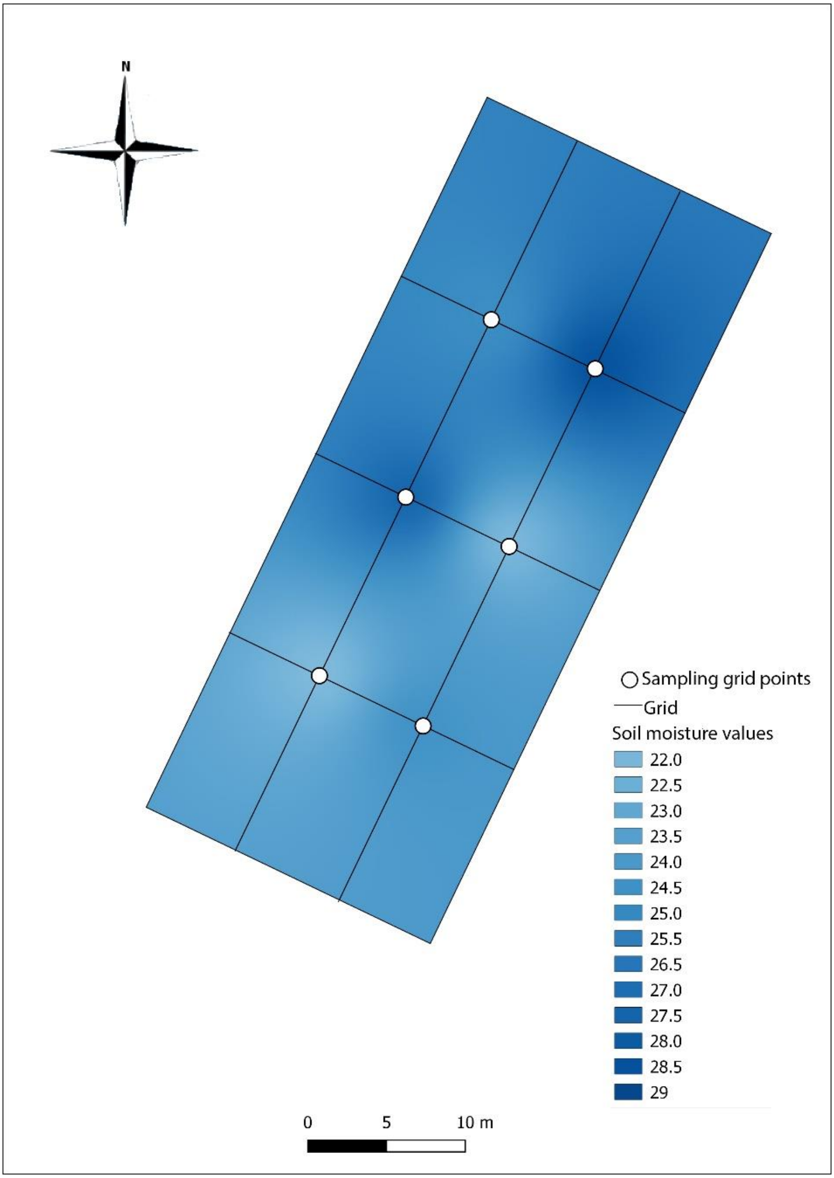
From the moisture values determined on the two sampling dates, soil moisture maps were interpolated within the sampling plots. The maps offer us the opportunity to compare the data and establish correlations.

Autumn sampling was preceded by a dry period of low rainfall. Our soil moisture determinations reached values between 16 and 21% on the southern slope. On the northern one, they were similar, i.e. between 16 and 21%.

Spring sampling soil moisture values were lower on the northern slope (compared to the southern slope values), ranging between 19.8 and 22.5% (*Map 3*), and so higher than in autumn with an average of 1%.



Map 3. The northern slope soil moisture distribution map in March



Map 4. The southern slope soil moisture distribution map in March

On the southern slope, these values were higher, between 22 and 28.4% (*Map 4*), with an average difference of 6% compared to the autumn determinations.

The values of the total organic carbon are evaluated through the graphs below, where the standard deviation can also be seen. On the northern slope, the values were between 2.7 and 4.2%, and just two samples reached 4% (*Fig. 5*). On the southern slope, all values were over 4% (*Fig. 6*).

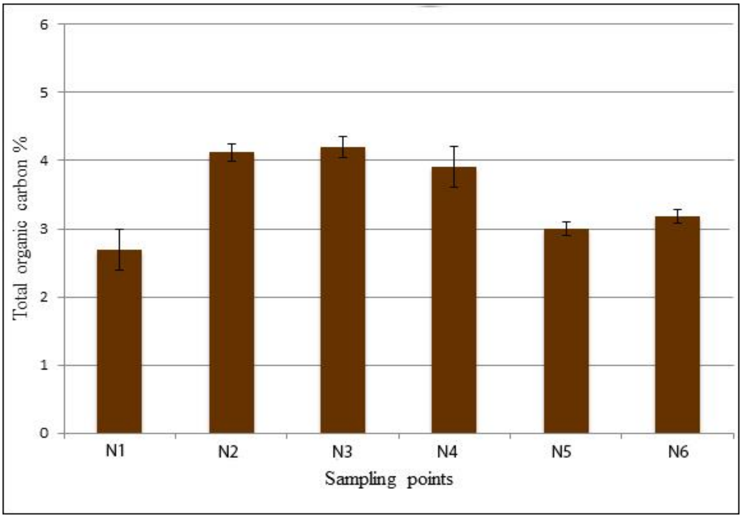


Figure 5. The northern aspect slope – total organic C content in percentage

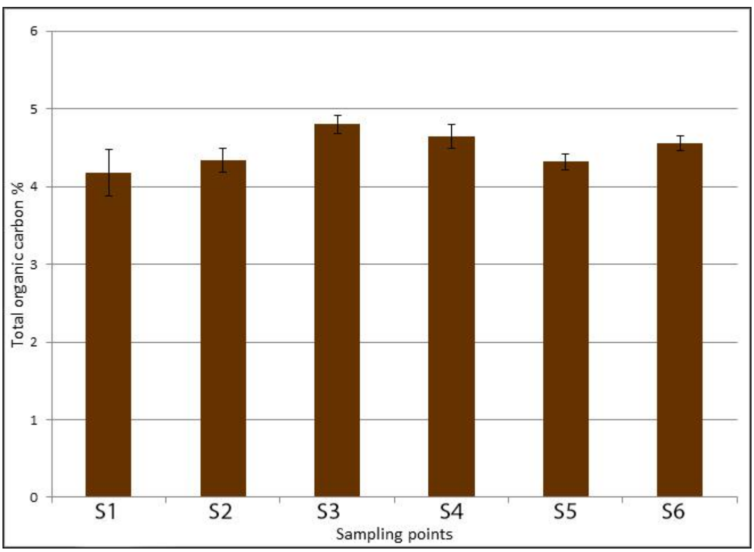


Figure 6. The southern aspect slope – total organic C content in percentage

4. Conclusions

These new methods like GIS and precision soil sampling provide new opportunities for a more complex study of the effect of slope aspect on soil moisture.

By georeferencing the soil map of Sfântu-Gheorghe in the Romanian projection system (Stereo 70), we opened up the opportunity for the information provided by this map to be compared – overlaid in GIS – to information provided by other maps, topographic information, and orthophotos. Through this map, we could do soil moisture measurements on the same soil type on different aspect slopes (a northern and a southern one). In addition, on both selected sample plots, we had the opportunity to make "virtual" grids and to set the grid points on the field, which became the soil sampling points.

The method of grid-point sampling allowed us to do the sampling always from same spot on each sampling date. As a continuation of the research, besides soil moisture, we can determine other soil properties connected to moisture as the samples can be collected from the same grid points already set.

The soil moisture values obtained on the northern and southern slope were alike on the autumn sample collection date. At the spring sample collection, the measured values were different. On the southern slope, the values were higher than on the northern one.

As the total organic carbon content of the soil plays a significant role in the variation of the soil water retention capacity, we determined this parameter too for each grid point for both sample plots. According to our results, the southern aspect slope sample plot has reached higher values. Therfore, we can suppose that the higher soil moisture values on the southern slope sample plot are due to the higher total organic carbon content [5]. However, more accurate conclusions can be done by simply carrying on with the measurements, evaluating the precipitation values, and determining the dominant wind direction (as a factor in the value of evapotranspiration). By making soil moisture measurements on a weekly basis, it will be possible to get an image about the drying out of the soil.

On the other hand, it is necessary to determine the total soil clay content as, besides the total organic carbon content, the clay colloid is able to absorb a great amount of water.

By georeferencing the soil map and by grid point sampling, we introduced new research procedures that will facilitate comparability between the studied slopes.

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