

**Hungarian University of
Agriculture and Life Sciences**

DOCTORAL PhD DISSERTATION

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Gödöllő

2025



Hungarian University of Agriculture and Life Sciences

**THE IMPACTS OF COVER CROP INTERCROPPING
TOWARDS MAIZE GROWTH DEVELOPMENT AND YIELD
QUALITY**

DOI: 10.54598/006540

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TABLE OF CONTENTS

LIST OF ABBREVIATION.....	1
LIST OF FIGURES	3
LIST OF TABLES	6
1. INTRODUCTION.....	7
1.1 Background of the study	7
1.2 Justification and objective of the study	8
1.3 Objectives of the study	9
2. LITERATURE REVIEW.....	10
2.1 Origin and classification	10
2.2 Growing stages.....	11
2.3 Nutritional value and usage	13
2.4 Economic importance of maize	16
2.5 Abiotic challenge in growing maize	17
2.6 Nutrient management and improved tolerance on abiotic stresses	20
2.7 Benefits of cover crops	21
2.8 Intercropping of cover crops in maize cultivation.....	25
3. METHODOLOGY.....	30
3.1 EXPERIMENTAL RESEARCH 1: Germination test of cover crop under different temperature and salinity levels.....	31
3.1.1 Experimental Materials	31
3.1.2 Growing Conditions	31
3.1.3 Statistical Analysis	31
3.2 EXPERIMENTAL RESEARCH 2: Germination test of 16 maize varieties under different temperature and salinity levels.....	32
3.2.1 Experimental Materials	32
3.2.2 Growing Conditions	32
3.2.3 Statistical Analysis	33
3.3 EXPERIMENTAL RESEARCH 3: The Effect of Nitrogen Fertilization on Maize Growth and Yield in Drought Condition	34
3.3.1 Experimental Site	34
3.3.2 Treatment and Experimental Design	35
3.3.3 Measurement	35
3.3.4 Statistical Analysis	36

3.4	EXPERIMENTAL RESEARCH 4: The impact of intercropping in alleviating drought and salinity stress in maize cultivation (Pot trial).....	36
3.4.1	Experimental Site and Materials	36
3.4.2	Maize and Cover Crops Variety	36
3.4.3	Treatments and Experimental Design	37
3.4.4	Measurements.....	38
3.4.5	Statistical Analysis	38
3.5	EXPERIMENTAL RESEARCH 5 & 6: Integration of various cover crops species on maize cultivation under different nitrogen levels	39
3.5.1	Experimental sites	39
3.5.2	Experimental designs	39
3.5.3	Measurements.....	40
3.5.4	Statistical Analysis	40
4.	RESULTS AND DISCUSSION.....	41
4.1	EXPERIMENTAL RESEARCH 1: Germination test of cover crop under different temperature and salinity levels.....	41
4.1.1	Germination parameters at different temperatures and salinity levels	41
4.1.2	Radicle and plumule length in different salt concentrations	42
4.1.3	Discussion	43
4.2	EXPERIMENTAL RESEARCH 2: Germination test of 16 maize varieties under different temperature and salinity levels.....	44
4.2.1	Germination Duration and Seedling Growth	44
4.2.2	Interaction between Salinity and Temperature on Maize Germination.....	46
4.2.3	Comparison of Germination Performance between Varieties in Abiotic Stress Conditions	48
4.2.4	Discussion	51
4.3	EXPERIMENTAL RESEARCH 3: The Effect of Nitrogen Fertilization on Maize Growth and Yield in Drought Condition	53
4.3.1	Vegetative stage.....	53
4.3.2	Yield	55
4.3.3	Grain chemical composition.....	58
4.3.4	Discussion	59
4.4	EXPERIMENTAL RESEARCH 4: The effect of different cover crops on maize growth development under drought and salinity stress (Pot trial)	61
4.4.1	Soil Moisture and Temperature	62
4.4.2	Maize vegetative stage	63
4.4.3	Maize reproductive stage	65

4.4.4	Cover crop dry mass.....	67
4.4.5	Discussion	68
4.5	EXPERIMENTAL RESEARCH 5: Integration of different leguminous cover crops on maize cultivation under various nitrogen levels in 2023	71
4.5.1	Maize vegetative stage	71
4.5.2	Maize harvested yield.....	74
4.5.3	Grain chemical composition.....	77
4.5.4	Cover crop dry mass.....	80
4.5.5	Discussion	80
4.6	EXPERIMENTAL RESEARCH 6: Integration of leguminous and Brassicaceae cover crops on maize cultivation under various nitrogen level in 2024.....	83
4.6.1	Maize vegetative stage	83
4.6.2	Maize harvested yield.....	86
4.6.3	Yield chemical composition	89
4.6.4	Cover crop dry mass.....	92
4.6.5	Discussion	92
5.	CONCLUSION AND RECOMMENDATION	97
5.1	Conclusion.....	97
5.2	Recommendation.....	99
6.	NEW SCIENTIFIC RESULTS.....	100
7.	SUMMARY.....	101
	APPENDICES	104
	A1. Bibliography.....	104
	A2. Research Activities /Photos.....	132
	A3. List of Statistical Analysis Tables.....	135
	LIST OF PUBLICATIONS OF THE AUTHOR IN THE RESEARCH FIELD.....	148
	OTHER SCIENTIFIC PUBLICATIONS OF THE AUTHOR	148
	ACKNOWLEDGEMENT	150

LIST OF ABBREVIATION

%	: Percent
ABA	: Absciscic Acid
ANOVA	: Analysis of Variance
ASI	: Anthesis-Silking Index
ATP	: Adenosine triphosphate
CAT	: Catalase
CC	: Cover Crop
CO ₂	: Carbon dioxide
DAS	: Days After Sowing
DC	: Double Cross
df	: Degree of Freedom
DNA	: Deoxyribonucleic acid
DS	: Drought Stress
EC	: Electrical Conductivity
EU	: European Union
FAO	: The Food and Agriculture Organization
GP	: Germination Percentage
H ₂ O	: Water
IAA	: Auxin
kg/ha	: Kilogram per hectare
LAI	: Leaves Area Index
LER	: Land Equivalent Ratio
LSD	: Least Significant Difference
MANOVA	: Multivariate analysis of variance
MA	: Maize intercropped with alfalfa
MM	: Monocrop Maize
MMB	: Maize intercropped with mung bean
MRC	: Maize intercropped with red clover
MWM	: Maize intercropped with white mustard
N	: Nitrogen
NaCl	: Sodium chloride
NH ₄ -N	: Ammonium
NPK	: Nitrogen-Phosphorus-Potassium
NUE	: Nitrogen Use Efficiency
P	: Phosphorus
<i>p</i> -value	: The Probability under the assumption of no effect or no Difference
PGPR	: Plant Growth-Promoting Rhizobacteria
PL	: Plumule length
PNUE	: Photosynthetic Nitrogen Use Efficiency
POD	: Peroxidase
RCBD	: Randomized Complete Block Design
RL	: Radicle Length
ROS	: Reactive Oxygen Species
SC	: Single Cross

SOC	: Soil Organic Carbon
SOD	: Superoxide Dismutase
SPAD	: Chlorophyll meter
SS	: Salinity Stress
SVI	: Seed Vigor Index
TC	: Three-Way Cross Hybrids
t/ha	: Tonne per hectare
TKW	: Thousand Kernel Weight
USA	: United States of America
USD	: United States Dollar

LIST OF FIGURES

Figure 1. Phenotypic difference between maize and teosinte with the genes responsible for each characteristic.....	10
Figure 2. Internal structure of maize seed and the seed germination process	11
Figure 3. Maize vegetative and reproductive stages characteristics.....	12
Figure 4. Precipitation and temperature data at the Gödöllő experimental field from May to September 2022-2024 (2024, MATE – Gödöllő).....	35
Figure 5. The planting distance between maize and the cover crops (2023 & 2024, MATE Gödöllő)	40
Figure 6. Germination rate of different cover crops at two different temperature and salinity levels (2021, MATE-Gödöllő).....	41
Figure 7. Average radicle length of each cover crop developed after 10 days incubated at two different temperature and salinity levels (2021, MATE-Gödöllő).	42
Figure 8. Average plumule length of each cover crop developed after 10 days incubated at two different temperature and salinity levels (2021, MATE-Gödöllő).	43
Figure 9. Overall germination percentage of all maize varieties at different days of incubation under different temperatures and salt stress conditions (2022, MATE-Gödöllő).....	45
Figure 10. Growth of maize radicle at different days of incubation under different temperatures and salt stress conditions (2022, MATE-Gödöllő).	46
Figure 11. Growth of maize plumule at different days of incubation under different temperatures and salt stress conditions (2022, MATE-Gödöllő).	46
Figure 12. Overall germination performance of 16 maize varieties at two different salt concentration across three different temperatures (2022, MATE-Gödöllő).....	46
Figure 13. Radicle growth average of 16 maize varieties at two different salt concentration across three different temperatures (2022, MATE-Gödöllő).....	47
Figure 14. Plumule growth average of 16 maize varieties at two different salt concentration across three different temperatures (2022, MATE-Gödöllő).....	47
Figure 15. Maize seedlings root:shoot ratio comparison of at two different salinity levels treatments across three different temperatures (2022, MATE-Gödöllő).	48
Figure 16. Comparison of maize seed vigor index (SVI) at two different salinity levels treatments across three different temperatures (2022, MATE-Gödöllő).	48
Figure 17. Plant height comparison of maize cultivated at five nitrogen level between 2022 and 2023 (2023, MATE-Gödöllő).	54
Figure 18. Leaves area comparison of maize cultivated at five nitrogen level between 2022 and 2023 (2023, MATE-Gödöllő).	54
Figure 19. Comparison of the SPAD value of maize cultivated at five nitrogen levels between 2022 and 2023 (2023, MATE-Gödöllő).	54
Figure 20. Ear length comparison of maize cultivated in five different N levels between the year 2022 and 2023 (2023, MATE-Gödöllő).	55
Figure 21. Ear weight comparison of maize cultivated in five different N levels between the year 2022 and 2023 (2023, MATE-Gödöllő).	56
Figure 22. Comparison of G:C ratio produced by maize subjected to five different N levels in the year 2022 and 2023 (2023, MATE-Gödöllő).	56
Figure 23. Comparison of grain weight produced per maize ear subjected to five different N levels in the year 2022 and 2023 (2023, MATE-Gödöllő).	57

Figure 24. Maize thousand kernel weight (TKW) comparison between five nitrogen levels and two different years (2023, MATE-Gödöllő).....	57
Figure 25. Starch content of maize subjected to five different nitrogen levels between the year 2022 and 2023 (2023, MATE-Gödöllő)	58
Figure 26. Protein content of maize subjected to five different nitrogen levels between the year 2022 and 2023 (2023, MATE-Gödöllő)	59
Figure 27. Oil content of maize subjected to five different nitrogen levels between the year 2022 and 2023 (2023, MATE-Gödöllő).	59
Figure 28. Average soil moisture content in pots containing maize intercropped with various cover crops and exposed to different abiotic stress treatments (2024, MATE-Gödöllő).	62
Figure 29. Average soil temperature in pots containing maize intercropped with various cover crops and exposed to different abiotic stress treatments (2024, MATE-Gödöllő).	63
Figure 30. Average SPAD values of maize intercropped with different cover crops in different abiotic stress treatments (2024, MATE-Gödöllő).	64
Figure 31. The average height at R1 of maize intercropped with different cover crops in different abiotic stress treatments (2024, MATE-Gödöllő).	64
Figure 32. The average leaf area at R1 of maize intercropped with different cover crops in different abiotic stress treatments (2024, MATE-Gödöllő).	65
Figure 33. The anthesis: silking index (ASI) of maize intercropped with different cover crops in different abiotic stress treatments (2024, MATE-Gödöllő).	66
Figure 34. The average ear weight of maize intercropped with different cover crops in pots and subjected to different abiotic stress treatments (2024, MATE-Gödöllő).	67
Figure 35. The average ear length of maize intercropped with different cover crops in pots and subjected to different abiotic stress treatments (2024, MATE-Gödöllő).	67
Figure 36. The dry mass of above ground structure of alfalfa and bean at different stress treatments (2024, MATE-Gödöllő).	68
Figure 37. Average plant height at VT of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).	72
Figure 38. Average leaves area at VT of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).	73
Figure 39. Average leaves area index (LAI) at VT of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).	73
Figure 40. SPAD values at VT of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).	74
Figure 41. The average ear weight of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).	75
Figure 42. The average ear length of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).	75
Figure 43. The average G:C ratio of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).	76
Figure 44. The average kernel weight per ear of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).	76
Figure 45. Thousand kernel weight (TKW) average of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).	77
Figure 46. Oil content average in seeds of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).	78
Figure 47. Protein content average in seeds of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).	79

Figure 48. Starch content average in seeds of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).	79
Figure 49. Plant height average at VT of maize plant intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).	84
Figure 50. SPAD value average at VT of maize plant intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).	85
Figure 51. Leave area average at VT of maize plant intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).	85
Figure 52. Leave Area Index (LAI) average at VT of maize plant intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).	86
Figure 53. Ear weight average of maize intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).	87
Figure 54. Ear length average of maize intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).	87
Figure 55. G:C ratio average of maize intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).	88
Figure 56. Average kernel weight per ear of maize intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).	88
Figure 57. Thousand kernel weight (TKW) average of maize intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).	89
Figure 58. Oil content (%) average in maize seeds intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).	90
Figure 59. Protein content (%) average in maize seeds intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).	91
Figure 60. Starch content (%) average in maize seeds intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).	92

LIST OF TABLES

Table 1. Taxonomic classification of cultivated maize	11
Table 2. Average composition per 100 g of edible portion of maize	14
Table 3. Industrial usage of maize	15
Table 4. Hungary maize cultivation and production 2014-2023	17
Table 5. Maize varieties tested for the germination performances (2022, MATE - Gödöllő)	32
Table 6. Mean data of germination percentage of different maize varieties in incubated in 100 mM NaCl solution at different temperatures (2022, MATE-Gödöllő).....	49
Table 7. Mean data of radicle length of different maize varieties incubated in 100 mM NaCl solution at different temperatures (2022, MATE-Gödöllő).....	50
Table 8. Mean data of plumule length of different maize varieties incubated in 100 mM NaCl solution at different temperatures (2022, MATE-Gödöllő).....	50
Table 9. 2022 and 2023 maize grain weight converted from kg/ ear to tonnes/ha at five nitrogen level (2023, MATE-Gödöllő).	57
Table 10. Mean value \pm standard deviation of the days after sowing ((DAS) of maize tassel and silks emergence in different CC and irrigation treatments (2024, MATE-Gödöllő).....	65
Table 11. Mean \pm standard deviation of maize moisture content (%) in different nitrogen and cover crop treatments (2023, MATE-Gödöllő).	78
Table 12. Mean \pm standard deviation of cover crop dry mass at different nitrogen levels harvested within 0.2 m ² area between the maize rows (2023, MATE-Gödöllő).....	80
Table 13. Mean \pm standard deviation of moisture content (%) of maize seeds intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).	90
Table 14. Mean \pm standard deviation of cover crop dry mass harvested within 0.2 m ² area between the maize rows (2024, MATE-Gödöllő).	92

1. INTRODUCTION

1.1 Background of the study

Maize (*Zea mays* L.), or corn, is one of the world's most versatile and multifunctional crops. Originating from Central America, maize has evolved into a staple food, an essential industrial raw material, one of the sources of bioethanol, and a vital component in animal feed across the globe. Its adaptability and wide range of uses make it indispensable in various sectors (TORRES et al. 2015). As the human population increases and is predicted to reach 10 billion by 2050, it is agreeable that maize is one of the most important crops in ensuring world food security and future energy alternatives (LANGNER et al. 2019). The modern agriculture revolution, with the discovery of synthetic fertilisers, pesticides, herbicides, and modern mechanisation, allows farmers to improve land productivity allowing higher yield production per area compared to traditional farming methods (TOKATLIDIS 2013).

However, the current monoculture practice in maize cultivation around the world may not be a sustainable production method in a long term. Long-term monoculture maize can lead to soil nutrient imbalance, increase susceptibility to diseases such as *Fusarium* ear rot, and reduce aboveground biodiversity in the area (FUCHS et al. 2021; ASRADE et al. 2024). Furthermore, the monoculture maize is more susceptible to the impact of climate change, which increases the frequency of abiotic stress on the crop, proving to be challenging for farmers and producers to manage. In Europe, the prolonged drought and high temperatures in 2022 impacted both winter and summer crops, including maize, which was predicted to be 7.8% lower than the past 5 years' average (TORETI et al. 2022).

The future of food supply is threatened not only by abiotic stresses but also by the depletion of arable land to produce sufficient food to feed the 10 billion human population. In 2020, only 38% of the earth's surface was allocated for agriculture, with only one-third of this area used for crop cultivation, and the number continues to decrease steadily as more land is needed for human residential and commercial areas (FAO 2020). The threat is not only in the reduction of the total area of arable land but also in fertile soil quality, which is needed to produce maximum yield potential. Soil degradation can be accelerated by aggressive farming practices, especially by excessive tillage, which causes soil compaction and heavy use of chemical input, which eliminates the underground biodiversity. Soil acidification, salinisation, and chemical contamination are also factors that need to be considered in preventing soil degradation (BIRKÁS and DEKEMATI 2023). It is proven that uncontrolled soil degradation will eventually lead to land desertification

(KERTÉSZ and KŘEČEK 2019; NASCIMENTO 2023). The Corn Belt in the USA is one of the examples of significant soil degradation from high-intensity farming. Over 100 million acres of topsoil eroded from the area due to over-tillage practice, and fertiliser application was not enough to increase the yield produced to the same level of healthy soil (THALER et al. 2021). Furthermore, it was predicted that the Corn Belt will be unsuitable for maize cultivation by the year 2100 due to temperature increases in the region (BURCHFIELD 2022).

1.2 Justification and objective of the study

Intercropping may be one of the solutions to produce more food in a limited area. It has been estimated that about 20-30% of land use efficiency could be increased through this ancient cropping system, allowing sustainable agriculture intensification (MACLAREN et al. 2023). This method also produced 38% more gross energy in terms of yield while using 23% less land than in monoculture practice (MARTIN-GUAY et al. 2018).

However, possible yield lost mainly on the cash crop is one of the main concerns for farmers. While it is easier to practice intercropping in small-scale farming, it was proved to be trickier in larger commercial production as it will be more challenging to manage and potentially increase the production cost if not adequately planned (BLESSING et al. 2022). The additional costs, including cover crop seeds, extra pest control, increase in labour costs, and different mechanization may also put a barrier for farmers to change their production system (HUSS et al. 2022). Furthermore, factors such as crop combinations and mechanisms should be further investigated in order to maximise the yield while ensuring soil sustainability in the intercropping system (KHANAL et al. 2021).

Evidently, several studies have proven the ability of cover crops to be interseeded with the main crops, including wheat and maize, without negatively affecting the yield of cash crops (GAUDIN et al. 2013; LAW et al. 2022). BARIBUTSA et al. (2008) showed that interseeding red clover and chickling vetch did not decrease the maize yield at any maize planting density. Meanwhile, another study discovered that alfalfa intercropping with maize reduced the grain yield and maize biomass from 14% to 18.8% and 15.9% to 25.8%, respectively, compared to monocultured maize. However, alfalfa's forage yield could compensate for the loss in maize grain and biomass yield (BERTI et al. 2021). The latest results by ZHANG et al. (2022) revealed that maize cultivated with alfalfa without nitrogen treatment produced significantly similar maize yield amounts with maize monocropping with nitrogen treatments. The same study also found; application of N fertiliser also significantly increased maize yield in intercropped with alfalfa compared to monocrop maize.

Moreover, a legume-based intercropping system is supposed to stabilise crop yield under stressful conditions such as drought and nutrient deficit due to the effective interaction between the roots with plant growth-promoting rhizobacteria (PGPR) in the rhizosphere (CHAMKHI et al. 2022). It was revealed that maize and pigeon pea intercropping enhance soil micro and macro aggregates, increases organic phosphorus (P) storage (55 mg P/kg higher than monoculture), and increases 13% more biologically fixed N in soil compared to monoculture pigeon pea (GARLAND et al. 2017). TANG et al. (2021) also found that the intercropping system required 21% less P than the monocultured system to produce the same amount of yield. In a 7-year study, intercropped maize-legume showed 4% higher soil organic carbon content and 11% higher soil organic N content in the 20 cm top soil layer compared to monoculture practice while producing 23% higher root biomass (CONG et al. 2015). VORA et al. (2021) mentioned that cross-colonization of PGPR could occur between legume and cereal plants in intercropping practice due to the composition of different root exudates in intercropped cultivation compared to monocropped cultivation. It has been shown that a cross-colonization of bacteria from the roots of leguminous pigeon peas can occur within 28 days after sowing on the roots of the companion maize plant, even without artificial inoculation (VORA et al. 2021).

1.3 Objectives of the study

The objectives of this study are as follows:

- i. To evaluate the influence of intercropping different cover crop species on maize vegetative development, yield production, and yield quality.
- ii. To determine the effect of nitrogen levels in monocropped and intercropped maize cultivation
- iii. To investigate the potential use of cover crops in alleviating salinity and drought stress in maize growth development

2. LITERATURE REVIEW

2.1 Origin and classification

Maize or corn (*Zea mays* L.) have been consumed by humans for millennia, either directly or indirectly, as animal feed. Historically, maize was first cultivated around 8700 years ago in the lowland areas of Mexico. As the maize breeding knowledge and technology advance, the area where this crop is cultivated widens across the globe from the southern latitude of 40°S up to the northern latitude of 50°N and up to 3500 m above sea level (TENAILLON and CHARCOSSET 2011). The theory of maize originated from teosinte (*Zea mays* ssp. *Parviglumis* and ssp. *Mexicana*) was the most accepted among researchers due to the similarity in DNA structure with the same chromosome numbers and gene arrangements (BUCKLER and STEVENS 2006; SHARMA et al. 2021). Teosinte is a wild grass with distinctive phenotypic characteristics than maize. It was believed that teosinte experienced five major mutations to be transformed into maize (BEADLE 1939). The distinctive features between these two species are summarised in Figure 1.

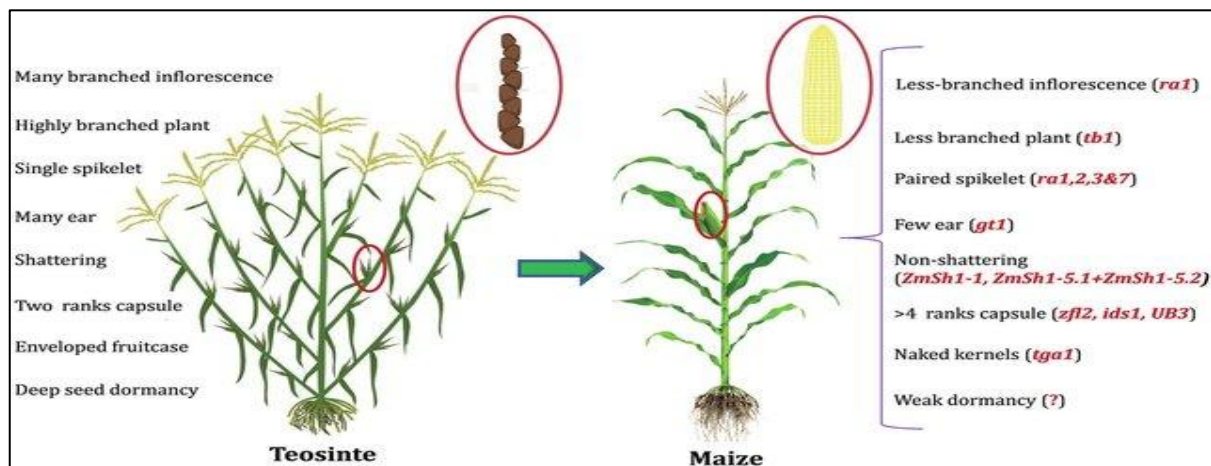


Figure 1. Phenotypic difference between maize and teosinte with the genes responsible for each characteristic
Source: (SHARMA et al. 2021)

Maize is classified as a type of annual grass and a C_4 species within the genus of *Zea* in the Poaceae family, with chromosome numbers of $2n=20$. This grass family also includes various important crops, including wheat, rice, barley, sorghum, and sugarcane. There are five species within the genus *Zea*, including the domesticated *Zea mays* L., *Zea diploperennis* (perennial wild grass), *Zea luxurians* (annual wild grass), *Zea nicaraguensis* (annual wild grass), and *Zea perennis* (perennial wild grass). *Zea mays* is further classified into four subspecies: *Zea mays* spp. *huehuetenangensis*, *Zea mays* spp. *mexicana*, *Zea mays* spp. *parviglumis*, and *Zea mays* with the first three are considered as the ancestor of the modern maize (DOEBLEY and ILTIS 1980). Table 1 simplifies the taxonomic classification of cultivated maize.

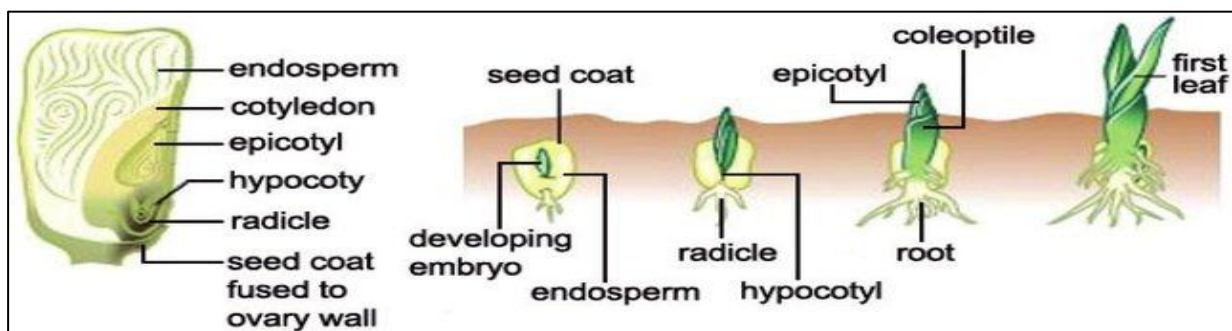
Table 1. Taxonomic classification of cultivated maize

	Scientific classification	Description
Kingdom	Plantae	Plants
Subkingdom	Tracheabionta	Vascular plant
Superdivision	Spermatophyta	Flowering plant
Division	Magnoliophyta	Monocotyledons
Class	Liliopsida	
Subclass	Commelinidae	
Order	Cyperales	
Family	Poaceae	Grass family
Genus	<i>Zea</i> L.	Maize
Species	<i>Zea mays</i> L.	Maize
Subspecies	<i>mays</i>	Maize

Source: (AWATA et al. 2019)

2.2 Growing stages

The life of the maize crop consists of three important stages which consist of germination, vegetative and reproductive stage. All three stages are crucial for producers in order to maximise the end production. Maize seeds, like other plant seeds, require optimal conditions, including temperature, moisture, and oxygen, to germinate and emerge at optimum levels. Water is crucial in seed germination, which is needed for seed imbibition. The presence of water allows the activation of protease and amylase enzymatic activities in the endosperm with the regulation of several hormones such as abscisic acid (ABA) and auxin (IAA) (MIRANSARI and SMITH 2014). The α -amylase and β -amylase enzymes are responsible for hydrolysing the starch and regeneration of energy (ATP) for the plant's early development (XUE et al. 2021). Germination ends when the radicle has grown out of the seed's coating layers and with the protrusion of the coleoptile (Figure 2). A recent study revealed that the temperate maize variety requires 20°C as the ideal germination temperature and showed the most rapid seedling growth after 11 days compared to the higher and lower temperatures. The study also shows that temperate maize varieties require 3 - 7 days to germinate within 5 - 35°C, while longer days are required at lower temperatures (KHAEIM et al. 2022). However, tropical maize varieties required a higher optimal germination temperature and higher tolerance to temperatures up to 35°C compared to the temperate family (KHALID et al. 2021).

**Figure 2.** Internal structure of maize seed and the seed germination process

Source: (ZHAO et al. 2012)

Maize growth stages are divided into two physiological stages, which are vegetative (V) and reproductive (R) stage. Figure 3 illustrates these two stages, with each stage labelled numerically. The vegetative stage starts with the emergence stage (VE), followed by V1, V2, V3 and so on, with each number representing the number of leaves with visible collars. Immediately after emergence, the seedling will rely on food reserve from the residue of the caryopsis under the ground until the V2 stage. The development of roots and leaves allows independent water and nutrient absorption as well as energy generation through photosynthesis (AWATA et al. 2019). The plant water and nutrient demand, especially nitrogen (N), is the highest during V11 until V17 stages to accommodate the plant's rapid growth (BELFIELD et al. 2009). VÁRI and PEPÓ (2014) concluded that irrigation and fertilization influence 21.5% and 45.2% of yield production, respectively. A study in Hungary also discovered that nitrogen fertilisation significantly increased chlorophyll content and grain yield while weather conditions influenced the effectiveness of nitrogen application (SZÉLES et al. 2012). Meanwhile, stresses during V18 could cause a delay in silking until after the pollen sheds (ESPINOZA and ROSS 2010). The vegetative stage ends with VT, indicating the tasselling stage approximately seven weeks after sowing.

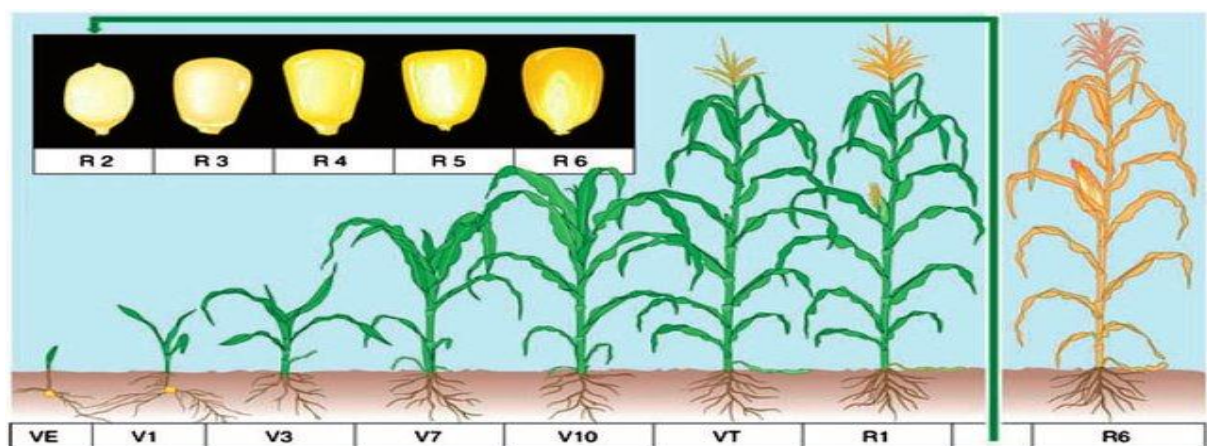


Figure 3. Maize vegetative and reproductive stages characteristics
Source: (NAFZIGER 2008)

The reproductive stages, denoted by the letters R1 (silking stage), R2 (blister stage), R3 (milk stage), R4 (dough stage), R5 (dent stage), and R6 (physiological maturity), begin during or right after tasselling. It is estimated that grain maize requires 105-120 days to achieve R6 after emergence (ESPINOZA and ROSS 2010). At R1, the tassel or the male reproductive part emerges at the top of the plant at the 14 to 15 leaves stage. Maize pollen is shed between 40 and 50 days after seedling emergence, and it is also highly influenced by the variety and environmental conditions. The emergence of the ear between the axil of the 11th to 13th leaf, exposing the silk of the maize, will allow pollination and fertilization to occur at this stage. The plant also requires a high amount of water and nutrients, especially N and P, for successful fertilization and preventing kernel abortion (SUBEDI and MA 2009; SANTOS et al. 2023). In Hungary, it was recommended

to apply N between 120 to 150 kg/ha depending on the maize variety to achieve the maximum yield (PEPÓ and KARANCSI 2014). It was also suggested that 60 kg/ha N as the optimum dose during dry years and 120 kg/h N in wet years (NAGY 2012). Pollination is only successful if the pollen grains are captured by the moist silk. Successful fertilization allows the kernel to enter the blister stage (R2) after two weeks of silk emergence, and starch starts to accumulate in the kernel (ESPINOZA and ROSS 2010). Fertilization significantly increased NPK uptake in maize hybrids, which is also influenced by genotype, fertilizer dosage, and crop year, leading to improved yields and nutrient content in both vegetative and generative parts (PEPÓ and KARANCSI 2017).

At kernel filling stages between R2 and R5, plant nutrient uptake is rapid, and higher energy is invested during these stages. Limited nutrient availability and environmental stresses will result in the reduction of kernel size and low grain yield (SÁRVÁRI and PEPÓ 2014; BISWAS and MA 2016). 20 days after silking, grain denting (R5) is observed, indicating completion of embryo development. At R6, kernels achieved physiological maturity two to three weeks after the dent stage. At this stage, the milk line disappeared with the kernel's moisture content reducing to 30%, and no increment in dry weight will be observed. A black layer of accumulated dead cells starts to form at the tip of each kernel 30 days after silking, which blocks further absorptions of assimilates and moisture into the kernel (VIEIRA et al. 1995). It is recommended to begin harvesting as the grain moisture reaches 20%, and grains should be dried to a moisture content of 13% or less for further use and storage to prevent mycotoxin and bacterial contamination (CHANNIAH and MAIER 2014).

2.3 Nutritional value and usage

In the ancient Greek language, *Zea* means “sustaining life”, while *mays* in the Taino language mean “life give” (SHAH et al. 2016). This description may be accurate as nowadays, as maize is one of the most important food sources, other than wheat and rice, and has higher bioenergy potential than the other two. Maize is not only consumed as a carbohydrate but also for its oil and protein content. Besides that, maize is also rich in vitamins and minerals, for example, vitamin C, vitamin E, vitamin K, vitamin B, folic acid, selenium, N-p-coumaryl tryptamine, and N-ferrulyl tryptamine (ERENSTEIN et al. 2022). Yellow maize grain is also rich in carotenoids, while purple or black maize is rich in anthocyanins, which are important antioxidant compounds (CERINO et al. 2019). Table 2 illustrates the average nutrient composition of 100 g maize.

Table 2. Average composition per 100 g of edible portion of maize

Source	Per 100g
Carbohydrate	71.88 g
Protein	8.84 g
Fat	4.57 g
Fiber	2.15 g
Ash	2.33 g
Moisture	10.23 g
Phosphorus	348 mg
Sodium	15.9 mg
Sulfur	114 mg
Riboflavin	0.10 mg
Amino acids	1.78 mg
Minerals	1.5 g
Calcium	10 mg
Iron	2.3 mg
Potassium	286 mg
Thiamine	0.42 mg
Vitamin C	0.12 mg
Magnesium	139 mg
Copper	0.14 mg

Source: (SHAH et al. 2016)

Shape of the kernel and the endosperm composition differentiate the name of and function of a maize variety. Starch contributes to 72 – 73% of maize kernel, and the distinguish physiochemical properties will differentiate the maize type (SANDHU et al. 2004). Grain maize types include flint, dent, floury and waxy kernels. They are the major contributor to the world maize production. Approximately 99% of maize grown in the U.S. consists of dent corn. The starch in dent corn consists of 23% amylose and 73% amylopectin is the key ingredient for livestock feed, cornmeal, corn syrup, and ethanol production (RANJAN et al. 2024). Dent maize is known for its high yield potential and adaptability to diverse environmental condition making it a staple crop in many regions (SEYE et al. 2019). However, the dent stage of maize kernels particularly favourable to the accumulation of fumonisins which is a harmful mycotoxin produced by *Fusarium verticillioides*, posing a health risks to humans and animals (PICOT et al. 2011).

Besides that, other maize types also include popcorn, sweet corn, and baby corn. Due to their favourable taste, these types of maize are usually consumed directly fresh, as snack or added in daily cooking. Sweet maize is classified as a tropical crop and has historically not been widely cultivated in temperate climates as it requires higher growing temperatures and greater soil moisture content compared to dent maize (FEKONJA et al. 2011). However, in the recent years, the cultivation of sweetcorn is also common in the temperate region thanks to the climate change, introduction of new hybrids and advanced in irrigation technology.

Maize is a versatile crop valued for its diverse components, particularly its grain, which serves as a vital source of food, feed, and industrial products. The starch component in maize

grains is the most utilised as a human food source as well as animal feedstock. Grain corn is usually converted into flour and used in various cultures as a staple food (RANUM et al. 2014). The high energy and nutrient content in maize grain makes it an excellent feed grain for animals such as poultry, cattle, and swine. Meanwhile, silage which is produced using the whole above ground plant structure of silage maize harvested at R5 is one of the important feeds in cattle industry and excellent source for provitamin A for ruminants (LOY and LUNDY 2019). The protein content in maize grain is also one of the most important protein sources for livestock after soybeans. Furthermore, maize produces the highest oil content compared to the other two important cereals (wheat and rice). Maize oil is extracted from maize germ (embryo), which contains 45 – 50% oil. The oil contains 14% saturated fatty acids, 30% monounsaturated fatty acids, and 56% polyunsaturated fatty acids, which is commonly used as cooking oil (SHAH et al. 2016).

In addition, due to its versatility maize has been in used in various industrial sector. Maize protein or zein has unique film-forming properties, which have a high potential to be used in various industries, including the food, medical, and pharmaceutical sectors (GITERU et al. 2021). Meanwhile the gelatinization properties of different type of starch molecules can be manipulated in generating products such as bioplastics and face talcum powder (OSTRANDER 2015). Besides that, maize grain contains high sugar levels in the form of glucose, sucrose and oligosaccharides, making corn one of the sweetener sources in the form of corn syrup as well as industrial and fuel ethanol production (HELSTAD 2019). Maize can also be transformed into paper, adhesive, organic solvent and cosmetics products. Table 3 summaries various industrial usage of maize.

Table 3. Industrial usage of maize

Industrial maize products			
Diaper	Acetic acid	Blankets and bedding	Antibiotics
Soap	Charcoal briquettes	Carpet tile	Aspirin
Bio-degradable utensils	Dyes and inks	Cosmetics	Citric acid
Paper, recycled paper	Insecticides	Electroplating	Enzymes
Cardboard	Building materials	Food packaging	Rayon fabric
Textiles	Matches	Mannitol	Medical syrups
Glues and adhesives	Metal plating	Industrial chemicals	Printer color carrier
Batteries	Organic solvents	Leather tanning	Surface coating
Bookbinding	Cleaners, detergents	Organic solvents	Crayons and Chalk
Fireworks	Plasticizing agents	Ethanol	Dyes

Source: (IOWA CORN 2025)

What distinguishes maize from other cereal crops is its greater ability to produce higher volume of biofuel in the form of bioethanol per hectare land compared to other main cereal crops. Bioethanol production from maize grains has gained significant attention as a renewable energy source, due to its high starch content for fermentation (YESMIN et al. 2020). There are various concerns in first generation maize biofuel production in term of utilising food crop as the biofuel

feedstock which may threatened the food security and increase in food prices (RANJAN et al. 2024).

However, various studies highlight the efficiency of different corn parts, such as cobs and stalks, in bioethanol production through chemical and biological processes. Research indicates that corn cob powder can be chemically degraded using hydrochloric acid, yielding up to 14% ethanol after fermentation with yeast (PANGGABEAN et al. 2024). Meanwhile, corn stalks can be pretreated with hydrolytic enzymes, achieving significant sugar extraction and resulting in a bioethanol yield of 0.77 g/g (BEHL et al. 2023). Bioethanol from corn contributes to energy security and rural economic development, particularly in the USA where it is already produced at an industrial scale (MOHANTY and SWAIN 2019). The use of agricultural waste, such as corn stover, for bioethanol production not only reduces fossil fuel dependency but also promotes sustainable waste management (FANSURI et al. 2024).

2.4 Economic importance of maize

The global production of maize has experienced a significant increase over the past several decades, facilitated by enhancements in maize yield and the expansion of cultivated areas. In 2022, 203 million hectares of land were cultivated with maize all around the globe, producing 1.16 billion tonnes of grains. In comparison, around 604 million tonnes of maize were harvested from 137 million hectares of land 20 years ago. Meanwhile, 808 million tonnes of wheat were produced from 219 million hectares of land in 2022 (FAO 2022). By 2018, 56.3 % of world maize production was utilised as animal feed and only 28% as human food. The European region contributed 75.7% of the maize production in the animal feed industry, while the African region is the highest in consuming maize as human food, with 54.3% while 30% of the production consumed as animal feed (ERENSTEIN et al. 2022).

The origin of the European maize is thought to come from the Caribbean by Christopher Columbus in Spain in 1493, which initially spread across the warm southern European region. Later, European hybrids were developed after World War II to adapt to the colder northern European climatic conditions (TENAILLON and CHARCOSSET 2011). Since then, maize has become one of the most important cereal crops in Europe, other than wheat and barley. In Hungary, maize is one of the most important crops after wheat and occupies the second-largest portion of arable land in the country. In the past 10 years, the harvested area and the total maize production in Hungary have fluctuated significantly (Table 3).

Table 4. Hungary maize cultivation and production 2014-2023

Year	Harvested area (hectares)	Total harvested production (tonnes)	Average yield (kg/hectare)
2014	1,191,420	9,315,104	7,820
2015	1,146,127	6,632,783	5,790
2016	1,011,563	8,729,915	8,630
2017	988,823	6,739,186	6,820
2018	939,080	7,976,941	8,490
2019	1,027,592	8,277,813	8,060
2020	981,006	8,414,350	8,580
2021	1,054,566	6,462,205	6,130
2022	816,643	2,781,774	3,410
2023	770,694	6,278,912	8,150

Source: (KSH 2024a)

In 2022, almost 817,000 ha of land was used in Hungary to produce 2.8 million tonnes of maize in comparison to 979,000 ha of land used for wheat cultivation to produce 4.4 million tonnes yield (KSH 2024a). In the same year, 1.99 million tonnes of Hungarian maize was exported with a value of USD 874 million. The export volume was higher in 2021, with 3.27 million tonnes traded for USD 996 million due to the higher total production (6.46 million tonnes) (FAO 2024). A long-term study in Hungary showed that optimal soil cultivation, fertilization, and plant density could yield up to 8.59 t/ha maize grain, while, unfavourable conditions could drop maize grain yields down to 2.09 t/ha (BERZSENYI and DANG 2008). The estimate is still applicable as the country's maize productivity was 8.15 t/ha in 2023, while the drought conditions in 2022 generated 3.41 t/ha grain maize (KSH 2024a). In comparison, the United States of America (USA), the world's biggest maize producer, managed to produce around 10.5 t/ha to 11.0 t/ha maize in recent years (FAO 2024).

In conclusion, maize plays a crucial role in Hungary's agricultural economy, significantly impacting both domestic production and international trade. Its cultivation occupies a substantial portion of agricultural land, making it a key crop for farmers and the national economy.

2.5 Abiotic challenge in growing maize

Even with advanced technology in the form of improved germplasm, fertiliser, and irrigation systems nowadays, there are still many challenges for maize producers in producing high yields and good quality maize in abiotic stress conditions. Abiotic challenges in maize cultivation include high or low temperature, drought, nutrient deficiency and soil salinity. Even though maize is considered highly polymorphic among crops with high genetic variability, all maize varieties will need optimal growing conditions to achieve their yield potential. Under favourable conditions, maize seedlings usually take 4 to 5 days to emerge, but under unfavourable conditions, the seedlings may take up to 21 days to emerge (ESPINOZA and ROSS 2010). Research indicated that temperature significantly impacts germination and seedling growth. It was published that 20°C

is ideal for maize germination, and a range below 30°C is optimal for the growth of temperate maize variety (KHAEIM et al. 2022). In North China Plain, farmers adjust the sowing dates to control temperature variations around flowering stages, which enhances summer maize yield, with early or delayed sowing having improved the photosynthetic capacity and, ultimately, grain yield (GUO et al. 2022).

Drought could cause problems for maize at multiple growth stages, from germination to seed filling stage (LIU et al. 2022b). In order to produce one kg of maize product, it was estimated that 1222 L of water is required (MEKONNEN and GERBENS-LEENES 2020). Water deficit stress leads to decreased cell viability and altered phenolic acid contents in maize leaves, affecting drought responses (ATTA et al. 2022). Implementing water deficit treatments at different growth stages of waxy maize also results in reduced fresh ear yield and grain quality, particularly when deficits occur during the V6–VT and VT–R2 stages (HASSAN and HADI 2022). Drought at the flowering stage decreases ovary sink potential and limits photosynthates allocated for ovary development, thus reducing the seed set number (WESTGATE 2015). Water deficits during hot and dry periods can indeed also impact maize silking and flowering, affecting pollination and seed set (AL-NAGGAR et al. 2022). Drought stress at the flowering stage significantly reduces grain yield and quality traits in maize, with notable decreases in grain yield, ears per plant, and kernels per plant (KOLO et al. 2022). As the drought periods are more frequent in Europe, cultivating a longer-season maize variety may not be a wise choice anymore (WEBBER et al. 2018).

Meanwhile at high temperatures, maize yield is reduced due to disruption of carbohydrate metabolism and starch biosynthesis needed for both male and female reproductive organs. At high temperatures, maize plants export higher amount of photosynthates and assimilates to vegetative plant parts compared to ear development (SUWA et al. 2010). Meanwhile, maize plants exposed to a maximum/minimum temperature of 40/30°C produced advance tassel inflorescence, advance pollen shedding, and broadened the anthesis-silking interval. Even though the high temperature did not affect silking but affected plants will produce low kernel numbers due to low number of viable pollens showing that the male reproductive organ in maize is more sensitive to high-temperature stress than female organs (WANG et al. 2019).

Furthermore, drought conditions will also elevate the impact of heat stress on maize. BHEEMANAHALLI et al. (2022) presented that pollen germination was reduced by 19 – 42% due to drought, heat and both stressors combined. Combined drought and high heat stress at the flowering stage reduced 18.0 – 37.6% kernel weight (LIU et al. 2022b). Moreover, seeds produced at water deficit and high heat conditions have lower endosperm cells and are smaller in size (LIU

et al. 2022b). It was estimated that 1.7% of the maize final yield would be reduced each day spent above 30°C under drought conditions compared to only 1% under optimal rainfed conditions (LOBELL et al., 2011). Importantly, abiotic stresses at the reproductive stage, could contribute to pollen sterility, pollen tube deformation, flower drop, and ovule abortion and cause a reduction in the final yield (SINHA et al. 2021). Besides that, the combination of heat and drought stress affects the kernel quality in maize in terms of biochemical content such as starch, protein and oil. Heat stress suppresses multiple enzymes involved in starch biosynthesis, thus reducing the starch accumulation between 21.8 - 22.2% (YANG et al. 2018). However, protein content increases in heat stress conditions due to an increase in the production of glutamate synthase (YANG et al. 2018). Studies show glutamate suppressed leaf senescence in grass by preventing chlorophyll degradation and improving the heat tolerance in maize seedlings (LI et al. 2019b; ROSSI et al. 2021). Besides that, maize plants that experienced both stresses showed a reduction in kernel starch, protein and oil content (BARUTCULAR et al. 2016; YOUSAF et al. 2022).

Moving on, soil salinisation poses a significant threat to agricultural productivity, which is driven by both natural and anthropogenic factors, including climate conditions and agricultural practices. Saline soil is characterized by soil with electrical conductivity (EC) exceeding 4 dS mL⁻¹ (equivalent to approximately 40 mM NaCl) within the root zone at 25 °C, along with an exchangeable Na concentration of 15% (YUNUS et al. 2024). Salt-affected soils cover about 600 thousand hectares in Hungary (BIRKÁS and DEKEMATI 2023). Without proper agriculture practices, it was estimated that 50% of global croplands would be salinized by 2050 (MEASHO et al. 2022). It was discovered that maize plants are more susceptible to salt stress at germination and early vegetative growth stage (BLANCO et al. 2007; FAROOQ et al. 2015). Salinity stress during the growth and reproduction stage will affect both green fodder and kernel yield of sensitive maize hybrid by decreasing the grain weight and count (NIU et al. 2012; CUCCI et al. 2019). A study reported that salt stress significantly inhibits maize acid invertase activity up to 50% in soil experiments and can affect about 50% kernel reduction in maize ears. Acid invertase activity is responsible for sucrose hydrolysis in the phloem to be transferred into the maize kernel (HÜTSCH et al. 2014). In the salt-tolerant maize variety, it was observed that high phenolic compounds such as anthocyanins and polyphenols are produced by the plant, which exhibits high antioxidant activities which limit the oxidative damage by the Reactive Oxygen Species (ROS) (HICHEM et al. 2009).

In Hungary, it was predicted that a yield reduction of up to 30% would occur every 9 years with an increase in frequency due to climatic changes (HUZSVAI et al. 2024). It was estimated that around 20% of yield will be lost in Europe by 2050 if the growers continue with the current

agricultural practices, such as sowing dates, relying on the rainfed system, and cultivating the non-tolerant maize genotypes. Therefore, farmers need to invest in additional inputs such as irrigation systems to fight against inevitable and unpredictable stresses such as extreme temperature, limited water, and salinity to achieve their forecasted yield value (SHIFERAW et al. 2011). In Hungary, projections indicated that the volume of summer precipitation will exhibit a declining trend as we near the year 2100, whereas winter precipitation is anticipated to experience a considerable increase (BARTHOLY 2007). As maize is cultivated between spring and late summer, this forecast acts as a warning for farmers for the future production cycles. Therefore, new agricultural technology and cropping systems need to be adapted in order to produce high quantities of good quality food while maintaining the sustainability in the inputs usage, including soil, water, and nutrients.

2.6 Nutrient management and improved tolerance on abiotic stresses

Optimum nutrient management is important in helping plants to develop tolerance against stress. A study published that the optimal nitrogen application during drought years is 120 kg/ha, and farmers should avoid excessive top dressing as it does not compensate for low rainfall during drought periods (SZÉLES et al. 2023). Optimum application of macronutrients such as nitrogen, potassium, magnesium and calcium increase the concentration of antioxidant enzymes such as superoxide dismutase (SOD), peroxidase (POD) and catalase (CAT) but reduces the ROS. Moreover, micronutrients such as boron, iron, and zinc are also important in activating defence mechanisms against stress, including activating various physiological changes in plants and improving the metabolic activities in the plants to allow adaptation to various detrimental stresses (KUMARI et al. 2022). However, as the price of chemical inputs continuously increases and the environmental impact of the residues, the dependence on just chemical fertilizer against climate change may not be a sustainable approach.

It was discovered that the application of organic fertilizer and amendments, including vermicompost, vermiwash, biochar, and bio-fertilizer, could also improve salt and drought tolerance in plants. It was discovered that these organic amendments are not only rich in nutrients but able to improve antioxidant enzyme activities (SONG et al. 2022), reducing osmotic and oxidative stress (HOQUE et al. 2022), trapping excess sodium ions and decreasing electrical conductivity in saline soil (CHINSAMY et al. 2013). The usage of organic materials in assisting plants in salinity and drought stress conditions also includes microorganisms, including PGPR (ANSARI et al. 2019; ABDELAAL et al. 2021). A study reported that the incorporation of *Enterobacter cloacae* PM23 in saline conditions enhanced maize growth, biomass production, photosynthetic pigment contents, carotenoids, and relative water content in contrast to the control

treatment. Plants inoculated with this bacterium showed boosted radical scavenging capacity, elevated antioxidant enzymes, osmoprotectant, soluble sugars, proteins, total phenolic, and flavonoid content upon exposure to salinity stress condition (ALI et al. 2022). A study in Hungary revealed that maize plants inoculated with arbuscular mycorrhizal fungi produced higher fresh and dry biomass under control, salt, and high-temperature stress conditions compared to non-mycorrhizal plants. However, the ability of the fungi to produce defence enzymes and gene expressions under different stress conditions has varying effects depending on the type of stress and the age of the plants (MAYER et al. 2019).

Throughout time, researchers and breeders are continuously developing biotic and abiotic stress-tolerant varieties in crops. Traditionally, crops with desirable traits that occur naturally were chosen by farmers to be cultivated. The discovery of the Law of inheritance by Gregor Mendel unlocks the effectiveness of cross-pollination in producing desired characteristics in plants. Today, our understanding of the genes responsible for stress tolerance, along with our ability to modify them, enables genetic adjustments to produce plants with higher stress tolerance (GONZÁLES GUZMÁN et al. 2022). Based on a study by SZÉLES (2012), the genetic background of maize hybrids and environmental factors influenced their nitrogen uptake efficiency, which influenced the chlorophyll levels and yields. In Hungary, technological advancement, especially by improving soil cultivation methods, may mitigate the adverse effects of climate change on maize yields, with predictions indicating an average yield of 8.2 t/ha by 2050 despite heat stress (HUZSVAI et al. 2020).

2.7 Benefits of cover crops

A cover crop is defined as a plant cultivated to cover and improve the soil (BENEDICT et al. 2014). A cover crop is also defined as a fast-growing crop, such as rye, buckwheat, cowpea, or vetch, planted to prevent soil erosion, increase nutrients in the soil, and provide organic matter (BRITANNICA 2024). Cover crops can be used as soil mulching in the living or dead form or as green manure by incorporating them into the soil. Any type of plant can be used as a cover crop as long as it can provide full coverage and benefits to the soil. However, it is common to categorise them as leguminous, non-leguminous or grass cover crops (BENEDICT et al. 2014). Alfalfa (*Medicago sativa* L.), red clover (*Trifolium pratense*), hairy vetch (*Vicia villosa* Roth), annual ryegrass (*Lolium multiflorum* Lam.), and *Brassicaceae* species are a few examples of common cover crops used by farmers and producers in the temperate regions. Meanwhile, in the tropics, species such as *Pennisetum glaucum* (pearl millet), *Crotalaria juncea* (sunn hemp), and legumes

like *Mucuna pruriens* (velvet bean) and *Canavalia ensiformis* (jack bean) are more adaptable with the climate to be used as cover crops (PEDRINHO et al. 2018).

Cover crops play a crucial role in enhancing soil health, managing weeds, improving water retention, and increasing crop productivity. They offer a multi-functional approach by restoring soil ecosystem services, aiding in extreme weather adaptation, and potentially boosting farm profitability (BLANCO-CANQUI 2023a). Cover crops can influence soil water dynamics, organic carbon levels, and microbial activities, leading to improved crop yields and quality (QUINTARELLI et al. 2022). Cover crops such as grasses, legumes, and small grains species are beneficial for soil health, quality, and fertility by improving soil's physical, chemical, and biological properties. A study revealed that cereal rye pre-crop increased P availability and scavenging excess $\text{NH}_4\text{-N}$ from the soil after 3 years of study (HARUNA and NKONGOLO 2020). Besides that, the incorporation of cover crops back into the soil not only increases the organic matter in the soil but also the nitrogen content in the case of leguminous cover crops (FAGERIA et al. 2005). The use of cover crops also proved to reduce the loss of soil organic matter and soil organic carbon in a rainfed vineyard (LÓPEZ-VICENTE et al. 2020). The decomposition of cover crops belowground biomass also increases the soil carbon content in the soil up to 36% compared to non-cover crop soil (HARUNA et al. 2020).

Besides that, full soil coverage by the leaf structure of cover crops can provide soil protection against extreme weather and avoiding soil leaching by water and wind, especially in a vacant area (LÓPEZ-VICENTE et al. 2020; CHEN et al. 2022). It was discovered that cover crops with fine branched roots, such as ryegrass, are more effective against soil erosion than cover crops with thick roots, such as white mustard and fodder radish (DE BAETS et al. 2011). Besides that, the infiltration of the cover crop roots enhances water infiltration, soil aeration, and soil stability, thus preventing soil compaction and crusting formation (DELGADO et al. 2021). The benefits of cover crops in eliminating soil compaction also allow the following main crop to access subsurface water sources in the dry growing period (CHEN and WEIL 2011). In comparison to soil without cover crops, the presence of cover crops could reduce soil bulk density by 4%, increase 33% higher macropores, and 62.9% water infiltration (HARUNA et al. 2020). Furthermore, it was also found that the presence of cover crop helps in controlling pest populations such as nematode and weed pressure by smothering and allelopathic effects (ACHARYA et al. 2021). Cover crops can suppress weeds through competition and allelopathic effects, and their success in weed management varies based on species, planting time, density, biomass, and termination method (GILL et al. 2023).

Moreover, cover crops have been shown to impact soil microbial communities positively (GIUSTI et al. 2023). These crops can induce shifts in the soil microbiome that persist over time, potentially enhancing nutrient cycling and disease suppressiveness (RODRIGUEZ-RAMOS et al. 2022). Different cover crop species can lead to specific microbial footprints in the soil (CAZZANIGA et al. 2023). While the overall functional diversity of microbial communities may not differ significantly among cover crop treatments, differences in how the communities metabolize carbon sources have been observed. Conservation agricultural practices, including cover crops, can influence soil microbial communities, with variations in specific microbial taxa that could impact organic matter decomposition and plant growth (LIU et al. 2022a). Cover-cropped alley soils in pecan orchards have been found to contain higher relative abundances of microbes utilizing soil substrates, enhancing soil nutrient and moisture contents (RODRIGUEZ-RAMOS et al. 2022).

Therefore, the benefits provided by cover crops may be utilised and manipulated by farmers to improve soil conditions against abiotic stresses. It is mentioned that cover crops can be manipulated to alleviate the effect of climate change through soil carbon sequestration and emissions reduction from fertiliser production (JANSSENS et al. 2019). Cover crops play a crucial role in retaining soil moisture in drought-prone areas by influencing soil water dynamics and enhancing water-holding capacity. Research indicated that the timing of cover crop termination is a key factor affecting soil water retention, with early termination leading to greater soil water preservation at planting time and throughout the growing season (ZHANG et al. 2023). Additionally, the amount of cover crop biomass produced, tillage systems, and soil texture interact to impact soil water content, with cover crops being particularly beneficial in high precipitation regions (BLANCO-CANQUI 2023b). Meanwhile, pre-cropping grass species cover crops such as barley and rye improve the soil resistance to penetration during main crop sowing due to modified soil moisture content (BLANCO-CANQUI and JASA 2019). The larger volume of residue produced by the grass species crop contributes to improved water retention when compared to other species, such as legumes (GABRIEL et al. 2021).

Furthermore, studies have shown that integrating cover crops such as crimson clover, cereal rye, and turnip into cropping systems can significantly improve soil moisture dynamics, leading to higher soil moisture levels compared to systems without cover crops, especially during critical growth periods (MENDIS et al. 2022; LEBEAU et al. 2023). MENDIS et al. (2022) also suggest that the extended utilization of cover crops can enhance the soil moisture processes within maize cultivation systems by enhancing the levels of soil organic matter. Besides that, a study reported that long-term use of winter rye as a cover crop improves the soil water storage at 0 to 30

cm depth with an increment of field capacity water content between 10 - 11% and 21 - 22% of plant water available in the topsoil. The study also shows the presence of winter rye as the cover crop, which did not reduce the yield of the subsequent main cash crop, including maize and soybean (BASCHÉ et al. 2016). Consequently, this practice could serve as a feasible approach to promoting soil moisture preservation and enhancing soil quality, especially in drought-prone areas.

Furthermore, studies found that various plant species can be used as cover crops in high-salinity areas to mitigate salt stress in the ground. Plants such as *Hordeum vulgare* L. (barley), *Trifolium alexandrinum* (berseem clover), *Vigna marina* (beach pea) and *Brassicaceae* species are classified as moderate to high tolerance to salt stress (MITCHELL et al. 1999; YUNUS et al. 2024). Basically, these plants help desalinise the soil by extracting salt ions into their plant parts and lowering the harmful ions below the toxic level. The phytoremediation also reduces the soil's electrical conductivity and increases the soil water dynamic around the root zone. Some plants also acquired phytotransformation ability, which allows them to transform toxic compounds from salt ions into less toxic or non-toxic compounds. The presence of cover crops also improves the soil structure, temperature, and soil organic matter, which also influence the phytoremediation (MOHAMMAD et al. 2016). High organic matters in the soil also increase the beneficial microorganism that acts as osmolytes, which absorb the excess salt in the soil while providing other beneficial functions in promoting plant growth (ZAHIR et al. 2019; GAO et al. 2022). Besides that, the soil surface covered by the cover crops reduces water evaporation, which prevents capillary pulling of soluble salt ions to the plant root system (CAO et al. 2012). Another study also found that symbiotic interaction with specific rhizobial strains will allow leguminous cover crops such as *Vicia sativa* L. to increase the presence of nitrogen in soil with salinity issues (VENTORINO et al. 2012). Cover cropping also improves microbial diversity in saline soil, which includes bacterial and fungal populations (DASGUPTA et al. 2023).

The benefits of cover crop are also acknowledged by farmers in the European region especially in improving soil protection and health. In the effort to encourage sustainable agriculture within the European Union (EU) region, the practice of cultivating cover crops during non-growing seasons in autumn and winter is common in the region and one of the policies under the European Nitrate Directive (EUROPEAN COMMISSION 1991). The cultivation of catch and cover crops in the European Union regions has been acknowledged as a feasible approach for reducing greenhouse gas emissions and as protection in agricultural areas against erosion and nutrient leaching (JANSSENS et al. 2019). Currently, cover crops are commonly incorporated in one of the crop rotations but not integrated with the main crop growing period mainly due to additional cost and workload in their management (PEIGNÉ et al. 2016). Various factors have

influenced farmers to avoid incorporating cover crops into their rotations, such as the increase in production cost, extra labour requirements, lack of benefits, lack of awareness, and unsuitable climate (JANSSENS et al. 2019). It was discovered that a higher percentage of farmers in the northern European region implement cover crops in their rotation compared to farmers in the southern region due to the drier climate in southern Europe (VINCENT-CABOUD et al. 2017).

2.8 Intercropping of cover crops in maize cultivation

The environmental impact of growing maize varies depending on farming practices, but could contribute to soil degradation, excessive water use, and pesticide reliance. Conventional maize cultivation typically involves monocropping, which depletes soil nutrients over time and reduces biodiversity, making crops more susceptible to pests and diseases. This increased vulnerability leads to higher applications of chemical fertilizers, herbicides, and pesticides, which can contaminate nearby water sources and harm beneficial insects and wildlife (BELETE and YADETE 2023). Moreover, maize cultivation requires around 600 – 1000 mm water per growing period to grow optimally. Depending on the region climatic condition and water sources additional irrigation may be needed which contributing to groundwater depletion and water scarcity (BAGULA et al. 2022). Fertilizer runoff from maize fields is a major contributor to nutrient pollution, leading to issues such as algal blooms and hypoxic zones in lakes, rivers, and coastal areas, which disrupt aquatic ecosystems and also polluting human drinking water source (HALL 2024). However, implementing sustainable agricultural practices such as crop rotation, reduced tillage, cover cropping, and precision nutrient management can help mitigate some of these environmental impacts by improving soil health, reducing chemical inputs, and conserving water.

Intercropping is defined as an agricultural practice that involves the co-cultivation of two or more crop species within a particular land area, adhering to a methodical row configuration (LAYEK et al. 2018). Intercropping may take the form of strip, row, mixed or relay intercropping. Row intercropping is defined as growing two or more crops simultaneously where one or more crops are planted in rows. This specific form of intercropping promotes significant interspecific interactions, which include shading, root interaction, and competition for essential resources, including water, nutrients and oxygen. In addition, this system also allows beneficial symbiosis, such as disease and weed suppression (GLAZE-CORCORAN et al. 2020). In strip intercropping, multiple crops are cultivated concurrently in adjacent strips of sufficient width to allow for separate cultivation and harvesting yet close enough to enable agronomic interactions among the crops (LI et al. 2021). Intercropping may be one of the ways to maximize crop productivity in one land unit, reducing yield fluctuations while suppressing soil degradation and improving the quality.

Successful intercropping may improve crop performance with higher resistance to disease, pests, and extreme weather conditions, with a reduction in pesticide and fertiliser demand. In general, an intercropping system should have higher productivity than a monocropping system due to the efficient utilization of limited resources like water, light, and nutrients (LAYEK et al. 2018).

Intercropping or integration of maize with other crops has begun to gain popularity in several countries such as China, the United States of America and India. Alfalfa (BERTI et al. 2021), clover species (ZIYOMO et al. 2013), *Brassica*, oil radish (CHI et al. 2024), grass species such as rye (LIESCH et al. 2011) and various pulse legumes (TANG et al. 2021) are several examples of crops with high potential to be integrated with maize cultivation. Maize and soybean intercropping are currently being practised by farmers in several countries, and the yield produced was significantly higher than in monocultured land (MONZON et al. 2014; LIU et al. 2017). In China, maize cultivations are integrated with various crops, including soybean (WEI, LIU, et al. 2022), faba bean (ZHANG, CHEN, et al. 2012), pea (HU et al. 2016), and peanut (LI et al. 2019a). In several areas in Eastern and Southern Africa, smallholder farms proved to gain up to a 35% increase in maize production through the incorporation of pigeon pea (*Cajanus cajan*) in maize cultivation (CHAMKHI et al. 2022). In Indonesia (HARSONO et al. 2020), Bangladesh (ALI et al. 2017), Mozambique (TSUJIMOTO et al. 2015) and Tanzania (RENWICK et al. 2020), maize intercropped with soybean or pigeonpea in drought-prone areas generated higher yield than the monoculture system. In the maize-rye intercropping system, the intense rye rooting structure improves the water permeability around the root system and the soil structure, which is beneficial for the companion silage maize. It was also revealed that land with multiple plant species is more resilient compared to a monocultured area due to higher soil biodiversity (TIBBETT et al. 2020).

It also important to highlight the potential of cover crop in improving the growth and yield quality of maize cultivated in contaminated soil. Maize-cowpea intercropping increases the phosphorus availability and maize yield in alkaline soil rhizosphere compared to monocultured maize and cowpea. The change in pH level, rhizobial symbiosis and increase in soil respiration due to microbial and root activity in the intercropping system are linked to the increase in phosphorus level (LATATI et al. 2014). CHI et al. (2024) reported that *Brassica juncea* L. planted between maize rows helped in increasing maize yield cultivated in cadmium-polluted areas. The green mustard species improves the maize rhizosphere microecological properties by extracting the metal particles from the soil into their parts (CHI et al. 2024). Pearl millet intercropped with mung bean and cowpea shows an increase of up to 55% in yield compared to monocropped millet (TRAIL et al. 2016). Furthermore, the intercropping of maize and legume also improves the maize yield production exposed to high saline conditions (WANG et al. 2022). The presence of microbial

communities in the rhizosphere bacteria and fungi enhanced maize growth, net photosynthetic rate, and crucial antioxidant enzyme activities. Antioxidant enzymes activation triggered the reduction in proline content and ROS production thus, diminishing the oxidative and osmosis stress under salt stress conditions (WANG et al. 2022). Similar observation was recorded with sorghum-soybean intercropping, which is positively beneficial to the soil quality and sorghum yield cultivated in coastal salt pans (ZHU et al. 2022). The intercropping system produced the highest sorghum yield elevation compared to row-cropped systems. Similarly, the intercropping system reduced the soil salinity level while increasing soil organic matter content, soil microbial population, soil enzyme activity and soil nitrogen and phosphorus contents (ZHU et al. 2022).

In Europe, several maize intercropping researches conducted with wheat (GOU et al. 2016), beans (FISCHER et al. 2020), and red fescue (MANEVSKI et al. 2015) were published. In Northern Germany, intercropping silage maize with several bean cultivars produced significantly higher yield volume compared to monoculture maize. Besides that, the presence of beans in the harvest increases the protein content of the animal feed (FISCHER et al. 2020). Moreover, intercropping maize with red fescue (*Festuca rubra* L.) on sandy soils in Northern Europe has been shown to significantly reduce 15 - 37% nitrogen leaching while maintaining crop yields (MANEVSKI et al. 2015). Additionally, maize and bean intercropping also proved to improve the wild honeybees and bumblebee activities in the area (HÜBER et al. 2022). In contrast, a study in the Netherlands showed that the intra and interspecific competition in wheat and maize intercropped negatively affected the maize biomass and thousand kernel weight (GOU et al. 2016). Meanwhile, in Hungary, several studies involving winter cereals and winter peas have been carried out in recent years, proving the benefits of the intercropping system to be adopted by cereal farmers in the country (KRISTÓ et al. 2022; RÁCZ et al. 2023; VÁLYI-NAGY et al. 2024).

In the long term, intercropping may allow sustainable agriculture intensification to ensure sustainable food security and a better replacement for high-intensity monoculture practices. The benefits of cover crops in the agriculture sector are well-known and accepted by farmers and agriculture producers, especially in improving soil health and protection. Nowadays, it is a common practice to incorporate cover crops in one of the crop rotation cycles before or after the production of the main cash crop. A recent study reported the effect of pre-season cover crops in increasing subsequent maize growth and yield, which was achieved by improving soil water content and maize leaf and root water potential. The incorporation of cover crops, especially those with fibrous root architectures and extensive root distribution, increased rainwater infiltration while simultaneously reducing evapotranspiration due to the mulching effects provided by the cover crops (ALI et al. 2024).

However, several challenges were identified in intercropping cover crops into the main crop, which leaves wide research gaps to be explored. Intercropping limitations include challenges in mechanization, potential damage during harvesting, shading effects on light use efficiency, and perceived economic inefficiencies in existing systems (BLESSING et al. 2022). Besides that, factors such as appropriate cultivar selection, planting ratio, and interspecific competition can significantly influence the efficacy of intercropping systems (LAYEK et al. 2018). A successful intercropping is significantly influenced by soil nitrogen levels. In nitrogen-limited soils, intercropping can enhance soil organic carbon (SOC) more effectively than in nitrogen-rich soils (SUN et al. 2024).

Therefore, it is important for farmers to acquire adequate knowledge, especially on crop combination, planting density, and nutrient application to implement intercropping on their land. Selecting compatible crops is essential to minimize competition and maximize yield. Incompatible species can lead to reduced productivity due to competitive inhibition (GLAZE-CORCORAN et al. 2020). Research undertaken in Switzerland regarding the direct sowing of wheat into an existing cover crop has revealed the competitive interactions between the cover crop and the primary agricultural crop (HILTBRUNNER et al. 2007). Despite these limitations, intercropping remains a promising strategy for sustainable agriculture, particularly in resource-limited settings and addressing these challenges is essential for maximizing its potential benefits (VINCENT-CABOUD et al. 2017).

Currently, intercropping systems have caught the attention of EU members as a sustainable way to produce the main cereals and legumes in the region. Due to the expanding population and the changing climate, which is threatening food production in the EU, a more resilient agriculture system is needed to secure food security. Therefore, the EU has granted a fund for a sustainability project called LEGUMINOSE, which focuses on improving the cereal monoculture cultivation methods in the region with cereals and legume intercropping system. The researchers and farmers work together to identify the best intercropping combination between various cereals and leguminous species and the best agricultural management to produce maximum yield with this agriculture practice. With only 2% of Europe's arable land currently adopting intercropping, this project is one of the ways to encourage the farmers within the region to implement the new practice in their land (LEGUMINOSE 2024).

Therefore, in this research, the impacts of cover crop intercropping in influencing maize crop cultivation were explored. The nutrient management was also investigated to determine the optimum nitrogen levels required. At the end of the studies, the best crop species and nitrogen

level will be recommended. Besides that, the potential use of these cover crops in alleviating abiotic stress was also studied. Alfalfa (*Medicago sativa* L.), mung bean (*Vigna radiata*), white mustards (*Sinapis alba*), crimson clover (*Trifolium incarnatum*) and red clover (*Trifolium pratense*) were chosen in this study with the focus of maximizing the maize yield production in the intercropping system and the potential of the cover crops in protecting the maize during abiotic stress conditions. Our previous study found that alfalfa has the highest tolerance against salinity stress compared to the other crops, while other study showed the root structure of this crop helps their survival during drought periods (KANG et al. 2011; KHALID et al. 2023a). Intercropping maize and soybean proved to produce higher crop yields in semi-arid conditions while saving 20-50% of water and land (RAZA et al. 2022). RAPHOLO et al. (2020) revealed that maize intercropped with lablab (*Lablab purpureus*) produced 242 kg/ha more yield than monocultured maize with similar soil water used between them.

3. METHODOLOGY

This study comprises six experiments, conducted either in the laboratory or the experimental field. The first experiment investigated the germination activities of three different cover crop species under two different temperatures and five different salt concentrations. This experiment was conducted to determine the salt stress tolerance among the three leguminous crops and the influence of the temperature on the germination performance. Meanwhile, the second experiment was designed to assess the combination effect of salinity and temperature on maize germination. The germination performance of 16 maize varieties under three different incubation temperatures, with and without salt stress was analysed with the best variety was chosen to be planted in the field.

Meanwhile, Experimental Research 3 was carried out to evaluate the effect of nitrogen on monocropped maize. The study compared the effect of nitrogen fertilization on maize growth and yield quality in two different years, 2022 and 2023. Five different levels of nitrogen were tested in this trial. Furthermore, Experimental Research 4 was an open-field pot trial conducted in 2024, investigating the potential use of cover crops in alleviating salinity and drought stress in maize growth development. Three different cover crop species were intercropped with maize plants and subjected to three different irrigation treatments to introduce drought and salinity stress.

The fifth and sixth experiments were intercropping field experiments focused on the integration of various cover crop species in maize cultivation under different nitrogen levels. These two experiments were carried out to evaluate the influence of intercropping different cover crop species on maize vegetative development, yield production, and yield quality and to determine the effect of nitrogen levels in intercropped maize cultivation. Experimental Research 5, conducted in 2023, investigated the effect of three leguminous cover crop species on maize development and yield under five different nitrogen levels. Meanwhile, in 2024, Experimental Research 6 compared the effect of two leguminous and one *Brassicaceae* cover crop species on maize development and yield under the same five nitrogen levels as in 2023.

The methodology of each experiment will be described in details in the separated sub-chapters.

3.1 EXPERIMENTAL RESEARCH 1: Germination test of cover crop under different temperature and salinity levels

3.1.1 Experimental Materials

The trial was conducted in the laboratory of the Crop Production Institute of the Hungarian University of Agriculture and Life Sciences, Gödöllő, Hungary. For this trial, three species of leguminous crops were used: *Medicago sativa* var. Plato (alfalfa), *Cicer arietinum* var. Dora (chickpea), and *Trifolium pratense* var. Altaswede (red clover) were chosen. The seeds were germinated at four different saline treatments and two different temperatures. Sodium chloride (NaCl) was diluted with distilled water to produce 0.5%, 1%, 1.5% and 2% salt solution. 0% solution consisting of distilled water was used as the control. Seeds were surface sterilized with 5% Hypo solution for 5 minutes and rinsed with distilled water three times.

3.1.2 Growing Conditions

All seeds were germinated on 13.5 cm petri dishes containing single-layer Whatman filter paper. A Memmert-type climatic chamber with 70% moisture was used to control the growing temperature at 10°C and 20°C. Each treatment was repeated four times with each petri dish filled with 20 seeds, except for chickpea, with only 10 seeds per petri dish. All seeds were allowed to germinate at each treatment for 10 days. The number of seeds germinated was counted, and the length of plumule and radicles developed were measured using a ruler on the 10th day after the treatment started.

3.1.3 Statistical Analysis

The data collected were analyzed using Microsoft Excel 2010 for charts, while IBM SPSS Statistics 27 was used for the analysis of variance (ANOVA). Data were subjected to a three-way analysis of variance to explain the effect of different crops, salinity and temperature on the germination parameters. The normality test and the homogeneity of the variance were tested prior to ANOVA to ensure they were not violated. The mean value of the treatment was compared with the least significant difference (LSD) at $p < 0.05$. A post hoc test for multiple comparisons using the least significant difference (LSD) was also used at $p < 0.05$.

3.2 EXPERIMENTAL RESEARCH 2: Germination test of 16 maize varieties under different temperature and salinity levels

3.2.1 Experimental Materials

Sixteen maize varieties were used to study the interaction of salinity and temperature on maize seed germination quality, summarised in Table 4. The seeds were obtained from a local producer, Cereal Research Non-profit Ltd, Szeged, Hungary, and the Center of Agricultural Research, Martonvásár, Hungary. The maize variation studied in this trial consist of single cross (SC), double cross (DC), three-way cross hybrids (TC), parental lines, and one commercially available variety (V16). Variety 16 (V16) is a Hungarian dent maize variety called Margitta and is widely grown regionally. Margitta lies in FAO 280 group, is considered a cold tolerance variety, and has a short growing cycle. However, the producer did not publish the salinity and temperature tolerance.

Table 5. Maize variation tested for the germination performances (2022, MATE - Gödöllő)

Source	Genotype		Hybrid/parent
Martonvásár	V1	B1026/17	Parent
	V2	TK222/17	TC hybrid
	V3	TK15/DV	Parent
	V4	TK1083/18	Parent
	V5	TK623/18	SC hybrid
	V6	MCS901/19	Parent
	V7	TK256/17	DC hybrid
Cereal Research Non-profit Ltd	V8	GK155	Parent
	V9	GK131	Parent
	V10	GK 154x155	SC hybrid
	V11	Szegedi 521	SC hybrid
	V12	GK 154	Parent
	V13	GK 150	Parent
	V14	GK 140	Parent
	V15	GK 144x150	SC hybrid
Producer	V16	Margitta	Hybrid

3.2.2 Growing Conditions

The trials were conducted in the laboratory of the Crop Production Institute of the Hungarian University of Agriculture and Life Sciences, Gödöllő, Hungary. The seeds were germinated at three different temperatures (15°C, 20°C, and 35°C), with 20°C chosen as the optimal temperature for maize germination based on a recent study on a temperate maize hybrid (KHAEIM et al. 2022). Based on other studies, the best temperature for maize germination activity is between 25°C – 28°C, with temperatures below 10°C delaying seedling emergence (FAROOQ et al. 2008; BANO et al. 2015). Besides that, a study showed temperate maize hybrids are more vulnerable to high temperatures stress (>35°C) compared to tropical hybrids (CASALI et al. 2018).

Therefore, temperatures 15°C and 35°C were used to introduce the temperature stress conditions. Temperatures of 15°C and 35°C were considered the lowest temperature (LT) and highest temperature (HT), respectively, to initiate germination within nine days (KHAEIM et al. 2022). Two sodium chloride (NaCl) concentrations, 0 mM (control) and 100 mM (8.6 dS/m), were used to test the simultaneous effect of temperature and salt stress on germination. Normally, soil with electrical conductivity (EC) higher than 4 dS/m is classified as saline soil (HUQE et al. 2021). Several maize varieties were reported unable to germinate at EC of more than 8 dS/m, while several other varieties successfully germinated at EC of 15 dS/m (AHMAD et al. 2008; KHORASANI et al. 2017; MASUDA et al. 2021). Therefore, the experiment consisted of 6 different treatments: control (20°C + 0 mM NaCl), LT (15°C + 0 mM NaCl), HT (35°C + 0 mM NaCl), salinity (20°C + 100 mM NaCl), LT + salinity (15°C + 100 mM NaCl) and HT + salinity (35°C + 100 mM NaCl).

Before use, all of the seeds used in this trial were surface sterilized with 5% sodium hypochlorite for five minutes, rinsed with distilled water five times, and filtered to remove excess water. Each treatment was repeated four times for all 16 maize varieties. In each repetition, six seeds from each variety were placed on a 9 cm petri dish containing single-layer filter paper, which allowed enough space for the seedling to grow within 9 days. As the seed size varied between varieties, the solution volume was fixed at 10 mL for every petri dish. The Petri dishes were sealed with Parafilm sealing film to avoid moisture loss and were incubated in a Memmert climate chamber with a 70% humidity level. The germination percentage (GP) and the length of the radicle (RL) and plumule (PL) were recorded on days 3, 5, 7, and 9 of the experiment. Germination was considered to start when the radicle length was more than 0.1 cm. On the last day of the incubation, the root:shoot length ratio (R:S) were calculated based on the radicle and plumule length. The GP and SVI were measured using the following equations (MALIK et al. 2022).

$$\text{Germination percentage} = \frac{\text{Total number of seeds germinated}}{\text{Total number of seeds in petri dish}} \times 100$$

$$\text{SVI} = (\text{shoot length} + \text{root length}) \times \text{GP}$$

3.2.3 Statistical Analysis

Multivariate analysis of variance (MANOVA) and two-way analysis of variance (ANOVA) at 0.05 probability level was used to analyze the interaction between the independent variables and the dependent variables using IBM SPSS (Version 27, Armonk, NY, USA). Assumption tests, including normality, homogeneity, correlation, and data outliers, were also tested before the

parametric test. Before statistical analysis, GP and seedling growth data were arcsine and $\ln(x+1)$ transformed to ensure the normal distribution. However, untransformed data were presented in the figures for a more understandable visualisation. Bonferroni's correction was applied to the ANOVA result to correct the increased family-wise error. Besides that, the mean value of each treatment was compared with that of Games-Howell post hoc and tested at a 0.05 probability level. Microsoft Excel 2010 was also used for data management and to produce figures.

3.3 EXPERIMENTAL RESEARCH 3: The Effect of Nitrogen Fertilization on Maize Growth and Yield in Drought Condition

3.3.1 Experimental Site

A field experiment was conducted to investigate the effect of N levels on yield and quality of maize at an experimental plot of the Department of Agronomy, Hungarian University of Agriculture and Life Sciences, Gödöllő, Hungary, in 2022 and 2023 growing season from May to September. This experimental site is located in a hilly section at the coordinate of 47.595012°N, 19.369821°E, near-average climatic zone, and 242 m above sea level on sandy loam, brown forest soil (Chromic Luvisol). The humus content was 3.12%, with sand, silt, and clay contents 10%, 54%, and 36%, respectively, at the top of the 20 cm layer (TÓTH et al. 2018). The soil had a slightly acidic pH of 6.2 (H₂O) and a pH of 5.1 (KCl) (DEKEMATI et al. 2020). The data from the meteorological tower at the field recorded that in the growing season from May to September 2022, the total precipitation in the field was 209.4 mm and the average temperature was 20.6°C. July was the warmest month, with the daily average of 24.5°C, with maximum temperature reaching 38.6°C and minimum reaching 16.2°C. In the 2023 growing period, the total precipitation in the field was 288.7 mm, and the average temperature was 20.11°C during the same period. July was also the hottest month, with the daily average of 22.8°C with maximum temperature reaching 34.4°C. Figure 4 shows the comparison of the meteorological data in the field during the growing period for the year 2022 until 2024.

At the national level, Hungary received a higher amount of precipitation in 2023 compared to 2022, with 714 mm compared to 447 mm in 2022, which correlated with the higher number of rainy days compared to 2022. Furthermore, data also shows the country faced a higher number of heat days (days with daily maximum temperature was at least 30°C) in 2022, with 48 days compared to 42 days in 2023 (KSH 2024b).

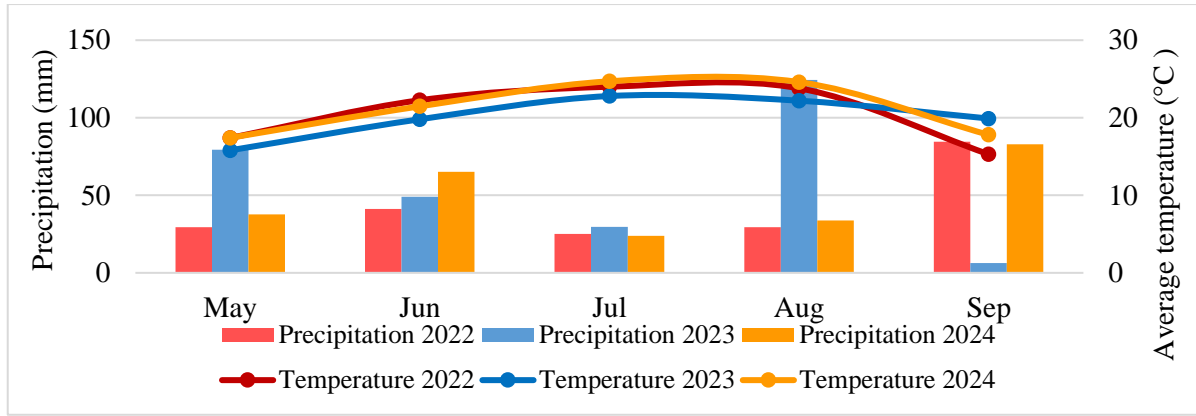


Figure 4. Precipitation and temperature data at the Gödöllő experimental field from May to September 2022-2024 (2024, MATE – Gödöllő).

3.3.2 Treatment and Experimental Design

The experimental design involved a randomized complete block design (RCBD) with three replications, with each replicate consisting of 10 plants. Maize seeds were sowed using a Wintersteiger Plotman planter, and the seeds were planted at a density of 75,000 plants per hectare, which was determined to be the optimal planting density for maize in this particular study. The experimental site consisted of five observation plots with N levels of 0 kg/ha, 50 kg/ha, 100 kg/ha, 150 kg/ha, and 200 kg/ha of net sizes 2 × 4 m. Ammonium nitrate were applied at week four after emergence as the main source of N in early June. Throughout the experiment period, weeding was carried out as needed to avoid any nutrient and water competition with the maize. All treatments were subjected to standard agronomic practices, including pesticides, weed control and other necessary inputs based on maize cultivation requirements.

3.3.3 Measurement

In general, all data collection consists of 3 phases: the vegetative stage data, physical yield parameters, and grains chemical quality. At the vegetative stage, the plant height, leaves area, and SPAD value (using SPAD-502 chlorophyll meter (Minolta Camera, Japan)) were measured from week four after emergence at the V6 stage up to the tasselling stage (VT). Following harvesting, the ear weight (g), ear length, total kernel per ear, kernel weight per ear and thousand kernel weight (TKW) were recorded. The 1000 kernels were counted using a Contador 2 seed counter, and the total weight was measured using a Scaltec electric weight balance. As the kernel moisture content reached around 12 – 11%, grain quality assessments including starch, protein, oil and moisture content, were carried out using MiniInfra Grain Analyser Scan-T Plus by Infracont.

3.3.4 Statistical Analysis

Multivariate analysis of variance (MANOVA) and two-way analysis of variance (ANOVA) at 0.05 probability level was used to analyze the interaction between the independent variables and the dependent variables using IBM SPSS (Version 27, Armonk, NY, USA). Assumption tests, including normality, homogeneity, correlation, and data outliers, were also tested before the parametric test. Pearson correlation test and Box's Test of Equality of Covariance Matrices were made prior to MANOVA testing on the dependent variables and were assumed to satisfy the assumptions. Bonferroni's correction was applied to the ANOVA result to correct the increased family-wise error. Games-Howell post-hoc test was used as $p < 0.05$. Microsoft Excel 2010 was also used for data management and to produce figures. The alphabets used in figures in the results section represent the post hoc results between each N treatment within the same year, with (a) representing the lowest value.

3.4 EXPERIMENTAL RESEARCH 4: The impact of intercropping in alleviating drought and salinity stress in maize cultivation (Pot trial)

3.4.1 Experimental Site and Materials

The pot trial was conducted in the open area of the experimental plot of the Department of Agronomy, The Hungarian University of Agriculture and Life Sciences, Gödöllő, Hungary, from June to October 2024. This experimental site is located in a flat section at coordinates of (47.594373°N, 19.368968°E), near-average climatic zone, and 242 m above sea level. Margitta maize variety was also used in this trial, while alfalfa (*Medicago sativa* var. Plato), white mustard (*Sinapis alba* var. Maryna) and mung bean (*Vigna radiata* L.) were used as the cover crops. Based on our findings, the Margitta maize hybrid was able to germinate in saline solution with 100 mM NaCl concentration, while alfalfa successfully germinated 1.5% (257 mM) NaCl (KHALID et al. 2023a; KHALID et al. 2023b).

3.4.2 Maize and Cover Crops Variety

This section explains the characteristic of the planting materials used in Experimental Research 4, 5 and 6. Margitta maize variety was used in both years, which is a Hungarian dent maize variety that is widely grown regionally. Margitta lies in the FAO 280 group, is considered a cold tolerance variety, and has a short growing cycle. In the Experimental Research 5 (2023), four different cover crop species were initially tested, including *Medicago sativa* var. Plato (alfalfa), *Trifolium pratense* var. Altaswede (red clover), *Trifolium incarnatum* var. Contea

(crimson clover) and *Trifolium repens* var. (white clover). The seeds were purchased from local producer. White clover was eliminated as the trial progressed as this species failed to be established in the trial plots. Whereas in Experimental Research 4 and 6 (2024), the trial with alfalfa (*Medicago sativa* var. Plato) were repeated in addition to white mustard (*Sinapis alba* var. Maryna) and mung bean (*Vigna radiata* L.).

The Plato alfalfa variety is a leafy German variety with an upright growing habit that is excellent winter-hardy and resistant to various diseases, with the highest height recorded at 88 cm (TURAN et al. 2017). This variety is commonly used as a livestock food source in hay and fodder form. This variety is also used as green fertilizer to improve soil structure while providing nitrogen and organic matter to the soil. The Altaswede red clover variety is a tall, quick-growing single-cut clover. This variety is also an excellent winter hardy variety and is able to flourish in a wide range of soil and growing conditions. Commonly used as a green fertilizer crop, this variety has long tap roots, allowing deeper root penetration, loosening soil and extracting nutrients from deeper layers, including phosphorus. This variety also proved to use less water in drought conditions and higher survival rates in terms of higher regrowing rates (LOUCKS et al. 2018). Furthermore, the Contea crimson clover or Italian clover is a winter annual clover species that is able to flourish even sown later in spring. It is recommended that this species be cultivated on dry sites, especially on light sandy loam soil with slight acidity or neutral pH. This species is also commonly used for grazing and hay production other than its soil amendment quality as green manure (GSM 2024a). *Vigna radiata* var. Crystal was the mung bean variety used in this experiment, and it originated in Northern Australia. This variety is suitable for spring planting and has good lodging resistance, with moderate resistance to diseases such as tan spots, powdery mildew, and halo blight. Able to produce 24-27% protein and up to 53.5% carbohydrate, which can produce up to 1420 kJ energy per 100g (AMA 2008; SKYLAS et al. 2017). Besides that, mung bean is suitable for intercropping due to its good shade tolerance (KHONGDEE et al. 2022). Lastly, *Sinapis alba* var. Maryna is a multifunctional variety that can be cultivated for seeds, green manure, and animal fodder. It has an excellent ability to improve soil quality and suppress soil-borne nematodes. However, it prefers fertile soils with good moisture retention, sufficient nutrients, and a neutral pH and requires higher water demand during the flowering stage (GSM 2024b).

3.4.3 Treatments and Experimental Design

Three maize seeds were initially sown in 26 cm pots and thinned to two plants per pot after germination. Each cover crop was sown around the pots with maize seeds in the middle on the

same day of the maize sowing. Alfalfa and white mustard were applied at the rate 2 g/m², while the mung bean was maintained at four plants per pot. The control pots only contain maize without any cover crops. All intercrop and monocrop pots were subjected to four replications. The pots were subjected to three different treatments: no stress (NS), non-irrigated to impose drought stress (DS), and salinity stress (SS), which were applied weekly at 100 mM NaCl (Photo 1.1 in A2 Appendices section). The NS and SS pots were irrigated with an automated irrigation system at the rate of 1.3 L per day (Photo 1.2 in A2 Appendices section), while the DS pots were irrigated once a week manually at the rate of 1.3 L per pot until the V7 stage. As the trials were carried out in the open area, all pots were exposed to 236.5 mm amount of rain during the trial period.

3.4.4 Measurements

The maize vegetative growth and development, including height, SPAD value (using SPAD-502 chlorophyll meter (Minolta Camera, Japan)), and leaves area, were recorded weekly one month after emergence until the beginning of the R1 stage. The soil moisture and soil temperatures were recorded using SGS07 digital soil meter (INHOCN, China) every two weeks prior to the daily and weekly irrigation schedule until R1. The number of flowering days was recorded as the tassel appeared, and cob weight and length were also recorded at harvest time (Photo 1.5 in A2 Appendices section). The dry mass of the aboveground material of the cover crop was also recorded at the end of the study.

3.4.5 Statistical Analysis

Multivariate analysis of variance (MANOVA) and two-way analysis of variance (ANOVA) at 0.05 probability level was used to analyze the effect of the independent variables on the dependent variables using IBM SPSS (Version 27, Armonk, NY, USA). Assumption tests, including normality, homogeneity, correlation, and data outliers, were also tested before the parametric test. Pearson correlation test and Box's Test of Equality of Covariance Matrices were made prior to MANOVA testing on the dependent variables and were assumed to satisfy the assumptions. The variables skewness and kurtosis value of all the variables were below 1. Tukey's post hoc test was used in this trial to determine the significant difference between the groups.

3.5 EXPERIMENTAL RESEARCH 5 & 6: Integration of various cover crops species on maize cultivation under different nitrogen levels

3.5.1 Experimental sites

The trials were conducted at the experimental plot of the Department of Agronomy, The Hungarian University of Agriculture and Life Sciences, Gödöllő, Hungary, in the 2023 and 2024 growing seasons with different cover crop species tested in each year. This experimental site is located in a hilly section at the coordinate of (47.595012°N, 19.369821°E), near-average climatic zone, and 242 m above sea level on sandy loam, brown forest soil (Chromic Luvisol). The humus content was 3.12%, with sand, silt, and clay contents 10%, 54%, and 36%, respectively, at the top of the 20 cm layer (TÓTH et al. 2018). The soil had a slightly acidic pH of 6.2 (H₂O) and a pH of 5.1 (KCl) (DEKEMATI et al. 2020). The data from the meteorological tower at the field recorded that during the growing season from May to September 2023, the total precipitation in the field was 288.7 mm, and the average temperature was 20.1°C. In 2024, the field received lower precipitation at 243.1 mm and a higher average temperature of 21.2°C during the same growing season. July was the hottest month with the daily average of 24.7°C and the maximum temperature reaching 36.1°C.

3.5.2 Experimental designs

The 2023 trial were conducted using randomized complete block design (RCBD) with three replications, with each replicate consisting of 10 plants, while the 2024 trial were conducted using a strip plot design with three replications, with each replicate consisting of 5 plants. Maize seeds were sowed using a Wintersteiger Plotman planter and were planted at a density of 75,000 plants per hectare, which was determined to be the optimal planting density for maize in this particular study. The cover crop seeds were applied one day after the sowing by broadcasting the seeds into the soil between the maize rows (Figure 5). The beans were planted at the rate of 4 plants per maize, while the alfalfa and white mustard were applied at the rate of 6 g/m². The clover species, including the white clover, were broadcasted at the rate of 4 g/m², and the control plot only contained maize plants. Photo 2.1, 2.2, and 3.3 in A2 Appendices section shows early stage of plant development in 2023 and 2024. After four weeks of maize emergence (V5-V6 stage), N treatment in the form of ammonium nitrate was applied at the levels of 0 kg/ha, 50 kg/ha, 100 kg/ha, 150 kg/ha, and 200 kg/ha in each of the intercrop and monocrop plot (Photo 2.2 in A2 Appendices section). Throughout the experiment period, weeding was carried out by hand as needed to avoid any nutrient and water competition with the maize and the cover crops. All

treatments were subjected to standard agronomic practices, including pesticides, weed control and other necessary inputs based on maize cultivation requirements.

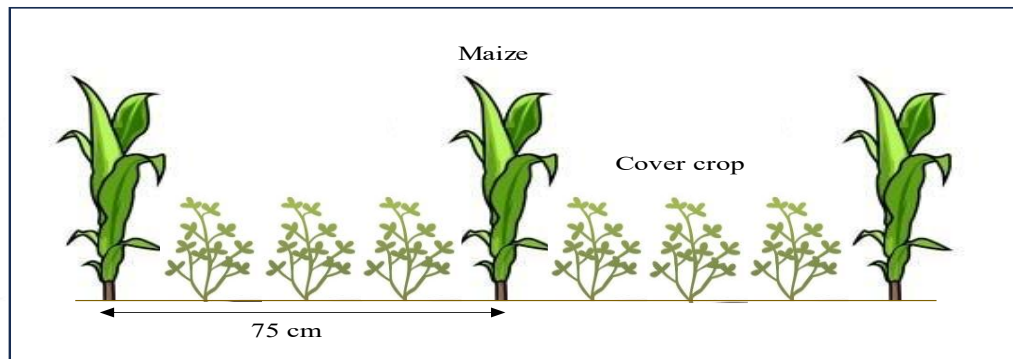


Figure 5. The planting distance between maize and the cover crops (2023 & 2024, MATE Gödöllő)

3.5.3 Measurements

At the vegetative stage, the plant height, leaves area, and SPAD value (using SPAD-502 chlorophyll meter (Minolta Camera, Japan)) were measured every two weeks from week four after emergence at the V6 stage up to the silking stage (R1). The leaf area of every plant was measured by multiplying the leaf length and the broadest midportion width of the leaf, which was multiplied by 0.75, which is a correction factor for maize leaf (OCHIENG et al. 2021). Meanwhile, the leaves area index was measured between the maize rows using LP-80 AccuPAR (METER Group, USA). At the beginning of August, the aboveground biomass of the cover crops was collected within an area of 0.5 x 0.4 m and dried to measure the dry mass. Following harvesting in September, the ear weight, ear length, total kernel per ear, kernel weight per ear and thousand kernel weight (TKW) were recorded. The 1000 kernels were counted using a Contador 2 seed counter, and the total weight was measured using a Scaltec electric weight balance. As the kernel moisture content reached around 12 – 11%, grain quality assessments including starch, protein, oil and moisture content, were carried out using MiniInfra Grain Analyser Scan-T Plus by Infracont.

3.5.4 Statistical Analysis

Multivariate analysis of variance (MANOVA) and two-way analysis of variance (ANOVA) at $p < 0.05$ was used to analyze the interaction between the independent variables and the dependent variables using IBM SPSS (Version 27, Armonk, NY, USA). Assumption tests, including normality, homogeneity, correlation, and data outliers, were also tested before the parametric test. Tukey's post hoc test was carried out to determine the significant differences between the groups. Microsoft Excel 2010 was also used for data management and to produce figures.

4. RESULTS AND DISCUSSION

4.1 EXPERIMENTAL RESEARCH 1: Germination test of cover crop under different temperature and salinity levels

4.1.1 Germination parameters at different temperatures and salinity levels

The three-way ANOVA results as shown in Table A.1 in the Appendices section, reveal that the three dependent variables significantly affected the three germination parameters, including germination percentage (GP), radicle length (RL), and plumule length (PL). The interaction of these variables also significantly affected all the germination parameters except the interaction of cover crop and the temperature treatment on the plumule length. Furthermore, the partial η^2 also shows salinity had a larger effect size on the three germination parameters compared to temperature. Meanwhile, the interactions between the dependent variables showed a small effect on the independent variables except for C x T which had a medium effect size on the RL (RICHARDSON 2011).

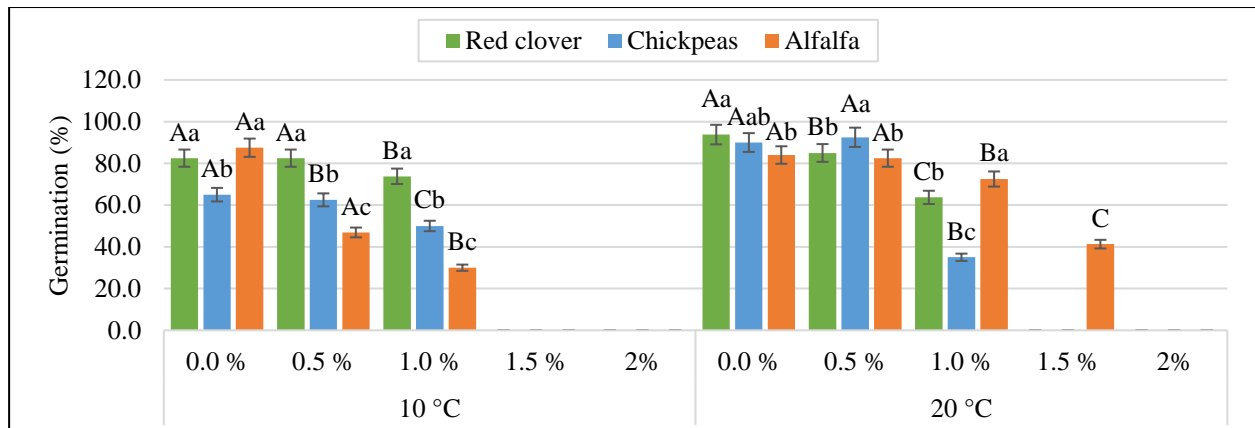


Figure 6. Germination percentage of different cover crops at two different temperature and salinity levels (2021, MATE-Gödöllő).

Capitals letters compare the difference between the same crop at the same temperature, incubated in different salt concentrations. The lowercase letters compare the difference between different cover crops exposed to the same salt concentration and temperature. Different letters indicate significant difference at $p < 0.05$.

In general, the GP of all three cover crop species decreased as the saline concentration reached 1% in both temperature conditions, and all three crops failed to germinate at 2% NaCl. Figure 6 shows there was no significant difference between the GP of red clover and chickpeas at 0% and 0.5% NaCl at 10°C. However, alfalfa showed a very significant decline in the GP as the salinity increased to 0.5% at the same temperature condition. The increase in temperature from 10°C to 20°C improved the GP of all three species that germinated in 0.5% and 0% saline solution. At 1% NaCl, alfalfa had the highest GP at 20°C but the lowest GP at 10°C. Meanwhile, red clover

produced the highest GP at 10°C compared to the other crops, while chickpea was severely affected by the 1% NaCl as the temperature increased to 20°C. Finally, only alfalfa seeds germinated at 1.5% NaCl at 20°C.

4.1.2 Radicle and plumule length in different salt concentrations

Figure 7 shows that the increase in salinity suppressed the radicle growth in all cover crop species in both temperature conditions. The increase in temperature only severely affected the radicle growth as the salt concentration reached 1% for red clover and chickpeas but not for alfalfa. Meanwhile, alfalfa produced the longest radicle at 20°C in the presence of NaCl, followed by red clover, while chickpeas produced the longest radicle in the presence of 0.5% NaCl at 20°C. Similarly to the GP, alfalfa also produced significantly the shortest radicle in 1% NaCl at 10°C compared to the other two crops in the same condition. Interestingly, alfalfa exposed to 1.5% NaCl at 20°C produced longer radicles compared to 1% NaCl at 10°C.

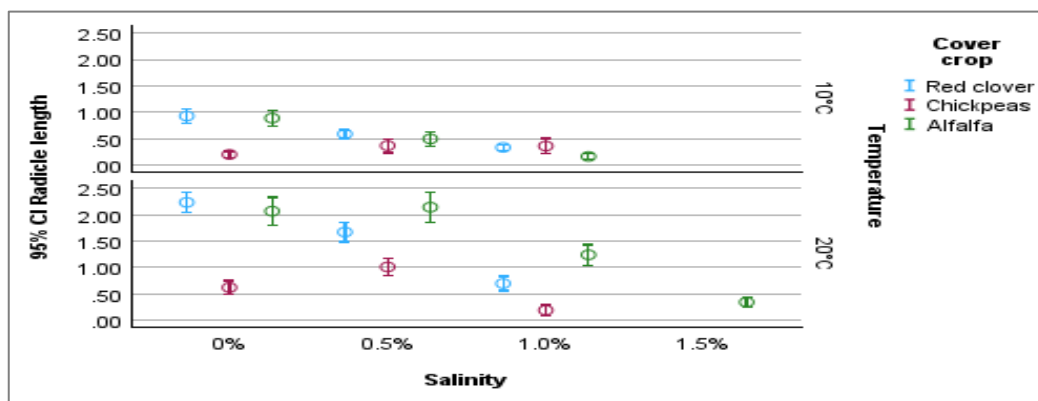


Figure 7. Average radicle length of each cover crop developed after 10 days incubated at two different temperature and salinity levels (2021, MATE-Gödöllő).

Based on Figure 8, similar results can be observed for plumule growth as the increased in salinity suppressed the plumule growth in all cover crop species at both temperatures. The temperature increased also negatively affected red clover and chickpea plumule growth at 1% but improved the growth of alfalfa. At 20°C, alfalfa produced significantly the longest plumule on average as the NaCl concentration gradually increased compared to the other crops. Meanwhile, in the presence of salt at 10°C, red clover produced the longest plumule but only significantly different from chickpea. Identically to the RL, chickpea also produced the longest plumule at the presence of 0.5% NaCl but were severely affected as the salt concentration increased to 1%. Finally, at 20°C and 1.5% NaCl, alfalfa also managed to grow longer plumule compared to at 10°C and 1% NaCl.

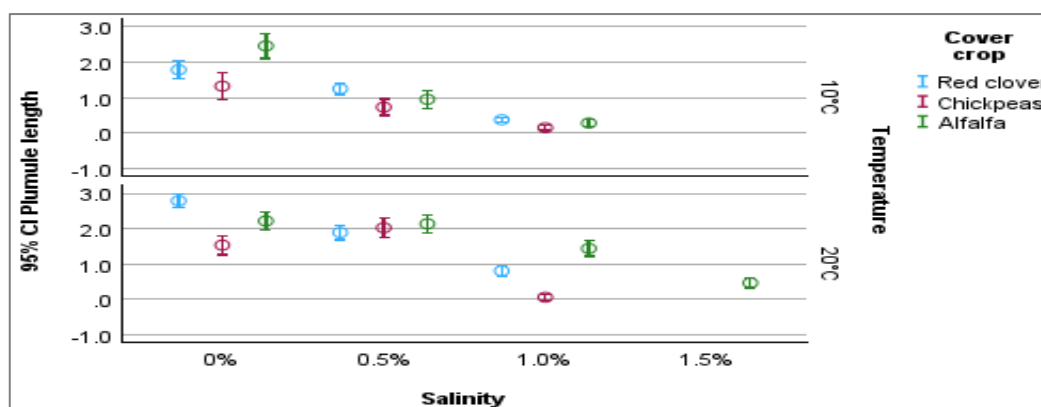


Figure 8. Average plumule length of each cover crop developed after 10 days incubated at two different temperature and salinity levels (2021, MATE-Gödöllő).

4.1.3 Discussion

In conclusion, based on the investigation, increases in salt stress negatively affected the germination and seedling growth of all three crop species in both growing temperatures. In this trial, elevation in salt concentration reduced the germination rate and inhibited radicle and plumule elongation on all three legume species. A study on other different legume species reported that salt stress inhibits embryonic axis growth in legume seeds. This defence mechanism against salt stress caused delay and stunted growth in the plumule and radicle of the seedlings (TLAHIG et al. 2021). NADEEM et al. (2019) also mentioned that leguminous species are more sensitive to salt stress at the seedling growth stage than at the germination stage.

Among all three crops, chickpea was the most sensitive to high salt stress condition at 20°C compared to at 10°C. This result supported the finding by (SINHA et al. 2020) which showed the increase in temperature amplified the salt stress in chickpea with reduction in the germination percentage and seeds vigour. However, even though chickpea is classified as sensitive to salt stress (KATERJI et al. 2012), but based on our results, a slightly saline condition at 0.5% did not affect the germination percentage of the chickpea variety used in this study.

Furthermore, based on the germination parameter results, it can be concluded that alfalfa has the highest salt tolerance compared to red clover and chickpeas. Our findings supported the results of several previous studies which published the moderate salt tolerance of alfalfa compared to red clover and the tolerance mechanism involved (NISTE et al. 2015; BHATTARAI et al. 2021). However, SHARAVDORJ et al. (2021) published that red clover showed higher germination rate than alfalfa at salt concentrations between 25 to 100 mM and temperature between 15 – 30°C. For comparison, in our experiment we tested NaCl concentration of 85.5mM, 171mM, and 256.5 mM. Besides that, as the journal did not mention the variety of the alfalfa and red clover used, the genetic variability between different varieties may cause the difference in both stresses tolerance.

Meanwhile, it was discovered that; tolerant alfalfa variety has moderate cell membrane damage and lower ROS accumulation than the sensitive variety. The salt tolerant alfalfa also able to maintain the leaf water content and prevent cells water loss during salts stress (QUAN et al. 2016).

Besides that, the germination data revealed that alfalfa had higher salt stress tolerance during germination at 20°C and was more sensitive to salt stress at a lower temperature of 10°C. SHARAVDORJ et al. (2021) also found 20°C was the best temperature for alfalfa germination at 0 to 100 mM NaCl compared to 15°C, 25°C and 30°C. Besides that, another study published temperatures between 18 – 29°C are the optimum temperature for alfalfa germination and lower temperatures will delay the process (KANKARLA et al. 2020). Another study suggested that at lower temperatures, the seeds' germination was delayed due to the suboptimal temperature to initiate the germination process (ZHANG, IRVING, et al. 2012). Besides that, a low temperature condition will increase ROS concentration while reducing various essential plant enzymatic reactions required for plant and eventually inhibit plant development (NIKOLIĆ et al. 2023).

Meanwhile, red clover produced higher germination percentage in 1% NaCl at 10°C than at 20°C. SHARAVDORJ et al. (2021) results showed no significant effect of temperature on red clover germination percentage in various salt concentration levels. The possibility of different variety used in their experiment should also be considered in comparing the findings. However, in our results, the seedlings growth was higher in the 20°C than in 10°C with longer radicle and plumule length. It was published that even though red clover able to grow in temperature range between 7 – 38°C but the optimum temperature for maximum growth is between 20 – 25°C (BOWLEY et al. 1984).

4.2 EXPERIMENTAL RESEARCH 2: Germination test of 16 maize varieties under different temperature and salinity levels

4.2.1 Germination Duration and Seedling Growth

The MANOVA results in Table A.2 in the Appendices section show that the number of days, salt stress (SS), temperature, and their interaction significantly influenced the dependent variables with $p < 0.001$ with medium and large effect size. Thus, follow-up ANOVA was carried out to determine the individual factors and their interaction on each of the dependent variables. ANOVA results in Table A.3 in the Appendices section show a significant effect between the independent variables and their combination on each of the dependent variables, i.e., GP, RL, and PL, with probability values ranging from <0.05 and <0.001 . The interaction of the three factors generated a small effect size on GP but a large effect size on RL and PL. The partial η^2 values also show temperature, and the salinity greatly affected RL and PL, while GP were more affected by

temperature than salinity. Similarly, the interaction between salinity and temperature had a larger effect on RL and PL than GP. Therefore, based on this result, the interaction between salinity and temperature significantly affected maize seeds' germination and the elongation of radicle and plumule.

Figure 9 shows the SS reduced the GP as the temperature increased from 15°C to 20°C and 35°C with the highest GP achieved at 15°C after 9 days of incubation. Even though the seeds germinated rapidly for the first 3 days at 35°C, compared to 15°C and 20°C, the percentage reached a constant level after 5 days of incubation. At 35°C saline condition, the rate of seeds germinating did not increase after 5 days of incubation and produced the lowest GP compared to the other two temperatures. Therefore, the data shows that at higher temperatures, the GP was more susceptible to salt stress than at lower temperatures.

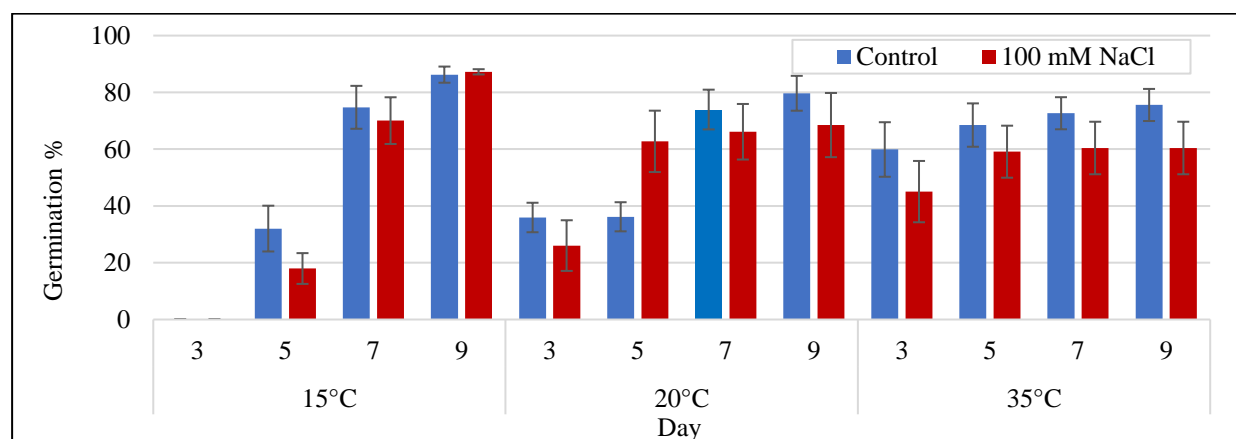


Figure 9. Overall germination percentage of all maize varieties at different days of incubation under different temperatures and salt stress conditions (2022, MATE-Gödöllő).

Meanwhile, the radicle and plumule growth results also produced a growing trend as the incubation days increased, especially for the control treatment (Figures 10 and 11). The results show that the control condition at 20°C was the optimum condition for the maize radicle and plumule growth compared to 15°C and 35°C. The results also show that the radicle and plumule growth had the shortest length at 15°C and only started growing on day 5, while the longest was at 20°C. However, at 35°C, the seedling growth, especially the radicle, was not improved as the incubation day increased for the SS seeds. Therefore, it can be concluded that SS inhibited the growth of maize radicle and plumule, and the temperature below and above the optimum level aggravates the stress responses. The significant interaction between SS and temperature will be further discussed in section 4.2.2.

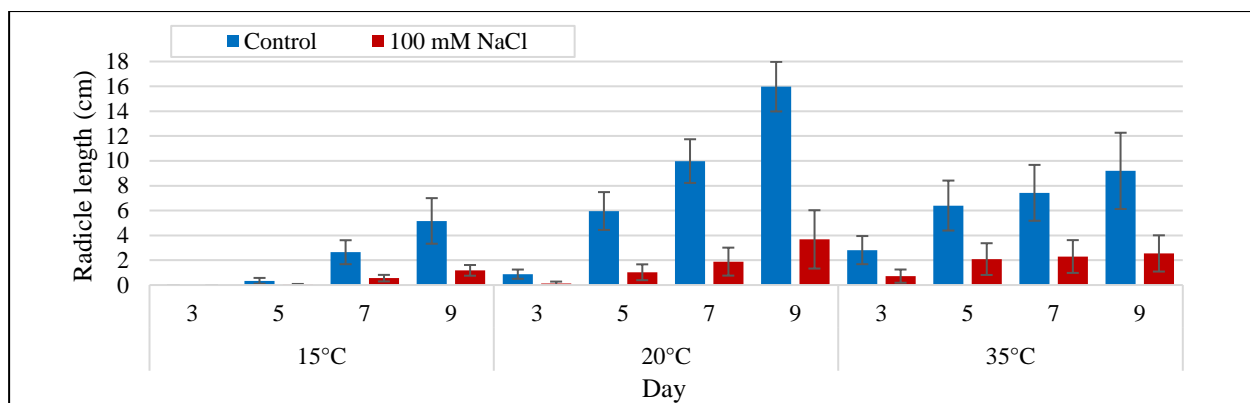


Figure 10. Growth of maize radicle at different days of incubation under different temperatures and salt stress conditions (2022, MATE-Gödöllő).

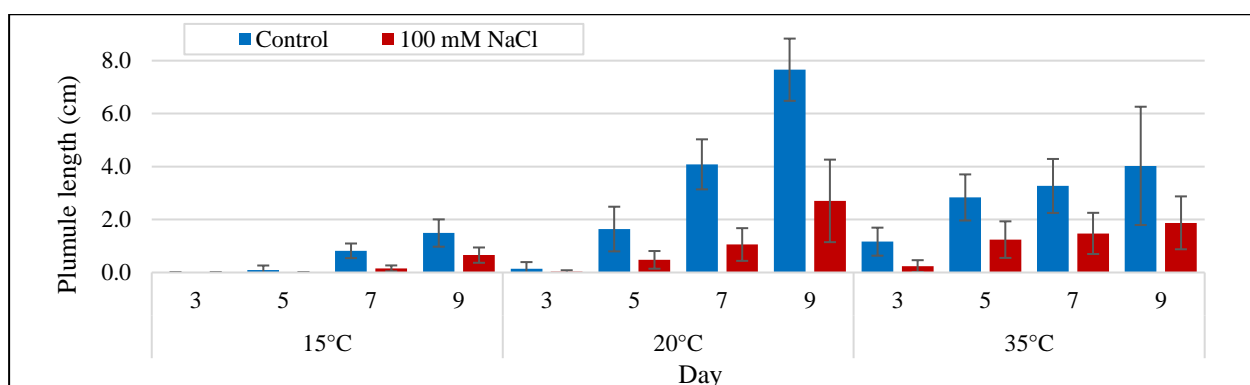


Figure 11. Growth of maize plumule at different days of incubation under different temperatures and salt stress conditions (2022, MATE-Gödöllő).

4.2.2 Interaction between Salinity and Temperature on Maize Germination

Figure 12 shows no significant difference in GP between the control and SS seeds at 15°C and 20°C, whereas, at 35°C, the control seeds show a significantly higher percentage than the SS seeds. Figure 12 also indicates that GP gradually increased as the temperature increased for the control seeds, whereas the salinity stress caused no significant difference in GP at 20°C and 35°C. The combination of salinity and 35°C caused the highest GP difference between the control and SS seeds at 12.3% than only 3.5% due to the salinity and 15°C combination.

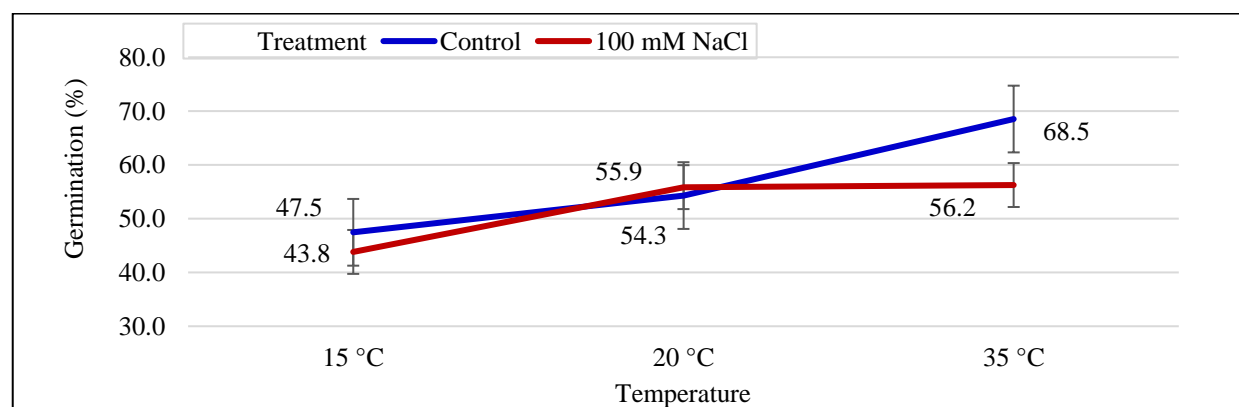


Figure 12. Overall germination performance of 16 maize varieties at two different salt concentration across three different temperatures (2022, MATE-Gödöllő).

Besides that, the presence of SS significantly reduced the length of both radicle and plumule as the temperature increased to 20°C and 35°C, while no significant effect of SS on maize germination at low temperatures was observed, as shown in Figures 13 and 14. The results also show that the RL and PL gradually increased as the temperature increased from 15°C to 20°C but reached a plateau as the temperature rose to 35°C. As the temperature increased to 20°C and 35°C, significant reductions in RL and PL were observed with the presence of SS compared to the control treatment.

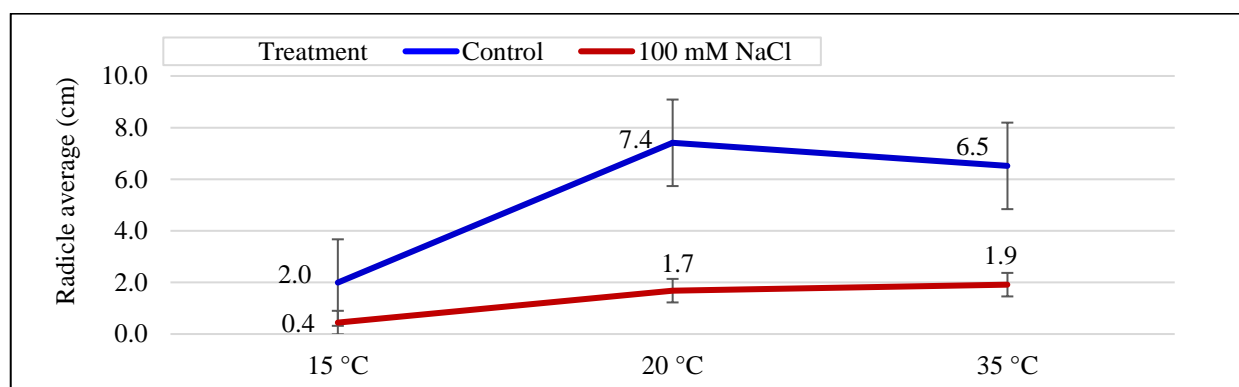


Figure 13. Radicle growth average of 16 maize varieties at two different salt concentration across three different temperatures (2022, MATE-Gödöllő).

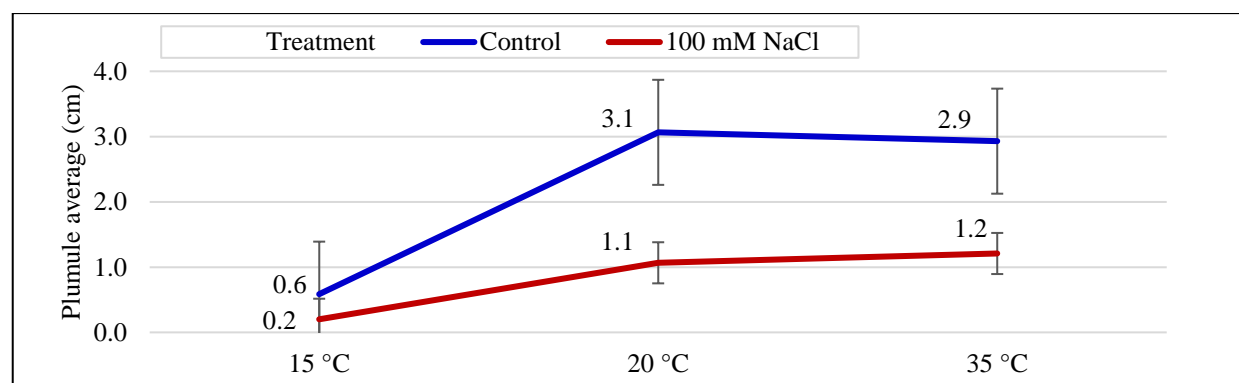


Figure 14. Plumule growth average of 16 maize varieties at two different salt concentration across three different temperatures (2022, MATE-Gödöllő).

Furthermore, the root:shoot length ratio (R:S) and seed vigor index (SVI) were also measured and tested with two-way ANOVA. The ANOVA results in Table A.4 in the Appendices section indicate a significant effect between the combination of salt treatment and temperature on the R:S and the SVI with $p < 0.001$. The partial η^2 shows that the interaction between the two factors had a greater effect on SVI compared to the R:S. Figure 15 shows that the seed produced the highest R:S ratio at 15°C, while there was no significant difference between the ratio at 20°C and 35°C in both control and SS conditions. The control had a higher ratio than the SS seeds, but both conditions produced a similar decreasing pattern as the temperature increased. Furthermore, Figure 16 shows that 20°C was the optimum temperature in both salt concentrations as it generated

significantly the highest SVI value compared to 10°C and 35°C. Meanwhile, the highest temperature at 35°C produced significantly higher SVI than at 10°C, showing that a temperature below the optimum has a more negative effect on seed vigor.

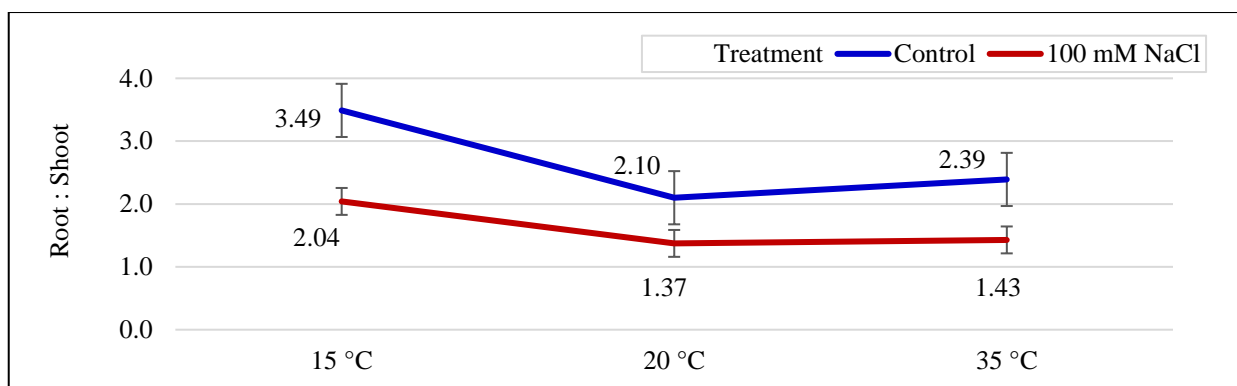


Figure 15. Maize seedlings root:shoot ratio comparison of at two different salinity levels treatments across three different temperatures (2022, MATE-Gödöllő).

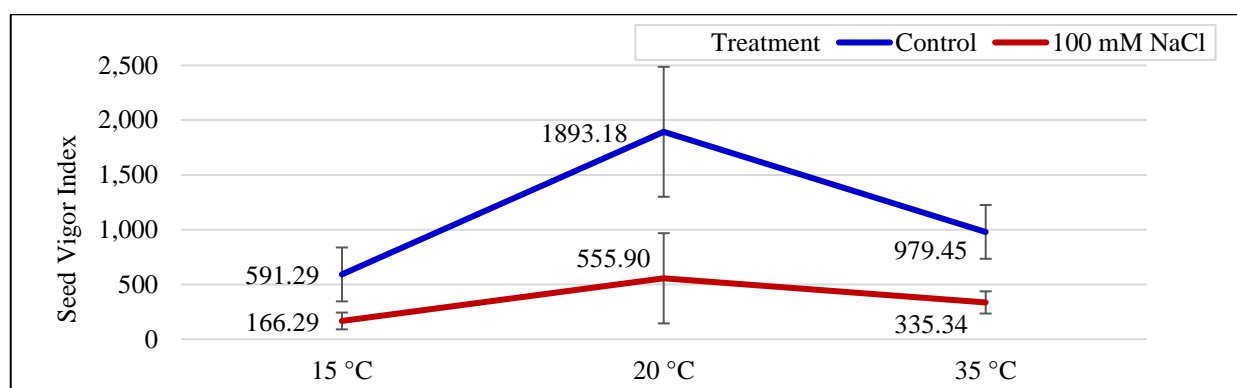


Figure 16. Comparison of maize seed vigor index (SVI) at two different salinity levels treatments across three different temperatures (2022, MATE-Gödöllő).

4.2.3 Comparison of Germination Performance between Varieties in Abiotic Stress Conditions

The Pillai's trace MANOVA results in Table A.5 in the Appendices section reveal that there were significant effects with p values <0.001 of variety, temperature, and their interaction on the germination parameters of the maize seeds incubated in 100 mM NaCl solution. The variety factor gave the largest effect size compared to the medium effect of temperature and the interaction between the factors. Thus, follow-up ANOVA was carried out and the result is presented in Table A.6 in the Appendices section.

The ANOVA results show that the independent variables, i.e., variety, temperature, and their interaction, significantly affected the GP, RL, and PL of maize seeds incubated in saline solution. Based on the partial η^2 , the maize variety showed a larger effect size on GP than RL and PL, whereas temperature had a larger effect size on RL and PL than GP. Finally, the interaction

between temperature and variety had a large effect size on GP and medium effect size on RL and PL (RICHARDSON 2011). Lastly, the Games-Howell post hoc test results and the impact of different temperatures on the germination performance of 16 maize varieties exposed to salinity stress were summarised in Table 6, 7 and 8.

Under 100 mM NaCl condition, each variety showed the highest germination performance at different incubation temperatures. Variety 16 produced the highest GP at 15°C compared to the other 15 varieties, but there was no significant difference with other varieties. At 20°C, V1 generated a significantly higher germination rate compared to several other varieties, while V9 and V11 showed the lowest germination rate at the same temperature. V16 also produced the highest GP at 35°C, significantly different from other varieties except V10 and V14.

Table 6. Mean data of germination percentage of different maize varieties in incubated in 100 mM NaCl solution at different temperatures (2022, MATE-Gödöllő).

Var	15°C	20°C	35°C
1	50.00 ± 42.60 Aa	87.50 ± 17.74 Bfg	65.63 ± 21.31 Abde
2	40.63 ± 43.87 Aa	75.00 ± 40.82 Bd-g	59.38 ± 29.17 ABb-e
3	45.83 ± 42.82 Aa	67.71 ± 31.90 Ac-g	55.21 ± 23.35 Abde
4	34.38 ± 37.75 Aa	48.96 ± 26.85 Abcd	54.17 ± 20.64 Aa-d
5	34.38 ± 36.75 Aa	48.96 ± 13.94 Acde	72.92 ± 13.44 Bbde
6	36.46 ± 36.12 Aa	47.92 ± 23.47 Acde	47.92 ± 29.74 Aabc
7	35.42 ± 39.85 Aa	62.50 ± 31.91 Bd-g	53.12 ± 23.74 ABbd
8	40.63 ± 44.71 Aa	59.38 ± 29.79 Ac-f	61.46 ± 14.55 Abd
9	42.71 ± 37.99 Ba	13.54 ± 16.35 Aa	30.21 ± 13.90 ABa
10	54.17 ± 46.55 Aa	82.29 ± 23.94 Bfg	58.33 ± 16.00 Aefg
11	29.17 ± 33.05 Aa	16.67 ± 18.26 Aab	34.37 ± 32.47 Aac
12	48.96 ± 36.24 ABa	26.04 ± 18.23 Aabc	64.58 ± 23.47 Bdef
13	46.87 ± 40.92 ABa	59.37 ± 27.87 Bc-g	28.13 ± 23.35 ABa
14	54.17 ± 40.60 Aa	60.42 ± 27.81 Ac-g	68.75 ± 36.45 Bfg
15	46.87 ± 39.07 Aa	55.56 ± 26.48 Ac-f	64.58 ± 29.11 Abde
16	60.42 ± 12.55 Aa	80.21 ± 18.48 Bfg	80.21 ± 20.38 Bfg

Capital letters compare the difference between a variety at different temperatures, while the lowercase letters compare the performance between different varieties at a particular temperature. Different letters indicate significant difference at $p < 0.05$.

Furthermore, at 15°C, there was no significant difference in RL and PL between varieties. V10 produced the longest radicle at 20°C and was only significantly different from V9, V11, and V12. On the other hand, V16 produced the longest radicle at 35°C and was significantly different in several varieties. Besides that, 13 out of 16 varieties showed a significant difference in RL between 15°C and 20°C. Meanwhile, V4, V5, and V14 produced significantly higher RL at 35°C compared to 20°C while RL of the other varieties was not significantly different at 20°C and 35°C. Meanwhile, V16 produced the longest plumule at 35°C and was only significantly different from V9, V11, and V13. Finally, all varieties revealed no significant difference in PL between 20°C and 35°C except V4, V5, V12, and V14.

Table 7. Mean data of radicle length of different maize varieties incubated in 100 mM NaCl solution at different temperatures (2022, MATE-Gödöllő).

Var	15°C	20°C	35°C
1	0.62 ± 0.67 Aa	2.94 ± 2.08 Bd	2.55 ± 1.91 Bb-e
2	0.44 ± 0.57 Aa	2.22 ± 1.92 Ba-e	1.97 ± 1.26 Bb-e
3	0.54 ± 0.60 Aa	2.40 ± 2.00 Bb-e	1.21 ± 0.54 Bb-e
4	0.29 ± 0.37 Aa	0.81 ± 0.48 Ba-e	1.90 ± 1.30 Cb-e
5	0.29 ± 0.36 Aa	1.02 ± 1.12 Ba-e	2.56 ± 1.36 Ccde
6	0.36 ± 0.44 Aa	1.17 ± 0.94 Ba-e	1.62 ± 1.23 Ba-e
7	0.37 ± 0.58 Aa	1.86 ± 2.10 Ba-e	1.46 ± 0.63 Bbcd
8	0.40 ± 0.51 Aa	1.43 ± 1.46 Ba-e	1.85 ± 0.84 Bb-e
9	0.49 ± 0.56 Aa	0.33 ± 0.55 Aa	1.15 ± 0.89 Aab
10	0.61 ± 0.71 Aa	3.30 ± 2.83 Bcd	2.53 ± 2.05 Bde
11	0.24 ± 0.40 Aa	0.54 ± 0.90 Aabe	1.38 ± 1.59 Aabc
12	0.41 ± 0.49 Aa	0.62 ± 0.47 Aabce	2.00 ± 0.88 ABcde
13	0.47 ± 0.52 Aa	1.70 ± 1.59 Bb-e	1.02 ± 1.16 ABa
14	0.60 ± 0.67 Aa	1.85 ± 1.77 Bb-e	2.77 ± 1.88 Cde
15	0.37 ± 0.41 Aa	1.62 ± 1.79 Bb-e	1.95 ± 1.19 Bbcd
16	0.60 ± 0.67 Aa	2.77 ± 2.47 Bcd	2.81 ± 1.41 Be

Capital letters compare the difference between a variety at different temperatures, while the lowercase letters compare the performance between different varieties at a particular temperature. Different letters indicate significant difference at $p < 0.05$.

Table 8. Mean data of plumule length of different maize varieties incubated in 100 mM NaCl solution at different temperatures (2022, MATE-Gödöllő).

Var	15°C	20°C	35°C
1	0.28 ± 0.35 Aa	2.03 ± 1.79 Bc	1.44 ± 1.22 Bbcd
2	0.22 ± 0.32 Aa	1.49 ± 1.68 Babc	1.10 ± 0.85 Ba-d
3	0.24 ± 0.34 Aa	1.76 ± 1.88 Bbc	0.86 ± 0.54 Bbcd
4	0.15 ± 0.22 Aa	0.81 ± 0.97 Babc	1.45 ± 0.77 Cbcd
5	0.17 ± 0.27 Aa	0.64 ± 0.71 Aabc	1.76 ± 1.13 Bbcd
6	0.11 ± 0.22 Aa	0.73 ± 0.80 Babc	1.13 ± 0.95 Ba-d
7	0.13 ± 0.22 Aa	1.08 ± 1.34 Babc	1.06 ± 0.63 Bbcd
8	0.13 ± 0.21 Aa	1.13 ± 1.53 Babc	1.14 ± 0.68 Bbcd
9	0.22 ± 0.34 Aa	0.24 ± 0.42 Aa	0.78 ± 0.62 Aabc
10	0.28 ± 0.39 Aa	1.52 ± 1.50 Babc	1.19 ± 1.12 Bbcd
11	0.08 ± 0.16 Aa	0.34 ± 0.68 ABab	0.86 ± 0.96 Bab
12	0.23 ± 0.35 Aa	0.49 ± 0.48 Aabc	1.54 ± 1.05 Bcd
13	0.18 ± 0.26 Aa	1.02 ± 1.11 Babc	0.53 ± 0.68 ABa
14	0.25 ± 0.35 Aa	1.11 ± 1.28 Babc	1.57 ± 1.25 Cd
15	0.23 ± 0.34 Aa	0.97 ± 1.06 Babc	1.25 ± 0.80 Bbcd
16	0.32 ± 0.44 Aa	1.5 ± 1.50 Bbc	1.79 ± 1.03 Bd

Capital letters compare the difference between a variety at different temperatures, while the lowercase letters compare the performance between different varieties at a particular temperature. Different letters indicate significant difference at $p < 0.05$.

In conclusion, based on the germination parameters studied in this trial, V1 (B1026/17), V10 (GK154x155), and V16 (Margitta) displayed the overall best germination performance in all three temperatures under saline stress. In contrast, V9 (GK 131), V11 (Szegedi 521) and V13 (GK 150) were the most vulnerable to salt stress, especially at 20°C and 35°C. Meanwhile, all varieties revealed significantly similar sensitivity to the low temperature of 15°C and while most varieties produced the highest growth at 20°C, V4 (TK1083/18), V5 (TK 623/18), V12 (GK 154), and V14 (GK 144) generated the highest seedling growth at 35°C showing higher tolerance to salt stress in high temperature.

4.2.4 Discussion

Based on our results, temperature significantly influenced the germination qualities of maize seeds incubated in saline solution conditions, which indicates the role of temperature in maize salinity stress response. Combining 100 mM NaCl and a temperature of 35°C severely decreased the GP and seedlings' growth compared to high-temperature stress alone. At a lower temperature of 15°C, the difference in germination performances was insignificant with or without salt stress. Lower temperatures also delayed the germination initiation and seedling's elongation compared to higher temperatures in both conditions. Besides that, similar to our results, several studies presented that a temperature of 15°C delays the emergence of maize seeds in temperate and tropical maize varieties (SANTOS et al. 2018; KHAEIM et al. 2022). In another recent study, it was observed that young maize seedlings' growth was more affected by a combination of salinity and 14°C than at a higher temperature of 24°C (ALSHOAIBI 2021). However, in our study, the combination of cold temperature and high salinity only affected the SVI and R:S ratio significantly but no significant difference was observed in GP, RL and PL with or without salt stress in cold condition.

The combination of the two factors also generated similar results in the germination of other plant species, including *Sorghum bicolor* (AL-SHOAIBI 2020), wheat (NEELAMBARI and KUMAR 2018), several medicinal plants (NADJAFI et al. 2010), and three salt-resistant halophytes (SONG et al. 2006). Temperature plays an important role in cell elongation and plant division, including during germination. High temperature disrupts the cell's production, thus affecting the elongation of radicle and plumule (RIBEIRO et al. 2014). It was stated that salinity could cause problems for crops in two ways. Salt in soil solution decreases the water availability to roots or seeds due to osmotic stress, while accumulated salt in plant cells can reach toxic levels in plant tissue (ACOSTA-MOTOS et al. 2017; ZHAO et al. 2021). A study revealed that maize germination speed reduced as salt concentration increased; NaCl concentration below 80 mM did not affect maize germination and seedlings growth, while a more than 320 mM concentration caused root deformation (AHMED et al. 2017). Their findings supported our results which showed that salt concentration at 100 mM NaCl were detrimental to the 16 maize varieties germination and seedling growth.

In our study, we observed that salinity decreases seedling growth, especially at 20°C and 35°C. In rice crop, the combination of high temperature and salt stress reduced the shoot's fresh weight and dry matter immediately 5 days after exposure, followed by irreversible leaf damage and desiccation as the exposure continues (NAHAR et al. 2022). ZANDALINAS and MITTLER

(2022) published that multiple stressors could form a synergistic interaction which amplify the stresses larger than when the stress occurs individually. It was also discovered that numerous unique genes were upregulated under multiple stresses conditions, while the individual stress response pathway are downregulated (ZANDALINAS and MITTLER 2022). Meanwhile, a study on *Arabidopsis thaliana* proved that a combination of salt, heat, and mannitol stress, activated unique biosynthesis pathways, which are not a simple combination of individual stress responses. It was suggested that under combined stress, the plant might only activate the most effective gene as a defensive mechanism under a limited resource situation instead of triggering all genes responsible for individual stress (SEWELAM et al. 2014).

Additionally, genetic variability within a species leads to differential phenotypic responses among its varieties, even under optimal growing conditions. In our previous study, differential in varietal performance between 16 similar maize varieties was observed from germination until the reproductive stage under optimum growing conditions (OMAR et al. 2023). Maize varietal performance on various abiotic stress at different growth stages has been unveiled by previous studies (HAJLAOUI et al. 2010; HUQE et al. 2021). A study reported a significant difference in salt stress tolerance between two maize varieties, with one of the varieties tolerating salt concentrations up to 12 dS/m (VENNAM et al. 2024). Our previous study discovered that grain maize was more susceptible to salt stress and a combination of salt and temperature stress than sweet maize (KHALID et al. 2021).

The variation in stress tolerance amongst different varieties may be caused by the different activation level of genes responsible for stress tolerance between varieties (ROYCHOUDHURY and CHAKRABOUTY 2013). Abiotic stress tolerance is achieved by expressing multiple genes responsible for producing stress tolerance metabolites and regulatory proteins such as osmolytes, antioxidants, and ABA in the case of exposure to triggering stress levels. It was discovered that barley with higher tolerance towards combined drought and salinity stress accumulated a lower $\text{Na}^+ : \text{K}^+$ ratio, higher $\text{Ca}^{2+}\text{Mg}^{2+}$ ATPase activities, proline and water use efficiency, and lower lipid peroxidation due to higher antioxidant activity compared to the susceptible variety (AHMED et al. 2013). A study on *Bromus inermis* showed that the optimum temperature of 20°C alleviated the salt stress and improved the tolerance as the salt level increased during germination compared to lower and higher temperatures (LIU et al. 2021).

Besides that, from our result, it was observed that salt tolerance was present in parent and single cross hybrids. It was also observed that several hybrids did not perform well with exposure to a combination of salt and heat stress. Stress tolerance between varieties may be triggered by genetic variation within species caused by various factors, including germplasm varietal, induced

mutation, genetic engineering, or intraspecific hybridization (CUSHMAN and BOHNERT 2000). Genetic variability allows multiple reactions to stress factors due to different signal-transmitting mechanisms that would enable appropriate physiological and biochemical responses to tolerate stresses (HASANUZZAMAN et al. 2013). A study on 18 maize varieties found that variation in genotypic component was the biggest contribution to the total variation in reaction against salt stress between varieties (HUQE et al. 2021). For example, some maize varieties tolerate high soil temperatures, while others tolerate low temperatures to allow the germination process to start (SANTOS et al. 2018).

4.3 EXPERIMENTAL RESEARCH 3: The Effect of Nitrogen Fertilization on Maize Growth and Yield in Drought Condition

4.3.1 Vegetative stage

MANOVA results in Table A.7 in the Appendices section show a significant main effect of nitrogen treatment, year, and the interaction on the vegetative stage parameters with $p < 0.001$. Both nitrogen and year factors had a large effect size on the vegetative stage growth but year was greater than nitrogen. Meanwhile, the interaction between the two factors caused a medium effect size on the dependent variables. Thus, follow-up ANOVA was carried out and the results are presented in Table A.8 in the Appendices section.

The ANOVA results show the year, N treatments, and the interaction between these two factors significantly affecting maize plant height, leaves area, and chlorophyll concentration (SPAD value) at VT stage with $p < 0.05$. Based on the partial η^2 , the N treatment had a large effect size on height and leaves area and only medium effect size on the SPAD value. Meanwhile, the year had a large effect size on all three parameters, while the interaction between the independent variables caused a small effect on height and leaves area but a medium effect on the SPAD value.

Figure 17 illustrates the performance of the plant height in 2022 and 2023 in different N levels. The maize planted in 2023 exhibited greater height compared to that of 2022 at all nitrogen application rates. Besides that, the increment in nitrogen rate caused a significant increase in the height in both studied years. However, the maize height planted in 2022 increased more rapidly with the N amount increment compared to 2023. In 2023, the highest height was reached at 150 kg/ha N, while in 2022, the height continued to increase as the N treatment increased up to 200 kg/ha but with no significant difference with 150 kg/ha N.

Furthermore, the leaves area also showed the same pattern as the plant height as the maize plant produced larger leaves in 2023 compared to 2022, as illustrated in Figure 18. In both years, the N fertiliser increment generated a significant increase in the leave area. Figure 18 also reveals

that the largest leaves area was achieved at 150 kg/ha N in 2023, while in 2022, the leaves area reached the maximum at 100 kg/ha N and did not increase as the N amount increased.

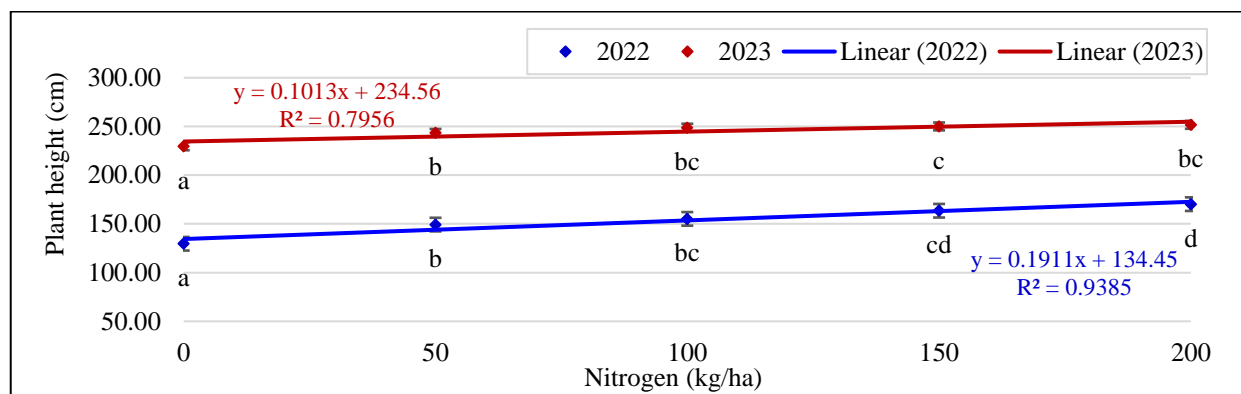


Figure 17. Plant height comparison of maize cultivated at five nitrogen level between 2022 and 2023 (2023, MATE-Gödöllő).

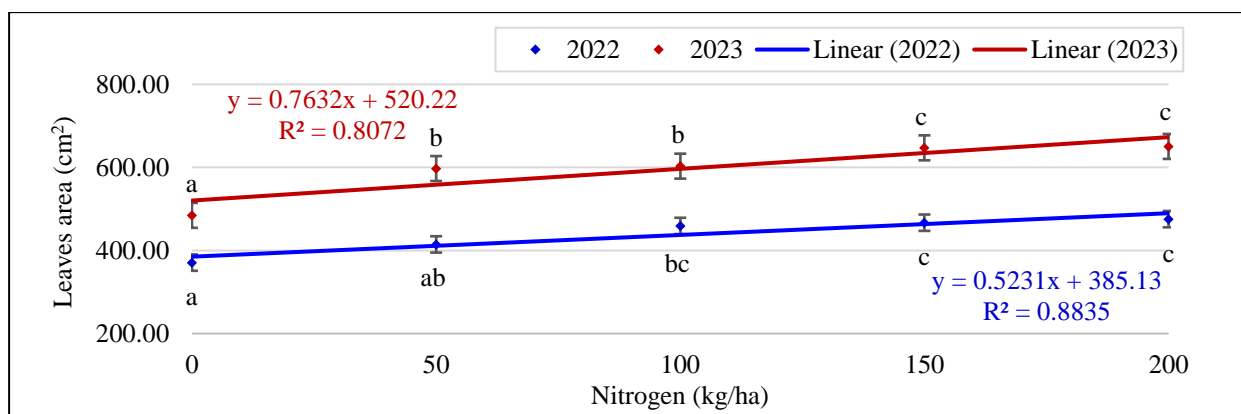


Figure 18. Leaves area comparison of maize cultivated at five nitrogen level between 2022 and 2023 (2023, MATE-Gödöllő).

Meanwhile, Figure 19 shows that maize planted in 2023 also produced a higher SPAD value compared to 2022, regardless of the N amount. However, the N treatment increment did not significantly affect the SPAD value of the maize planted in 2022 and showed a declining pattern as the N amount increased. In contrast, the SPAD value in 2023 increased as the amount of nitrogen increased.

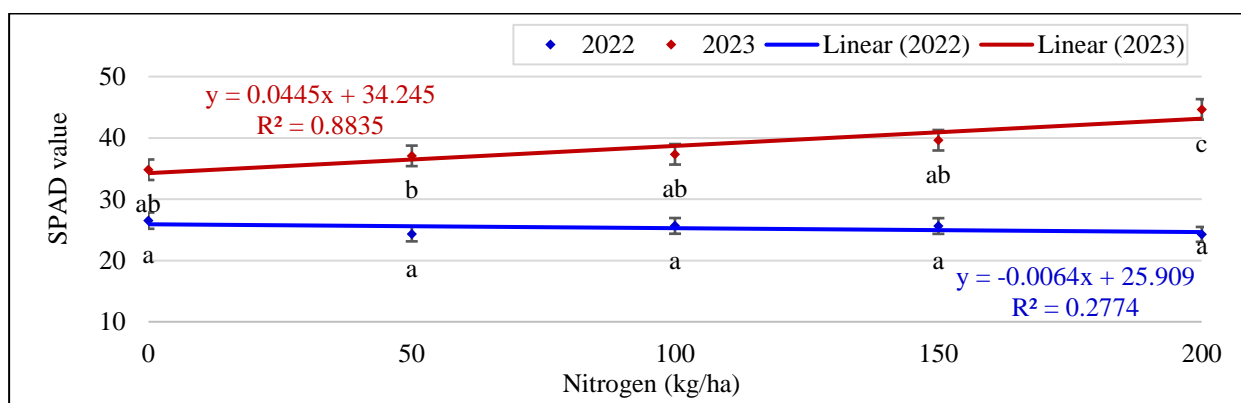


Figure 19. Comparison of the SPAD value of maize cultivated at five nitrogen levels between 2022 and 2023 (2023, MATE-Gödöllő).

4.3.2 Yield

The MANOVA results in Table A.9 in the Appendices section show, the year, N treatments, and the interaction between these two factors significantly affected the yield parameters. The partial η^2 and Pillai's trace values both show a large effect of the factors and their interaction on the dependent variables. Thus, ANOVA was carried out to determine the effect of the factors and their interaction on each of the parameters. The ANOVA results in Table A.10 in the Appendices section reveal, the year, N treatments, and their interaction significantly affected the maize ear weight, kernel weight, and kernel number per cob with $p < 0.001$. The ear length was not affected by the interaction between the factors. Based on the partial η^2 , both factors gave a large effect size on all the parameters individually. Meanwhile, the interaction between the factors also had a large effect on the yield parameters.

Figure 20 reveals that maize planted in 2023 produced longer ears compared to 2022, regardless of the nitrogen application rates. Besides that, an increment in nitrogen treatment caused a significant increase in the EL in both studied years. Maize planted in both years required an increment of 100 kg/ha N to produce a significant EL difference. In 2022, the EL reached the highest length at 150 and 200 kg/ha N as no significant difference was observed, while in 2023, the ear significantly increased in length as the N increased from 150 kg/ha to 200 kg/ha. Furthermore, the year 2023 also produced maize with ear weight bigger than 2022 in all N treatments tested (Figure 21). The weight increment in 2023 was also more rapid compared to 2022 as the N rate increased with both years reaching the highest weight at 150 kg/ha N.

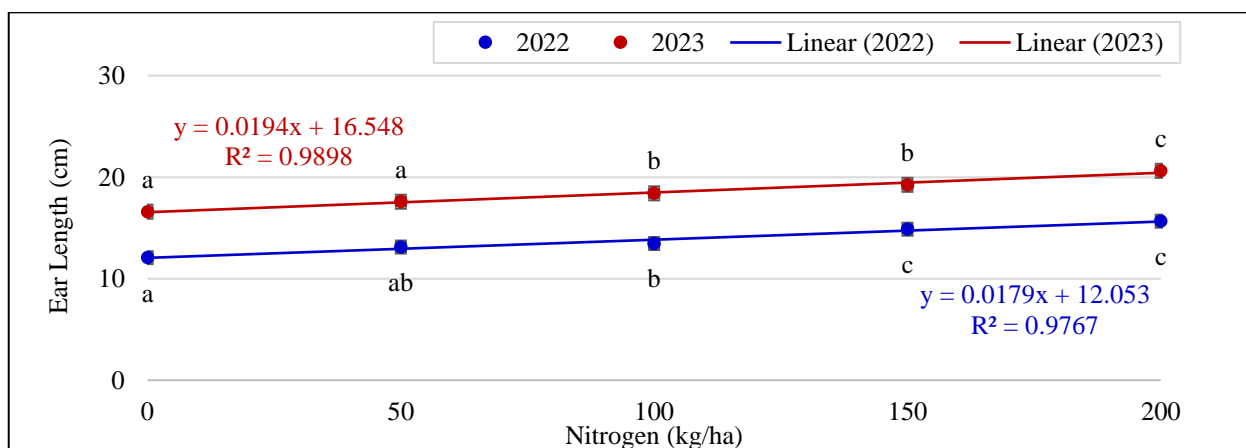


Figure 20. Ear length comparison of maize cultivated in five different N levels between the year 2022 and 2023 (2023, MATE-Gödöllő).

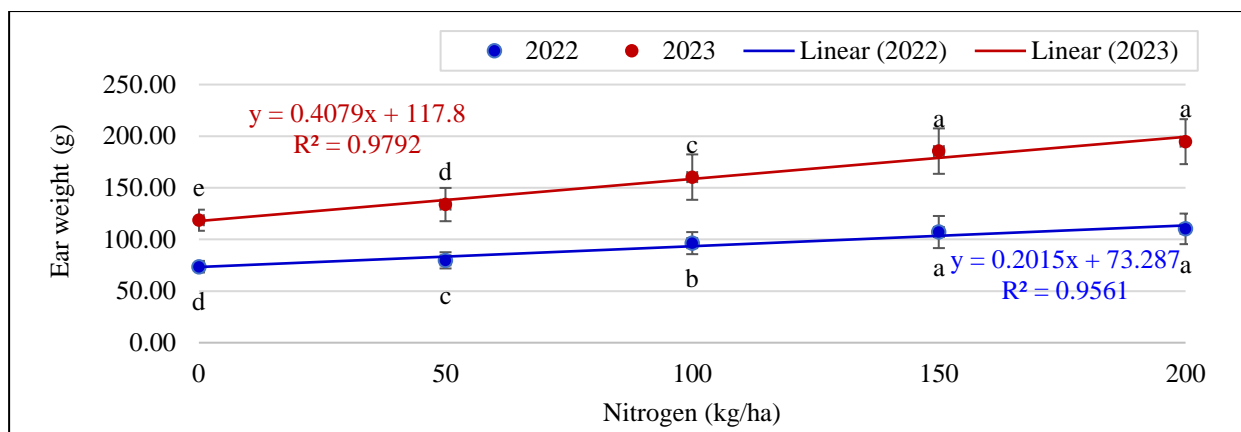


Figure 21. Ear weight comparison of maize cultivated in five different N levels between the year 2022 and 2023 (2023, MATE-Gödöllő).

Similarly, 2023 yield produced higher grain:cob ratio compared to 2022 in all N concentration tested (Figure 22). The year 2023 also produced a more rapid increase in GC ratio, while in 2022, the increase was more gradual as the N rate increased. In both years, the increased of N significantly increased the GC ratio up to 150 kg/ha N, while, the increased of N from 150 to 200 kg/ha also did not produce a significant difference for the GC ratio.

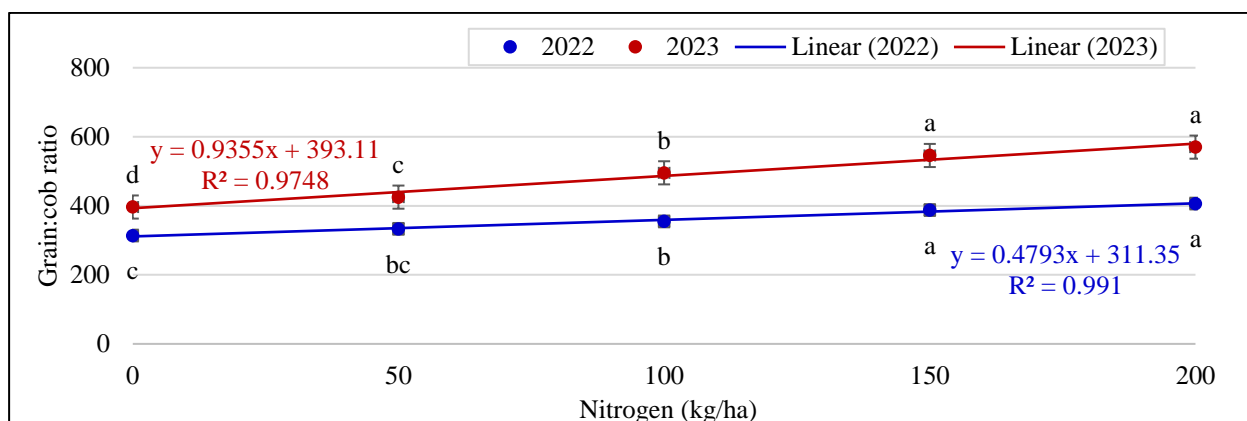


Figure 22. Comparison of G:C ratio produced by maize subjected to five different N levels in the year 2022 and 2023 (2023, MATE-Gödöllő).

A similar result was observed for the kernel weight per cob (KW) of the 2023 harvest, while the 2022 maize required a higher N rate to produce a significant KW difference (Figure 23). The 2023 harvest produced around 59.9% to 72.2% higher KW compared to 2022, depending on the N application rate. Table 9 reveals the estimation of GW that could be harvested in one hectare of land. The value was generated by multiplying the grain weight per cob with 75000 plants and converted to tonne/hectare. Based on the table, the values in 2022 and 2023 were higher than the national average for both years at 3.4 t/ha and 8.15 t/ha, respectively (KSH 2024a).

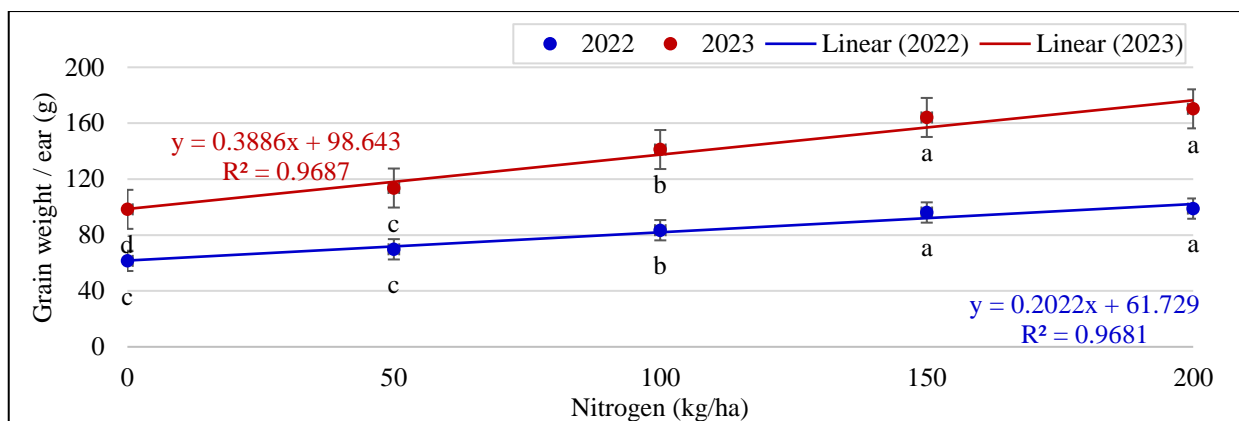


Figure 23. Comparison of grain weight produced per maize ear subjected to five different N levels in the year 2022 and 2023 (2023, MATE-Gödöllő).

Table 9. 2022 and 2023 maize grain weight converted from kg/ ear to tonnes/ha at five nitrogen level (2023, MATE-Gödöllő).

Nitrogen (kg/ha)	Yield estimation (tonne/ha)	
	2022	2023
0 N	4.61	7.38
50 N	5.23	8.52
100 N	6.26	10.59
150 N	7.21	12.31
200 N	7.42	12.77

Besides that, the ANOVA results in Table A.11 in the Appendices section show the year, N treatments, and their interaction significantly affected the thousand kernel weight (TKW) of the maize harvested with $p < 0.001$. Based on the partial η^2 , both factors and the interaction had a large effect size on the TKW.

Figure 24 illustrates that there was a significant difference between the TKW in both years, with 2023 producing higher TKW and more rapidly increased as the N rate increased compared to 2022. Besides that, the TKW in 2023 also showed significant increases at each of the N levels up to 150 kg/ha N. In contrast, the 2022 TKW was less sensitive to N rate increment and reached the maximum level at 150 kg/ha, while increasing the rate to 200 kg/ha did not significantly increase the TKW.

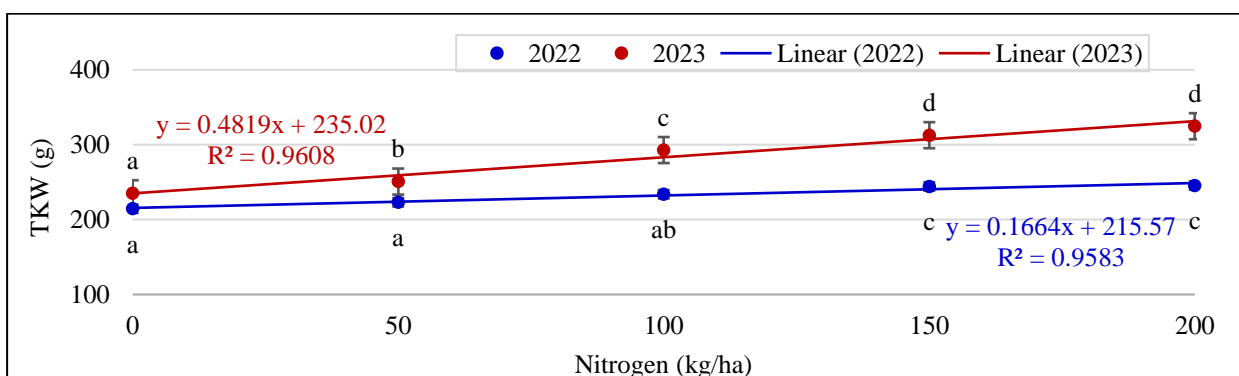


Figure 24. Maize thousand kernel weight (TKW) comparison between five nitrogen levels and two different years (2023, MATE-Gödöllő).

4.3.3 Grain chemical composition

The MANOVA results shown in Table A.12 in the Appendices section reveal the year, N treatments, and their interaction significantly affected the dependent variables with $p < 0.001$. The partial η^2 and Pillai's trace values both show a large effect of the factors and their interaction on the dependent variables. Thus, ANOVA was carried out to determine the effect of the factors and their interaction on each of the parameters. The ANOVA results in Table A.13 in the Appendices section show that the year significantly affected starch, protein, oil, and moisture content in the grains with $p < 0.001$, whereas the N treatment significantly affected all the components except the grain's moisture content. Besides that, the interaction of the two factors also had a significant effect on the grain chemical components, except the moisture content.

Similarly to the vegetative growth and the yield quantity traits, the 2023 grain starch content were higher than the grains from the 2022 harvest. Figure 25 reveals that the starch content in 2022 only significantly increased as the N levels reached 150 kg/ha, but increasing the rate to 200 kg/ha did not significantly increase the yield. In contrast to 2023, the starch content significantly elevated as the N level increased to 100 kg and continuously increased as the N reached 200 kg/ha. The starch content in 2023 increased around 1.4 – 2.5% from the 2022 values, depending on the N amount.

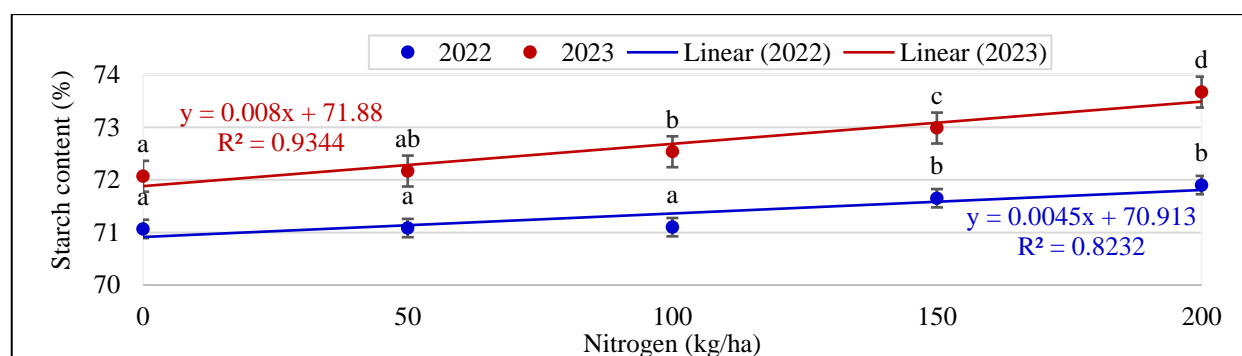


Figure 25. Starch content of maize subjected to five different nitrogen levels between the year 2022 and 2023 (2023, MATE-Gödöllő)

Interestingly, the 2022 grains produced higher protein content compared to the grains in 2023 (Figure 26). In both years, a significant increase in protein content was observed as the N fertilizer reached 100 kg/ha. In 2022, the protein reached the highest percentage at 100 kg/ha N, and further increments in N did not significantly increase this component. However, in 2023, increasing the N after 100 kg/ha continuously increased the protein content significantly.

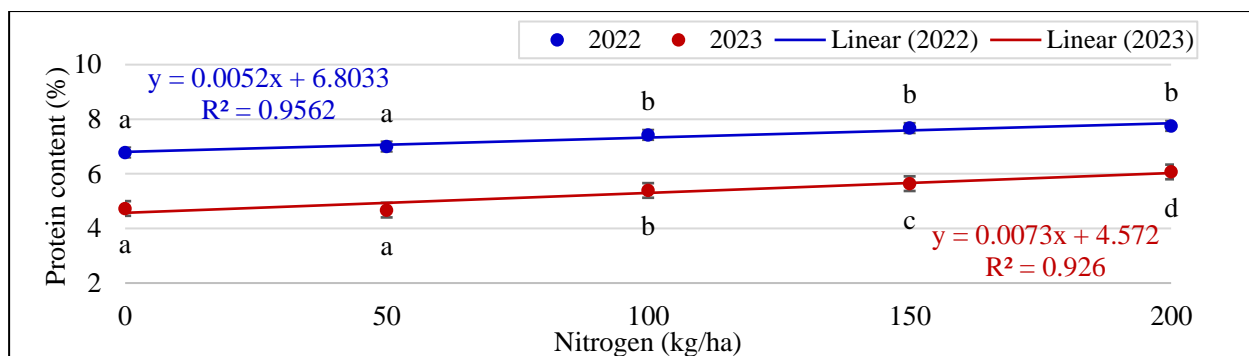


Figure 26. Protein content of maize subjected to five different nitrogen levels between the year 2022 and 2023 (2023, MATE-Gödöllő)

Similar to the protein content, the 2022 harvest produced higher oil content compared to the 2023 harvest, with both years showing an increasing trend as the N level increases (Figure 27). However, the significant difference could only be observed as the N level reached 150 kg/ha and above. In both years, the oil content increased significantly as we increased the N level, except for no significant difference at 100 kg/ha and 150 kg/ha N in 2023.

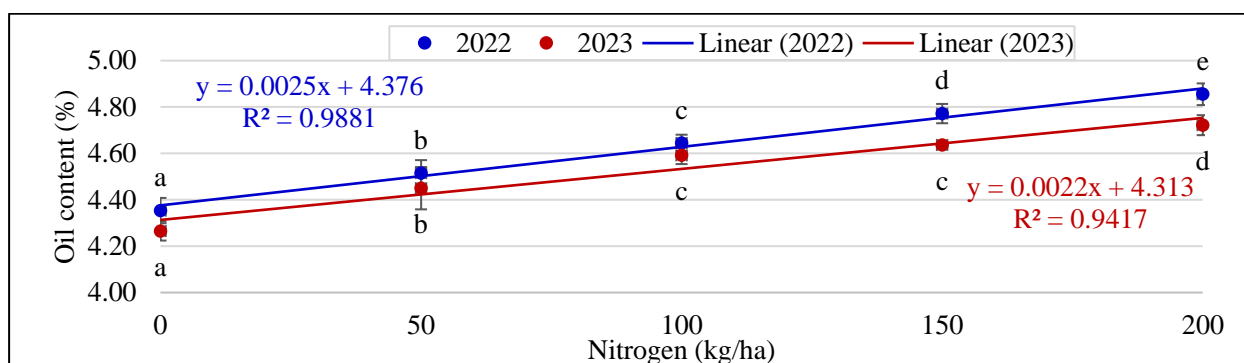


Figure 27. Oil content of maize subjected to five different nitrogen levels between the year 2022 and 2023 (2023, MATE-Gödöllő).

4.3.4 Discussion

As reported in the methodology section, the experimental field received 79.3 mm less precipitation in 2022 compared to 2023. The average temperature during the maize growing period in 2022 was also 1.7°C higher than the average temperature in 2023 (Figure 4). The year 2022 was considered a severe drought year in Hungary and the European region (TORETI et al. 2022; KSH 2024b). In Hungary, the drought period started in 2021 and ended in the early fall of 2022, which was in the path of the 2022 maize-growing period (BIRÓ and KOVÁCS 2023). In Hungary, the drought situation in 2022 not only affected the maize yield but also did considerable damage to other important crops, including wheat, barley, oat, soybean, sunflower, and rapeseed, with the production volume lower than in 2023 (KSH 2024a). In the European Union region, maize and sunflower were severely affected by the spring and summer drought of 2022, especially in the

Western Mediterranean and Carpathian-Balkan regions, contributing to revenue loss of up to 13 billion euros (PINKE et al. 2024).

Based on our results, the drought condition caused significant damage to the maize growth and yield harvested. It was discovered that among the important crops of Hungary, such as maize wheat, barley, and oat, maize is the most susceptible to precipitation changes, especially from June to August (CZIBOLYA et al. 2020). Increasing the N levels helps to increase the growth and yield, but it did not reach the same level as maize cultivated in better climatic conditions. BIRÓ and KOVÁCS (2023) reported that the peak drought in July and August delayed the plant cell development due to the absence of sufficient water, which caused a reduction in the final yield. A study in Debrecen, Hungary, shows drought stress in 2022 caused premature leaf senescence at the early vegetative stage and affected the leaf chlorophyll content (SPAD). However, they found that N fertiliser has less influence on the SPAD value compared to crop year (SZÉLES et al. 2023). A similar result was obtained in our study as the N fertilizer had no significant effect on the SPAD value in the dry year 2022 compared to 2023. Besides that, another study also highlights that dry and high-temperature conditions severely affected the SPAD value, which consequently reduced the final yield (HORVÁTH et al. 2019).

ZHAI et al. (2022) mentioned that one kilogram of nitrogen per hectare could produce around 0.023 – 0.057 tonnes of maize grain depending on the application rate, plant density, and hybrid used. Meanwhile, other studies show the rate of 0.0345 – 0.058 tonne grain generated by 1 kg N (WAJID et al. 2007; SHRESTHA et al. 2018; OCHIENG' et al. 2021). In our study, the rate of production was around 0.035 – 0.1 tonne in 2022 and 0.07 – 0.23 tonne in 2023 for 1 kg of N with 50 kg/ha N producing the highest grain weight per 1 kg N. SZÉLES et al. (2023) suggested the pre-sowing application of 120 kg/ha N is the optimum rate for base fertiliser and additional application of fertiliser at V6 and V12 did not improve the yield in the absence of rain. In our study, in the dry year of 2022, increasing the fertiliser rate to more than 150 kg/ha did not improve the vegetative growth and several yield parameters, including grain weight, TKW, and starch content significantly. Therefore, the results suggest application of N more than 150 kg/ha should be avoided by farmers to maintain the economic benefit. In contrast, increasing the N fertiliser in the wetter year significantly improved the maize growth and yield produced, except for protein and fat. In a rainfed agriculture system, water from rain has an essential function in solubilising and mobilising the N to penetrate the root zone and be absorbed by the plants (SUBHANI et al. 2012). NAGY (2012) suggested a higher N dose of 120 kg/ha should only be applied in the wet years and not in the dry years.

In addition, based on our results, the drought stress and high temperature of 2022 significantly decreased the starch content but increased the protein and oil content. Drought stress experienced throughout the grain-filling stage leads to a decline in photosynthetic activity, triggers premature senescence, and decreases the length of the grain-filling duration. Meanwhile, the high temperature inhibits the enzymatic activities responsible for starch synthesis (BARNABÁS et al. 2008). A study on wheat reported that in drought and high-temperature conditions, grains change their chemical composition, causing a reduction of starch accumulation but increasing in protein (DUPONT et al. 2006; RAKSZEGI et al. 2019). It was found that temperature and precipitation highly influenced maize protein content, with a 1.5% protein increase discovered in dry years compared to wet years in Hungary (GYŐRI 2017). Maize kernel increases their protein content, especially if the high-temperature stress occurs at the early grain-filling stage (MAYER et al. 2016). A study in wheat found drought stress increased the protein content and the β glucan in the grain, while the combination of drought and high temperature gave the highest protein (RAKSZEGI et al. 2014). In a study on maize, eliminating irrigation at the 12 leaf and flowering stages produced grain with higher protein content than the control treatment (AZADI et al. 2022). In addition, the phenotypic phase where the water stress occurs was proven to influence the final grain chemical composition in waxy maize (HUANG et al. 2023). Furthermore, a study also found that a dry period during the early reproductive stage (R1) will significantly threaten the yield produced (SZÉLES et al. 2023). In our study, R1 started in the first week of July, and the field only received 25 mm for the whole month of July 2022. A study on maize revealed that drought can enhance the oleic acid content in seed oil while reducing linoleic acid, thus altering the oil's fatty acid profile (ALI et al. 2013). Furthermore, sunflower crops under drought stress showed increased levels of palmitic and linoleic acids, although overall grain yield and oil quality were negatively impacted (GHAFARI et al. 2023).

4.4 EXPERIMENTAL RESEARCH 4: The effect of different cover crops on maize growth development under drought and salinity stress (Pot trial)

Based on our previous findings, the Margitta maize hybrid was able to germinate in saline solution with 100 mM NaCl concentration, while alfalfa successfully germinated 1.5% (257 mM) NaCl (KHALID et al. 2023a; KHALID et al. 2023b). In this trial, alfalfa and mung bean plants managed to be established and developed until the end of the experiment. However, the white mustard plants started to dry out in maize V6 stage in drought stress treatment (DS) and at the

beginning of maize R1 stage for the no-stress and salt stress treatment. The residues were left to cover the soil surface until the maize cob harvesting process.

4.4.1 Soil Moisture and Temperature

The results of MANOVA presented in Table A.14 in the Appendices section show a significant main effect of cover crop (CC) type, stress treatment, week and their interaction on the soil moisture and temperature with p values <0.001 . All factors and their interaction had a large effect size except for CC and stress treatment, which had a medium effect size. Thus, follow-up ANOVA was carried out and the results are presented in Table A.15 in the Appendices section. The ANOVA indicates that the three factors and their interaction significantly affecting the soil moisture and temperature with $p < 0.001$. Based on the partial η^2 , almost all the factors and their combination had a large effect size to the soil moisture and temperature.

Figure 28 shows that alfalfa and white mustard retained the soil moisture content in both no-stress (NS) and salt-stress (SS) treatments. Mung bean crops did not provide full soil coverage, causing significant moisture evaporation compared to the other cover crops. The absence of CCs in control pots also accelerates moisture evaporation from the soil in NS treatment. However, there was no significant difference in the moisture content in pots containing alfalfa and white mustard at SS condition. In contrast, alfalfa showed significantly higher moisture retention capability in drought stress treatment (DS) than the other cover crop treatments and control pots.

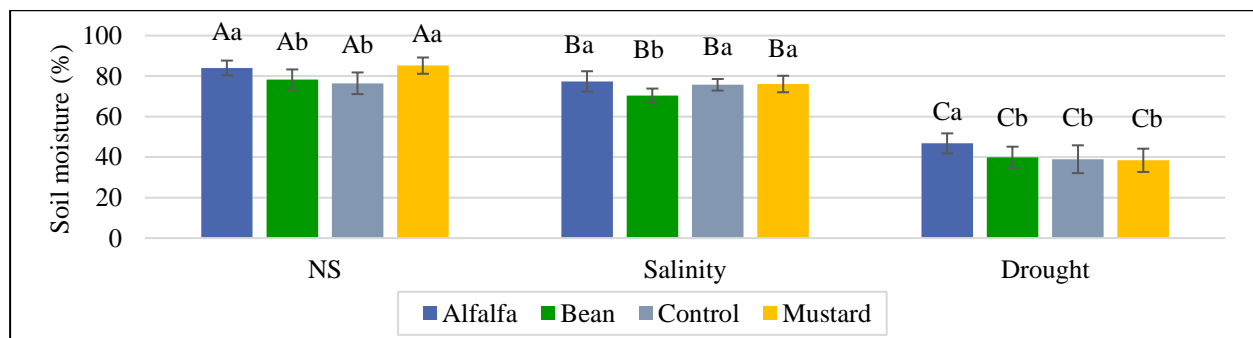


Figure 28. Average soil moisture content in pots containing maize intercropped with various cover crops and exposed to different abiotic stress treatments (2024, MATE-Gödöllő).

Capital letters compare the effect of the same CC treatment under different stress treatments while, lowercase letters compare the effect of different CC treatments under the same stress treatment on soil moisture. Different letters indicate significant difference at $p < 0.05$.

Meanwhile, the control pots retained the highest soil temperature in all stress treatments while, all three cover crops revealed no significant difference in reducing the soil temperature in NS and SS treatment (Figure 29). However, alfalfa proved to be superior in DS conditions,

followed by mung bean, while white mustard residues failed to lower the soil temperature in drought condition.

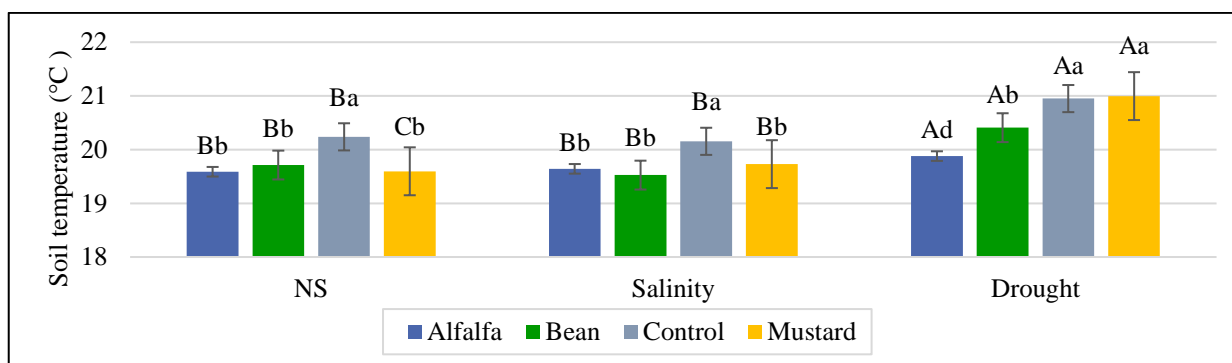


Figure 29. Average soil temperature in pots containing maize intercropped with various cover crops and exposed to different abiotic stress treatments (2024, MATE-Gödöllő).

Capital letters compare the effect of the same CC treatment under different stress treatments, while lowercase letters compare the effect of different CC treatments under the same stress treatment on soil temperature. Different letters indicate significant difference at $p < 0.05$.

4.4.2 Maize vegetative stage

Moreover, the results of the MANOVA in Table A.16 in the Appendices section show a significant main effect of CC type, stress treatment, week and their interaction on maize vegetative growth at $p < 0.001$. All factors show a large effect size on the dependent variables. Meanwhile, the factors interaction reveals a medium to large effect size on the vegetative growth parameters. Thus, follow-up ANOVA was carried out and tabulated in Table A.17 in the Appendices section. The ANOVA reveal the three factors individually significantly affecting maize SPAD value and height from V4 to VT stage with $p < 0.05$. The interactions between the factors also showed a significant effect except for all three factors interaction which had no significant effect on maize height. Based on the partial η^2 , almost all factors had large effect sizes individually, while the interactions had a small and medium effect size.

Figure 30 and Figure 31 show that the presence of leguminous CCs significantly improved maize SPAD value and height compared to the absence of CC in salt stress condition. In the NS condition, there was no significant difference in maize SPAD value and height between the pots incorporated with leguminous CCs and control pots, except for pots with white mustard which produced the lowest value compared to the other pots. Meanwhile, the DS treatment affected the height and SPAD value more severely than SS, but the presence of alfalfa and mung bean significantly improved the value compared to control and white mustard. Among all three CCs, white mustard negatively affected maize height and SPAD value in the DS condition compared to the control treatment and the maize intercropped with alfalfa and mung bean. Photo 1.3 and 1.4 in

the Appendices section show the vegetative growth of maize intercropped with alfalfa and mung bean.

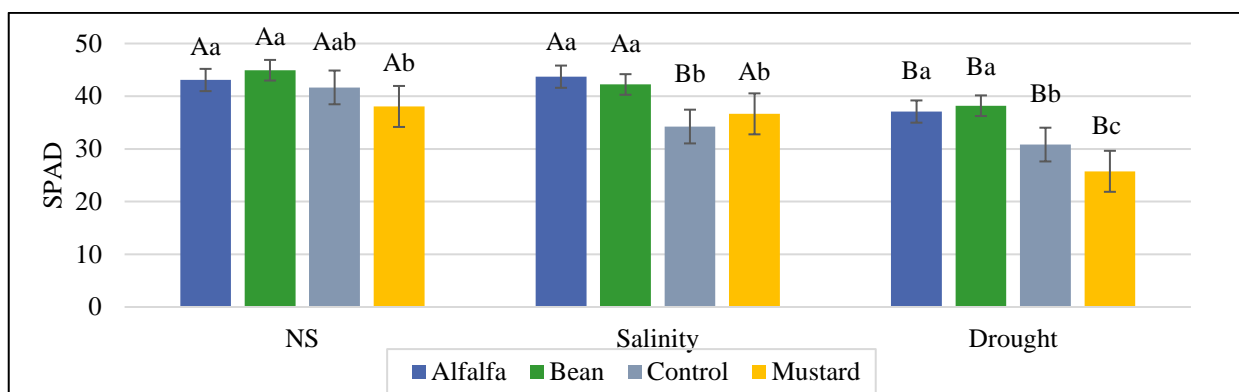


Figure 30. Average SPAD values of maize intercropped with different cover crops in different abiotic stress treatments (2024, MATE-Gödöllő).

Capital letters compare the effect of the same CC treatment under different stress treatments, while lowercase letters compare the effect of different CC treatments under the same stress treatment on SPAD value. Different letters indicate significant difference at $p < 0.05$.

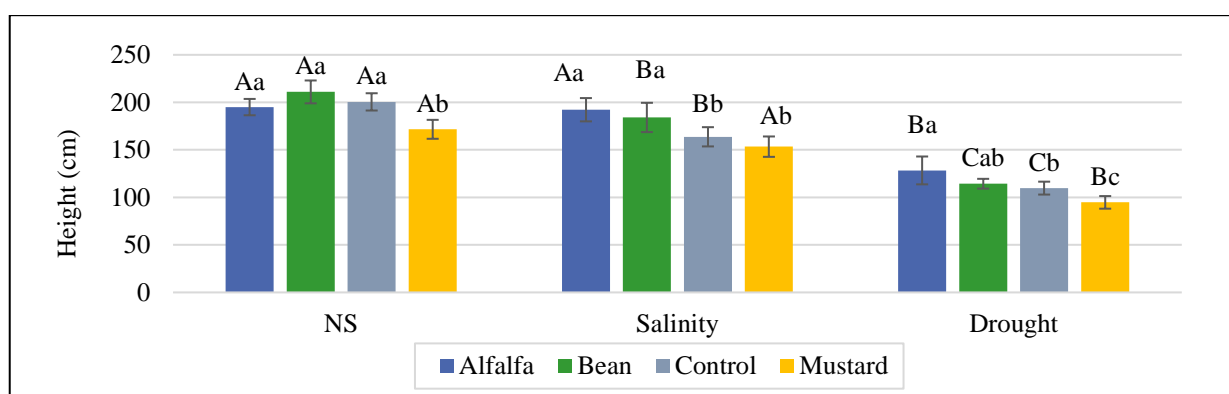


Figure 31. The average height at R1 of maize intercropped with different cover crops in different abiotic stress treatments (2024, MATE-Gödöllő).

Capital letters compare the effect of the same CC treatment under different stress treatments, while lowercase letters compare the effect of different CC treatments under the same stress treatment on plant height. Different letters indicate significant difference at $p < 0.05$.

Furthermore, the ANOVA results as shown in Table A.18 in the Appendices section also show that the CCs, type of stress treatments and their interaction significantly affected the maize leaf area with $p < 0.005$. Figure 32 shows that intercropping maize with mung bean significantly increased the leaf area in NS, while white mustard significantly reduced the maize leaf area. Leguminous CCs improved the leaf area in both stress conditions. DS condition severely affected maize in control and intercropped with white mustard more than the SS condition and when compared to maize intercropped with the leguminous crops.

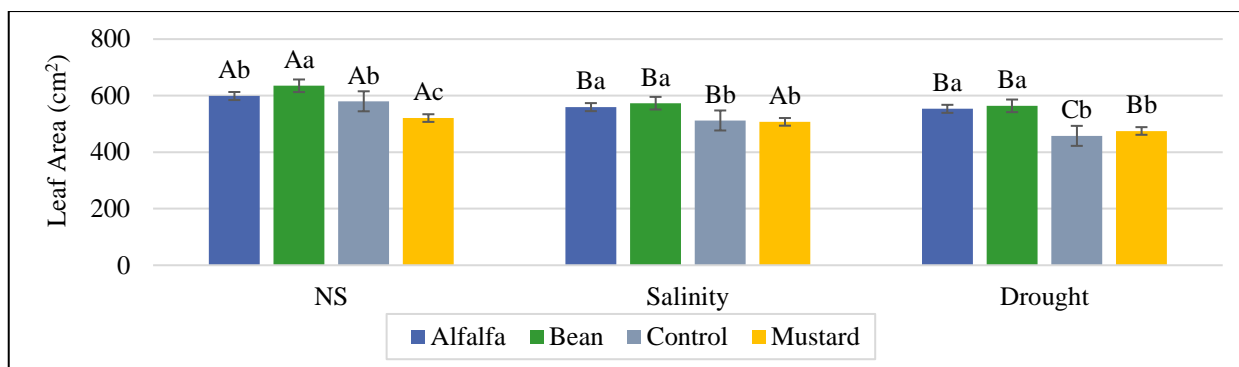


Figure 32. The average leaf area at R1 of maize intercropped with different cover crops in different abiotic stress treatments (2024, MATE-Gödöllő).

Capital letters compare the effect of the same CC treatment under different stress treatments, while lowercase letters compare the effect of different CC treatments under the same stress treatment on leaf area. Different letters indicate significant difference at $p < 0.05$.

4.4.3 Maize reproductive stage

The MANOVA results in Table A.19 in the Appendices section, present a significant main effect of cover crop type, stress treatment, and their interaction on maize flowering parameters with p values < 0.001 . Both factors and their interaction revealed a large effect size. The follow-up ANOVA results in Table A.20 in the Appendices section, shows that, the different stress and cover crop treatments and their interaction significantly affected the days of anthesis, silking and the anthesis-silking index (ASI) with $p < 0.05$ with large effect size.

Table 10 reveals the number of days after sowing (DAS) of the tassel and silk emergence in different CC and stress treatments. SS and DS delayed the emergence of both male and female florescence in maize regardless of the presence of any CC if compared to the NS treatment. However, the presence of leguminous CCs significantly improved the time for both tasselling and silking in SS, but only silking time was improved in the DS. No significant different in the tasselling days in DS in all CCs and control treatments was observed.

Table 10. Mean value \pm standard deviation of the days after sowing ((DAS) of maize tassel and silks emergence in different CC and irrigation treatments (2024, MATE-Gödöllő).

	NS		Salinity		Drought	
	Tasselling (DAS)	Silking (DAS)	Tasselling (DAS)	Silking (DAS)	Tasselling (DAS)	Silking (DAS)
Alfalfa	60.1 \pm 1.4Abc	65.4 \pm 2.1Ab	66.5 \pm 3.3Bb	75.1 \pm 3.5Bb	82.9 \pm 1.4Ca	94.8 \pm 1.4Cb
Bean	58.3 \pm 0.5Ac	63.8 \pm 1.2Ab	63.1 \pm 2Bb	71.8 \pm 2.3Bb	84.6\pm1.6Ca	96.4 \pm 1.5Cb
Control	68.3\pm4.7Aa	73.9\pm4.6Aa	74.8\pm2.6Ba	86.1\pm3.2Ba	84 \pm 2.8Ca	99.9 \pm 1.6Ca
Mustard	63.8 \pm 3.1Ab	70.5 \pm 3.2Aa	74.6 \pm 2.1Ba	85 \pm 2Ba	83.4 \pm 2.4Ca	99.9\pm2Ca

Meanwhile, the anthesis: silking index (ASI) in Figure 33 shows a similar pattern to the tasselling and silking days. In the NS condition, no significant difference was observed between the treatments, while SS and DS significantly increased the ASI value. However, the presence of

leguminous cover crops significantly reduced the ASI value in both SS and DS compared to white mustard, which produced significantly similar results with the control treatment.

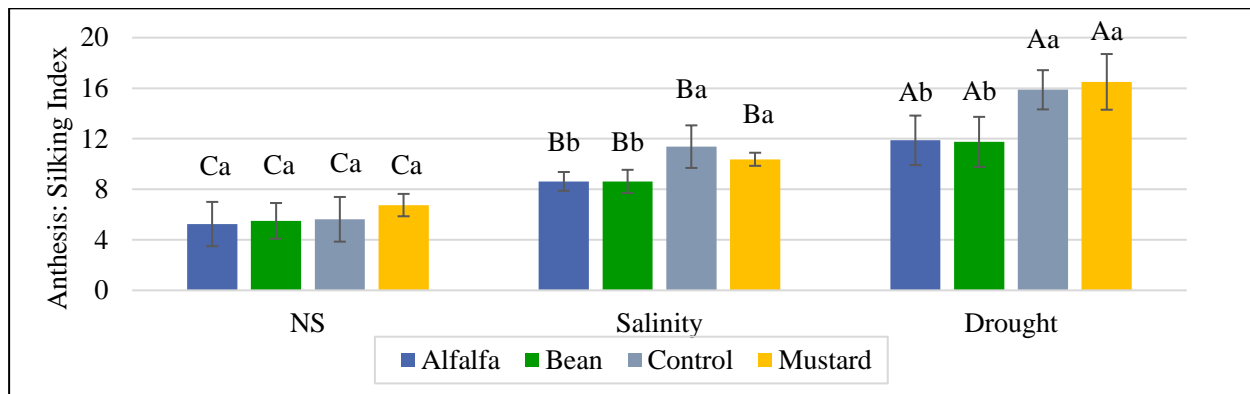


Figure 33. The anthesis: silking index (ASI) of maize intercropped with different cover crops in different abiotic stress treatments (2024, MATE-Gödöllő).

Capital letters compare the effect of the same CC treatment under different stress treatments, while lowercase letters compare the effect of different CC treatments under the same stress treatment on ASI. Different letters indicate significant difference at $p < 0.05$.

Furthermore, the MANOVA in Table A.21 in the Appendices section shows a significant large effect of cover crop type, stress treatment, and their interaction on maize yield produced with p values < 0.05 (Table 32). All factors and their interaction had a large effect, including the interaction of both factors. The follow-up ANOVA results in Table A.22 in the Appendices section show, individually, the different stress and cover crop treatments significantly affecting the ear weight and ear length with $p < 0.05$ and large effect size. However, the interaction between the two factors had no significant effect on the two parameters.

Meanwhile, the harvested cobs results show that both DS and SS reduced the maize ear weight (EW) and ear length (EL) compared to NS treatment with DS caused more detrimental effects than SS (Figure 34 and Figure 35). There was no significant difference in the EW and EL of maize intercropped with leguminous CCs in the NS condition, while the white mustard significantly reduced the values. In SS condition, both leguminous CCs also improved the EW, while white mustard had a deteriorating effect. In DS, alfalfa proved to improve the yield produced significantly more than mung bean. As the control and white mustard pots produced no cobs, no comparison could be carried out with these two treatments.

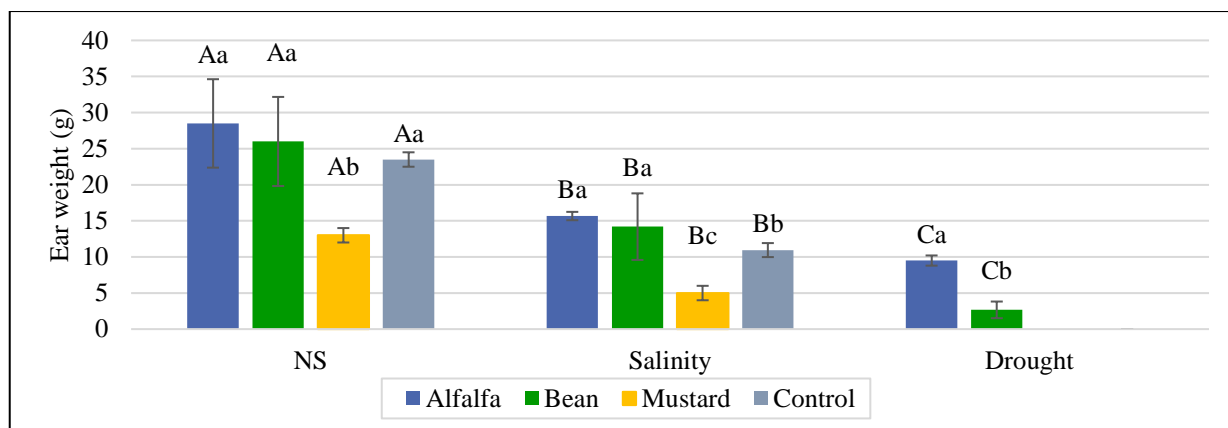


Figure 34. The average ear weight of maize intercropped with different cover crops in pots and subjected to different abiotic stress treatments (2024, MATE-Gödöllő). Capital letters compare the effect of the same CC treatment under different stress treatments, while lowercase letters compare the effect of different CC treatments under the same stress treatment on EW. Different letters indicate significant difference at $p < 0.05$.

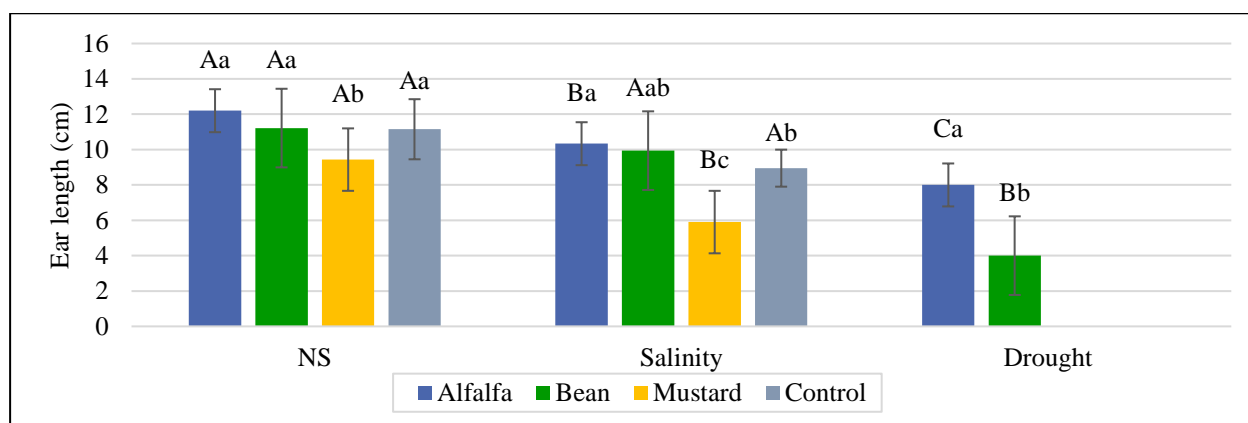


Figure 35. The average ear length of maize intercropped with different cover crops in pots and subjected to different abiotic stress treatments (2024, MATE-Gödöllő). Capital letters compare the effect of the same CC treatment under different stress treatments, while lowercase letters compare the effect of different CC treatments under the same stress treatment on EL. Different letters indicate significant difference at $p < 0.05$.

4.4.4 Cover crop dry mass

A one-way ANOVA was carried out to determine the effect of different stress treatments on the dry mass of alfalfa and bean aboveground residues harvested at the end of the trials. The mustard residue was omitted due to premature drying. Based on the ANOVA results in Table A.23 in the Appendices section, the stress treatments significantly affected the dry mass of alfalfa and mung bean at $p < 0.001$. Figure 36 shows that alfalfa produced significantly higher dry mass compared to beans in DS than SS. Meanwhile, mung bean dry mass was significantly higher compared to alfalfa in the SS than in DS.

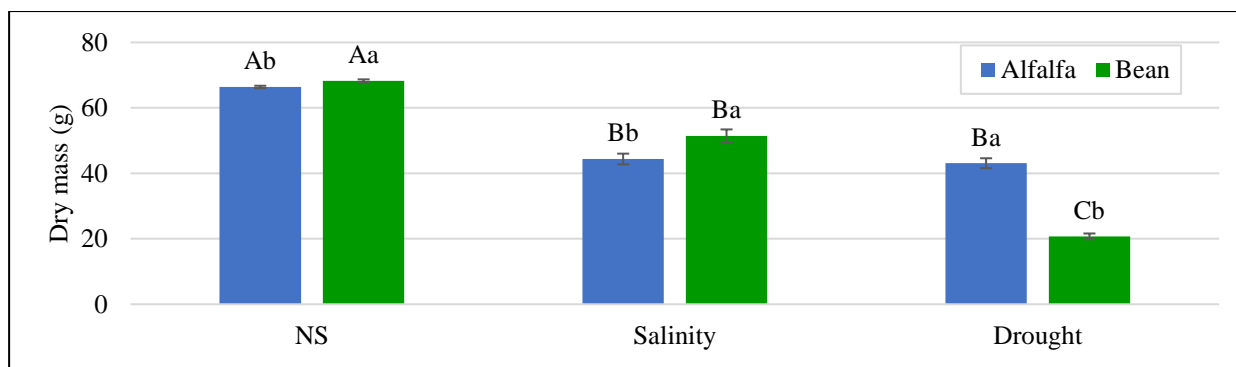


Figure 36. The dry mass of above ground structure of alfalfa and bean at different stress treatments (2024, MATE-Gödöllő).

Capital letters compare the effect of the same CC treatment under different stress treatments, while lowercase letters compare the effect of different CC treatments under the same stress treatment on CC dry mass. Different letters indicate significant difference at $p < 0.05$.

4.4.5 Discussion

Overall, our results discovered that the drought stress (DS) were more detrimental to the growth of maize than the salt stress (SS) in both monocrop and intercropped systems. DS has been reported in various publications to have more detrimental effects on maize than SS, impacting growth, development, and yield significantly. Our results supported the study of 36 maize varieties, which also discovered that DS affects maize height, SPAD value, transpiration rate, and yield more severely than SS (ZHANG et al. 2019). WEI, FAN, et al. (2022) revealed DS resulted in more severe wilting and yellowing in maize compared to SS, indicating a more significant detrimental effect on plant growth. It was discovered that DS caused more photosynthetic pigment damage than SS. Meanwhile, research indicates that DS significantly affects maize development compared to SS performance, including maize LAI, biomass, and the cob weight on five maize hybrids on various maize hybrids (KÓMLOSI et al. 2022). Our result also found that DS caused stunted growth in the maize plant compared to the SS. Similar results was published in another study which showed that DS during the vegetative stage influences the overall plant development in maize (ÇAKIR 2004). It was stated that water scarcity which caused a reduction in leaf water potential and stomatal aperture, consequently causing the down-regulation of genes associated with photosynthesis and reduced the CO_2 uptake. The reduction in photosynthesis rate eventually reduces the growth rate in plants (OSAKABE et al. 2014).

Meanwhile, the reproductive results also showed that DS and SS increase maize ASI index. Based on our result, both kind of stress significantly delayed the emergence of both male and female inflorescence in maize. However, drought stress severely affects the emergence of silk, causing a higher ASI value, which is highly influenced by water and nutrients available to plants. A large ASI value indicates unsynchronized anther extrusion and silk exposure that prevents

successful pollination from occurring, which reduces the final yield formation (ZHUANG et al. 2024). A study found that, under water-limiting conditions, the silk growth rate decreased and terminated two to three days after the first silk exposure (OURY et al. 2016). Besides that, as discussed in the literature review, drought conditions, especially at the reproductive stage, could also lead to pollen sterility, pollen tube deformation, flower drop, and ovule abortion, which also affects the final yield formation. At high temperatures, maize yield is reduced due to disruption of carbohydrate metabolism and starch biosynthesis needed for both male and female reproductive organs (SINHA et al. 2021). Nevertheless, our results found that incorporation of alfalfa and mung bean significantly reduced the ASI values in both salt and drought stress.

Besides that, in spite of SS being less damaging than DS in this experiment, the stress imposed by the presence of salt significantly reduced maize vegetative and reproductive growth. Even though the irrigation rate was similar in NS and SS, the SS caused higher moisture absorption by the plant, thus significantly reducing the soil moisture. Our results supported several similar findings published in the recent years. Other than a 47% reduction in plant height and 44% in leaves area, it was discovered that SS reduces stomatal conductance and transpiration rates, which increase the canopy temperatures up to 4°C (VENNAM et al. 2024). SS significantly reduces maize growth and reproduction by decreasing photosynthesis due to damage to photosynthetic pigment. The decline in assimilate translocation also caused poor kernel setting, which eventually affected the final grain number and weight (IQBAL et al. 2021). SS conditions also reduce spikelet growth, silk growth, and kernel setting in maize. It was discovered that the inability to utilize carbohydrate reserve during osmotic stress also caused kernel abortion in maize (HENRY et al. 2015). It was discovered that under SS conditions, plant increase their water uptake capacity as a survival mechanism (WIN et al. 2011).

Based on our germination test, the Margitta maize variety tolerated the 100 mM NaCl stress imposed on the seeds in (Section 4.2). Meanwhile, in this experiment, the variety was also able to develop and reproduce in the same salt concentration, especially with the assistance of leguminous cover crops. The presence of leguminous cover crops improved the performance of maize in both SS and DS compared to the uncovered soil surface. In one study, alfalfa proved the ability to reduce the salt content in the soil while increasing the nutrient content due to an increase in the soil microbial community (MEI et al. 2022). A study revealed that intercropping cowpea and maize, together with the application of N fertilizer, increases maize productivity in saline soil (EL-GHOBHASY et al. 2020). In SS conditions, elevation in proline, Na⁺, peroxidase while decreasing superoxide dismutase increases the alfalfa tolerance towards the stress (HOU et al. 2022). Alfalfa is also the best choice to plant in drought-prone areas as it improved maize performance and

produced significantly the highest biomass at the end of the experiment. Alfalfa is considered drought tolerant but not resistant. It was discovered that under stress conditions, alfalfa changes its root morphology by increasing the branching, which enhances water absorption from the soil (LI et al. 2022). Another study discovered that alfalfa increases the root mass and length to increase water absorption more rapidly only during the drought recovery period (ERICE et al. 2010).

Additionally, mung bean gave significantly similar benefits to maize crops like alfalfa except in drought conditions where alfalfa was superior. In SS condition, intercropping maize and mung bean improved the maize vegetative and reproductive growth at a similar level to alfalfa. Our results supported the findings of PATACZEK et al. (2018) which mentioned that mung bean is more tolerance to DS compared to SS, with the highest sensitivity in the germination stage. Nevertheless, salt tolerance varieties were widely cultivated in saline areas in Myanmar and were able to tolerate salt concentrations up to 225 mM NaCl (WIN et al. 2011). It was discovered that inoculation of mung bean seeds with salt tolerance rhizobium bacteria will improve their salt tolerance (AHMAD et al. 2013). Bacterial inoculation also improves the PGPR community, which significantly enhances soil fertility by aiding in soil aggregation, refining nutrient acquisition processes, and encouraging root proliferation, thus improving growth in stressful soil conditions (PATACZEK et al. 2018). Mung bean as pre-crop improved wheat yield up to 0.45 tonne per hectare, and incorporation of mung bean residue into the soil will provide N equivalent to the application of 74 - 94 kg of urea per hectare (SHARMA et al. 1995). In our study, mung bean was also able to reduce soil temperature in both DS and SS. Even though mung beans utilised more moisture in saline soil conditions, the moisture loss did not affect the vegetative and reproductive growth of maize intercropped with them. A study on cotton and mung bean revealed a higher water and nitrogen use efficiency in intercropped cotton than in monocrop cotton, which resulted in higher cotton yield (LIANG and SHI 2021). Finally, mung bean also produced heavier dry biomass at the end of the experiment compared to alfalfa in both NS and SS conditions. The fact that the mung bean plant is phenotypically bigger with a thicker stem and larger canopy than alfalfa may influence the end dry mass of this species. Besides the potential seeds harvested, the higher residue volume shows the multipurpose properties of this bean species.

In this experiment, the benefits of soil surface covering CC in retaining soil moisture and temperature were proven by alfalfa and white mustard in NS and SS conditions. However, several field studies reported that in a long-term monocrop system, alfalfa reduced soil water content due to its deep rooting system (SUN, HUANG, et al. 2018; WANG et al. 2023). However, a study in China's Corn Belt reported that the presence of alfalfa intercropped with maize significantly increases the relative soil water content in one of the treatments and improves the biomass water

use efficiency (SUN, LI, et al. 2018). Meanwhile, in this experiment, white mustard had a lower survival rate in dry conditions, causing soil surface exposure, which increase the temperature and increase moisture evaporation in drought conditions. It was mentioned that mustard cover crops require adequate soil moisture and can deplete soil moisture, especially in dry conditions, but can enhance subsequent crop performance (DONG et al. 2023). Furthermore, intercropping maize with white mustard revealed a competitive effect on the maize, leading to a reduction in both vegetative growth and reproductive development. It can be concluded that white mustard competed for water and nutrients with the main cash crop, maize, compared to the leguminous cover crop. A similar observation was recorded in the field trial with the white mustard intercropped with maize and will be discussed further in section 4.6.

The three cover crops tested showed the ability to lower soil temperature only if they were fully established and the above ground biomass provide effective soil surface covering. Surface coverage by cover crop protected the soil surface by shading the soil surface from direct solar exposure, alter canopy heat balance, lower thermal conductivity, and reducing soil water evaporation in contrast to exposed soil (YANG et al. 2021). BLANCO-CANQUI and RUIS (2020) found that cover crop reduced average springtime soil temperature about 1°C (BLANCO-CANQUI and RUIS 2020). While temperature provides important role in increasing root biomass and nutrient uptake, temperature above the optimum levels could negatively affecting the effectiveness of soil nutrients uptake including nitrogen and soil moisture conservation (XIA et al. 2024). Besides that, soil temperature also influences the movement of soil solution and the form of soil water. At high soil temperature condition, the soil water movement and gaseous form of water increase in frequency, while available solid water for crop roots decreases. Besides that, elevated root zone temperature causes elevated active oxygen build-up in the roots which caused membrane lipid peroxidation and enzyme inactivation. The chain of reactions negatively impacts the root absorption and synthesis function which eventually affecting the plant development (XIA et al. 2021). Furthermore, high soil temperature also disrupts soil organic matter decomposition while inhibits various enzyme and microorganism activities beneficial to plant growth (ROBINSON et al. 2020).

4.5 EXPERIMENTAL RESEARCH 5: Integration of different leguminous cover crops on maize cultivation under various nitrogen levels in 2023

4.5.1 Maize vegetative stage

Photo 2.1, 2.2, 2.4 and 2.5 in A2 Appendices section show the vegetative growth of both maize and their companion cover crops. The MANOVA results in Table A.24 in the Appendices

section show, the type of cover crops (CC), different N treatment and their interaction revealed a significant effect on the dependent variables with a small effect size. Thus, follow-up ANOVA was carried out and the results are presented in Table A.25 in the Appendices section. The ANOVA shows that, individually, N treatments and type of CC had a significant effect on maize plant height, leaves area and SPAD value with $p < 0.05$. Based on the partial η^2 , N had bigger size effects on height, SPAD value and leaves area compared to CC treatment. The CC treatment also had the biggest effect on SPAD compared to the other two parameters. However, the interaction between these two factors only had a significant effect on maize plant height and the SPAD value but not leaves area.

Figure 37 shows maize height was severely affected at 0 kg/ha N but gradually increased as the fertiliser rate increased. Maize intercropped with red clover (MRC) showed a significant height improvement at 0 and 50 kg/ha N compared to the plot integrated with alfalfa (MA) and crimson clover (MCR). The control plot, which contains only monocrop maize (MM), has a significantly lower height at 0 and 50 kg/ha N than the CC plots. However, the MM managed to achieve a relatively similar height with MRC and MCR and produced a higher plant than the MA plot at 100 kg/ha N. As the fertilizer rate increased to 150 kg/ha, only MA and MRC significantly increased in height while the MM and MCR showed no significant height increase at 150-200 kg/ha N. Lastly, all intercropped and MM show no significant difference height at 200 kg/ha N than at 150 kg/ha N.

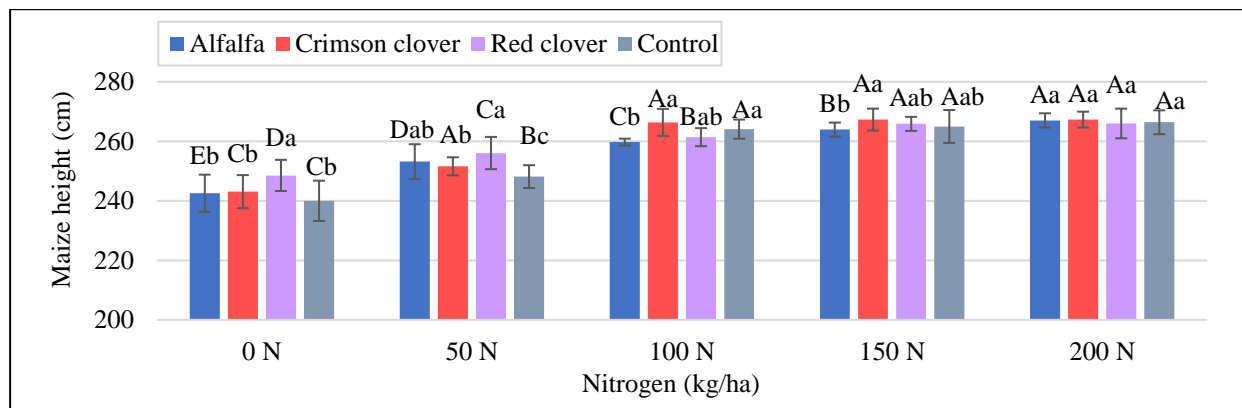


Figure 37. Average plant height at VT of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rates on plant height. Different letters indicate significant difference at $p < 0.05$.

Furthermore, Figure 38 shows that the application of crimson clover and red clover significantly increased maize leaves area, especially at a higher N rate. At a 0 N rate, alfalfa incorporation reduced maize leaves area compared to MM, while no significant difference in area was observed between these two treatments as the N concentration increased. Besides that,

application of N above 100 kg/ha did not improve the MM leaves area compared to the intercropped maize. Meanwhile, maize integrated with CC reached the largest leaves area at 150 kg/ha N.

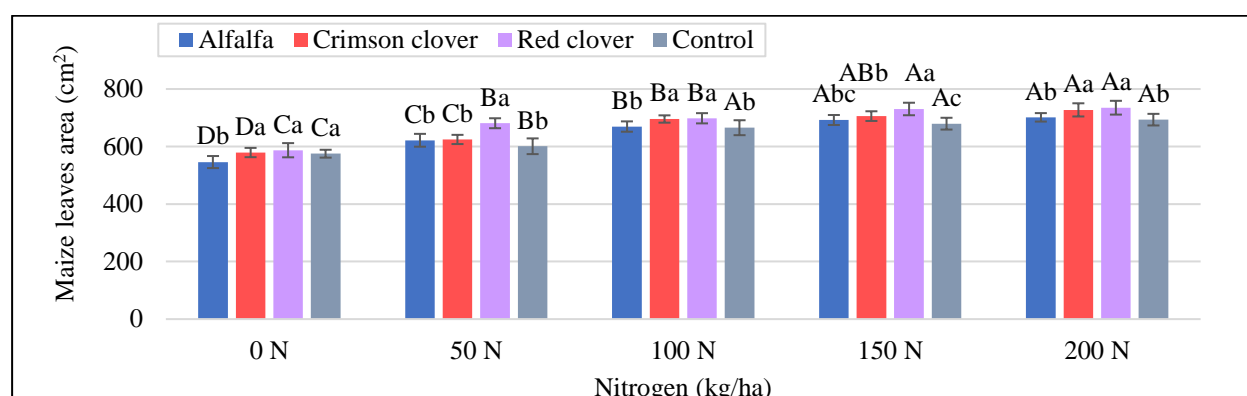


Figure 38. Average leaves area at VT of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rates on leaves area. Different letters indicate significant difference at $p < 0.05$.

The ANOVA result in Table A.26 in the Appendices section shows, the N levels, type of CC and their interaction also significantly influenced maize leaves area index (LAI) with large effect size. Figure 39 summarises that the MRC produced significantly higher LAI at all N rates compared to MM. Meanwhile, MCR produced the highest LAI at 0 kg/ha N compared to all three treatments. In contrast, MA produced a significantly lower LAI compared to MM until it reached a significantly equivalent value at a higher N rate of 150 kg/ha and 200 kg/ha. The data also show that the incorporation of red clover or crimson clover could improve the stagnant LAI value after 100 kg/ha N in MM.

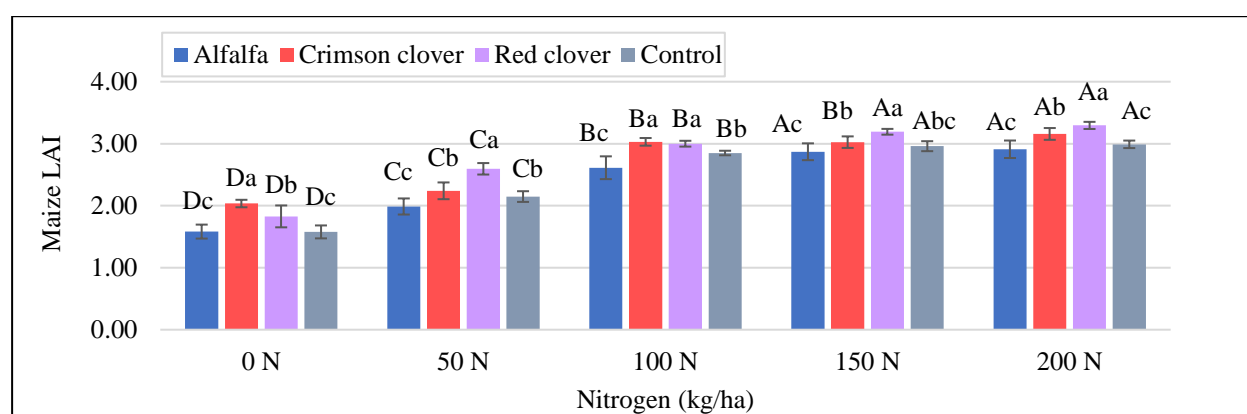


Figure 39. Average leaves area index (LAI) at VT of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rates on LAI. Different letters indicate significant difference at $p < 0.05$.

Moreover, the integration of red clover caused a significant improvement in the SPAD values of maize plants compared to the MM and the other two CCs. Alfalfa significantly improved maize SPAD value at low N levels but did not increase value more than MM at N between 100 – 200 kg/ha. Nevertheless, crimson clover also improved maize SPAD value compared to the MM at almost all N rates (Figure 40).

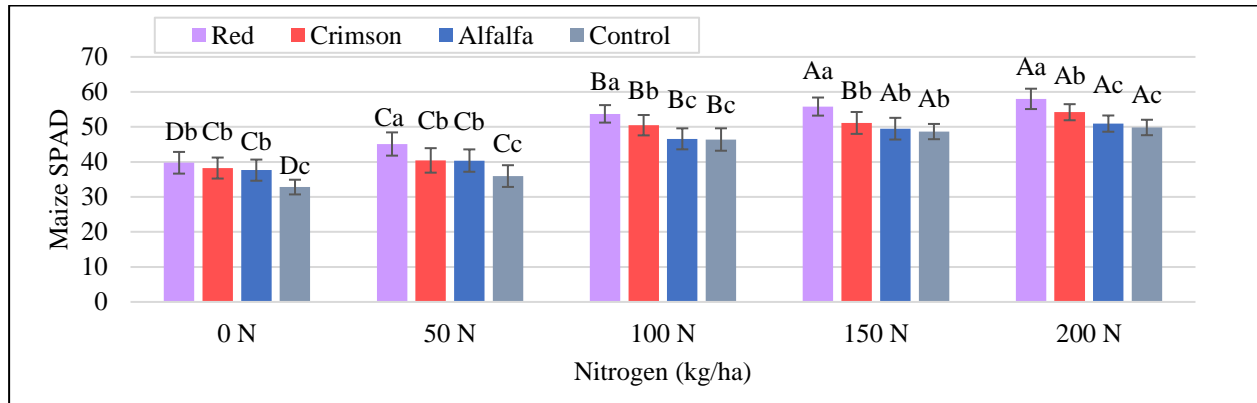


Figure 40. SPAD values at VT of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rates on SPAD value. Different letters indicate significant difference at $p < 0.05$.

4.5.2 Maize harvested yield

Meanwhile, photo 2.6 in A2 Appendices section show maize matured ear in MA intercropping system. The MANOVA results in Table A.27 in the Appendices section reveal, the type of cover crops (CC), different N treatment and their interaction show a significant effect on yield quantity parameters. Only N level had a large effect on the parameters, while CC and the interaction of factors had a medium effect on the dependent variables. Thus, follow-up ANOVA was carried out and presented in Table A.28 in the Appendices section. The ANOVA shows, individually, N treatments and CC had significant effects on maize ear weight (EW), ear length (EL), kernel weight (KW) and grain:cob ratio (G:C) with $p < 0.001$. However, the interaction between the factors only significantly affected the EW and KW. Based on the partial η^2 , N caused bigger effects on the yield quantity compared to CC treatment.

Furthermore, Figure 41 shows that maize EW significantly increased as the N rate increased from 0 to 200 kg/ha in all the treatments. Besides that, the intercropped maize produced heavier ears at low N levels of 0 and 50 kg/ha N compared to the MM. As the N rate increased to 100 kg/ha, MM produced a significantly heavier ear compared to MA and MCR. MM also produced no significant difference in weight with the intercropped maize at 150 kg/ha N. However, while the ear weight of MM showed no significant increase as the N rate increased from 100 to

200 kg/ha, the ear weights of MCR and MRC continued to increase with the increment in N levels. Furthermore, the integration of red clover with the application of 200 kg/ha of N produced the highest maize ear weight, averaging 246.03g.

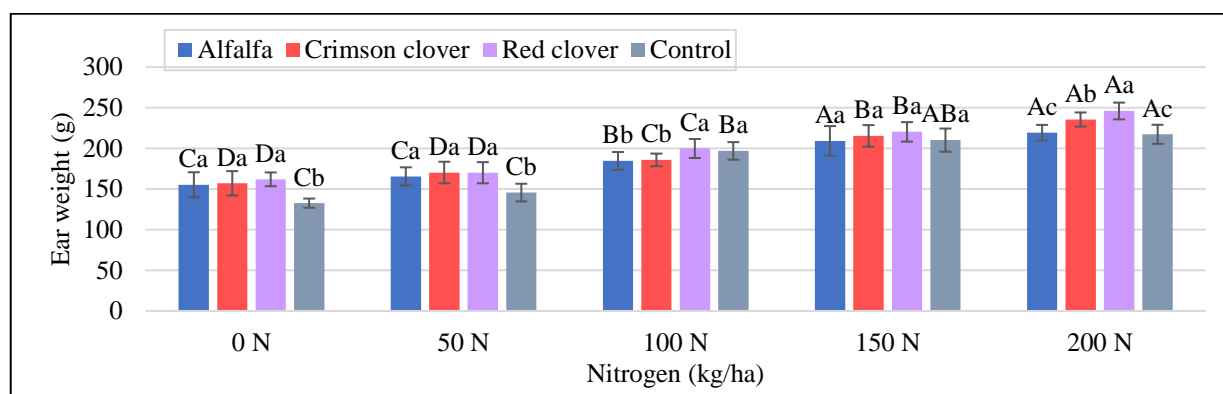


Figure 41. The average ear weight of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rates on EW. Different letters indicate significant difference at $p < 0.05$.

Besides that, the maize EL also shows a gradual increasing pattern as the N rate increases, as shown in Figure 42. Similar to the EW, the intercropped maize produced longer ears at low N levels of 0 and 50 kg/ha N compared to the MM. The MRC produced significant longer ear compared to MM at all N levels. Meanwhile MCR also produced significantly longer ear at all N levels except at 100 kg/ha N where no significant different was observed. Furthermore, there was no significant difference in EL between MM and MA at 100 and 150 kg/ha of N. However, at 200 kg/ha of N, MA had a significantly higher EL than MM, which produced the lowest EL among all intercropped maize. Lastly, all treatments reached the maximum EL at 150 kg/ha, as no significant increases were observed at 200 kg/ha.

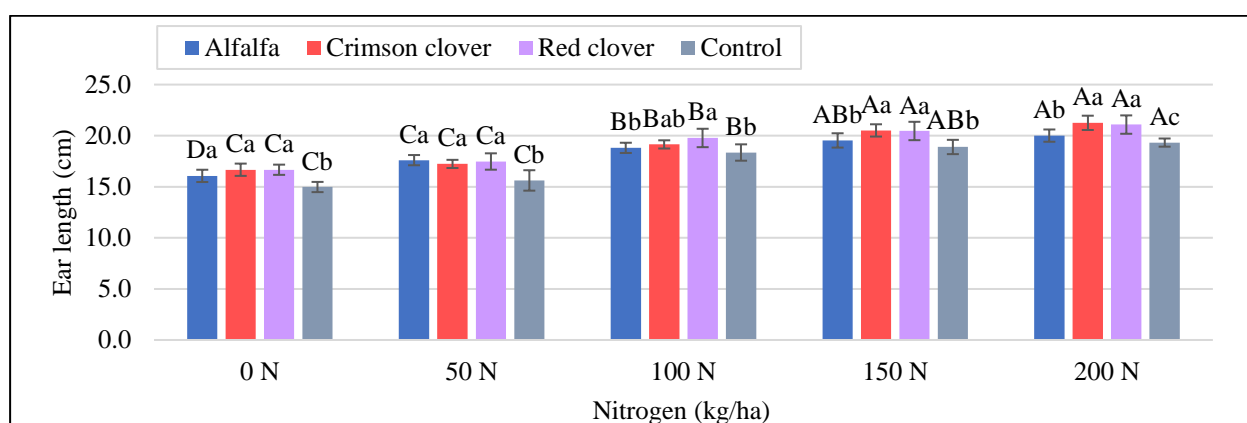


Figure 42. The average ear length of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rates on EL. Different letters indicate significant difference at $p < 0.05$.

Furthermore, Figure 43 and Figure 44 show that the N treatments also significantly increased the KW per ear and G:C ratio of maize regardless of the CCs treatment. At 0 kg/ha N, intercropped maize produced a higher value in both G:C and KW, while there was no significant difference in all CCs treatment at 50 kg/ha N. As the N rate increased to 100 kg/ha, MRC produced the highest G:C ratio, while MM produced relatively similar KW to the intercropped maize. MCR produced the highest G:C percentage increase at 200 kg/ha with a 15.8 % increase, while MRC produced the highest KW increment rate of 14.6 % at 150 kg/ha N compared to MM. MA produced significantly higher G:C value than MM at 150 and 200 kg/ha N but no significant difference was recorded in the KW at these two N levels. Red clover and crimson clover proved to increase the KW and G:C in the absence of N and at N rates higher than 100 kg/ha.

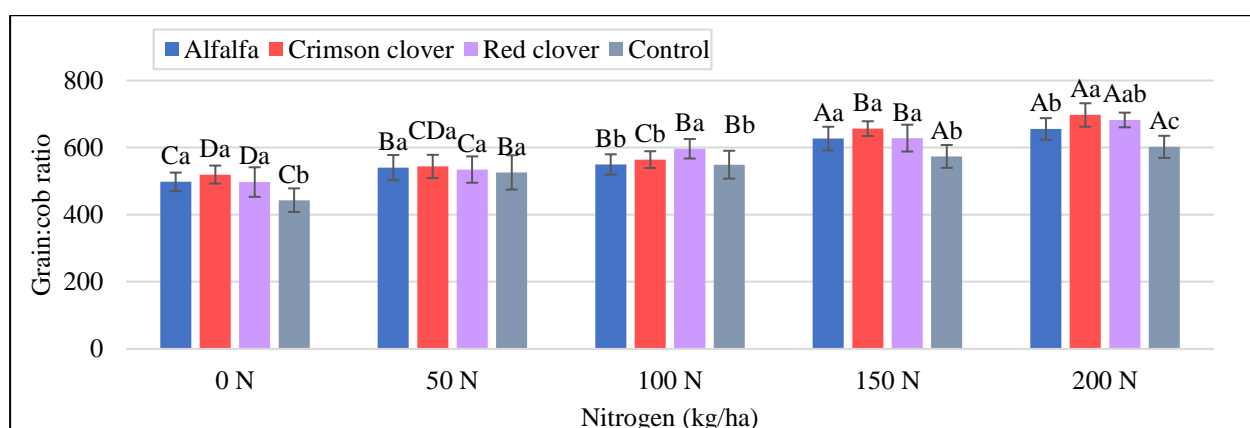


Figure 43. The average G:C ratio of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rates on G:C ratio. Different letters indicate significant difference at $p < 0.05$.

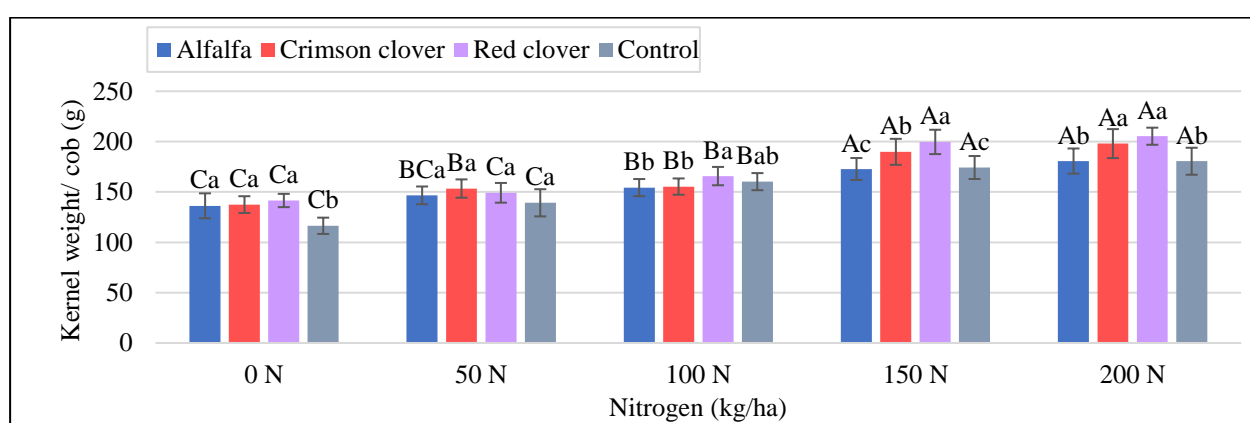


Figure 44. The average kernel weight per ear of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rates on KW. Different letters indicate significant difference at $p < 0.05$.

Furthermore, a univariate ANOVA was performed and presented in Table A.29 in the Appendices section. The ANOVA reveals the two factors and their interaction also significantly affected the thousand kernel weight (TKW) of maize with $p < 0.001$ with a large effect size. Based on Figure 45, increasing the N rate from 0 to 50 kg/ha did not improve the TKW in MM, MA, and MRC. However, all intercropped maize produced statistically similar TKW at 100 kg/ha of N. Despite producing the lowest TKW at 0 and 50 kg/ha N, MM produced the highest TKW at 100 kg/ha and higher value than MA and MCR at 150 and 200 kg/ha N. Meanwhile, red clover increased maize TKW as the N rate increased but only up to 150 kg/ha with a maximum percentage increase of 4.61%. Even though the application of 200 kg/ha N significantly reduced the TKW in MRC, the value was still significantly higher than the other two intercropped systems. Besides that, MCR produced significantly lowest TKW at 0 kg/ha N but showed significant increase with the presence of N and reached the highest level at 150 kg/ha N. Lastly, MA only improved the TKW at low N levels, while at higher N levels between 100 to 200 kg/ha, MA produced the lowest TKW compared to MM and the other intercropped maize.

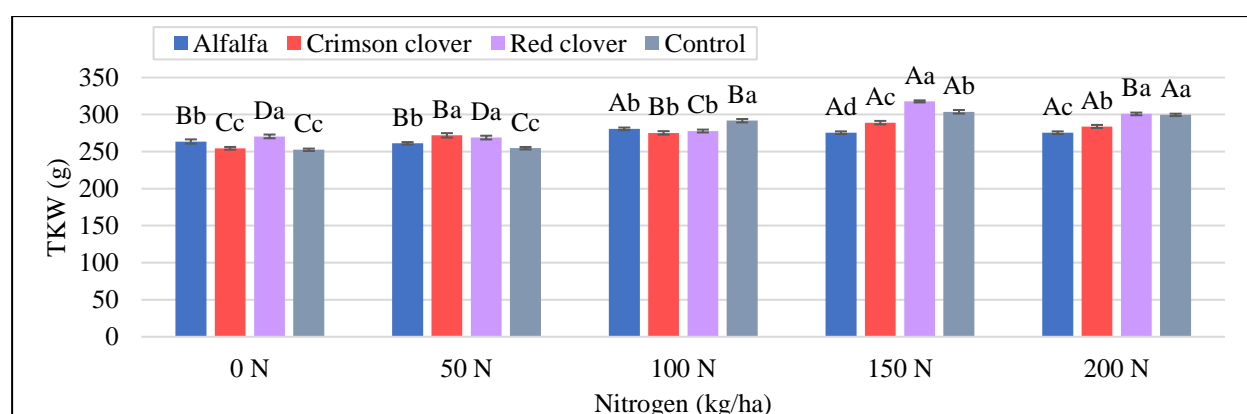


Figure 45. Thousand kernel weight (TKW) average of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rates on TKW. Different letters indicate significant difference at $p < 0.05$.

4.5.3 Grain chemical composition

The MANOVA results in Table A.30 in the Appendices section show that the type of cover crops (CC), different N treatment and their interaction had a significant large effect on yield chemical composition parameters with $p < 0.001$. The follow-up ANOVA results in Table A.31 in the Appendices section show both factors and their interaction significantly affecting the maize moisture, oil, protein and starch content at $p < 0.05$. Based on the partial η^2 , both factors and their interaction had a large effect on all the chemical composition parameters tested except the moisture content.

Table 11. Mean±standard deviation of maize moisture content (%) in different nitrogen and cover crop treatments (2023, MATE-Gödöllő).

N level (kg/ha)	Maize seeds moisture content (%)			
	Alfalfa	Crimson clover	Red clover	Control
0	11.12±0.12Ca	11.05±0.1Ca	10.98±0.16Ba	11.07±0.12Ca
50	11.07±0.08Ca	11.05±0.16Ca	11.17±0.15Ba	11.23±0.16Ca
100	11.43±0.12Ba	11.27±0.08Bab	11.13±0.1Bb	11.48±0.09Ba
150	11.53±0.1Bab	11.55±0.08Aa	11.65±0.1Aa	11.4±0.12BCb
200	11.78±0.12Aa	11.42±0.19ABa	11.77±0.1Aa	11.7±0.09Aa

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rates on moisture content. Different letters indicate significant difference at $p<0.05$.

Table 11 reveals that the moisture content increased as the N rate increased. Besides that, all CC treatments produced the highest moisture content at 200 kg/ha N except for MCR, which produced the highest moisture at 150 kg/ha N. At 0 and 50 N kg/ha, there were no significant differences in moisture content between all treatments. Meanwhile, at 200 kg/ha N, no significant difference in moisture content on all the intercropped maize and the control.

The oil content analysis illustrated in Figure 46 shows that alfalfa integration severely affected the maize oil content at 0 kg/ha N and 200 kg/ha N compared to other CC treatments and the MM. Meanwhile, red clover significantly increased maize oil content at all N levels, while crimson clovers only significantly increased oil content in maize at 100 to 200 kg/ha N. The oil content in MA was significantly higher than MM at 50 kg/ha N, but no significant difference was observed at 100 and 150 kg/ha. Increasing the N level by more than 50 kg/ha also did not improve the oil content in MA, but the MM, MRC and MCR continuously showed elevation in oil content as the N increased to 200 kg/ha.

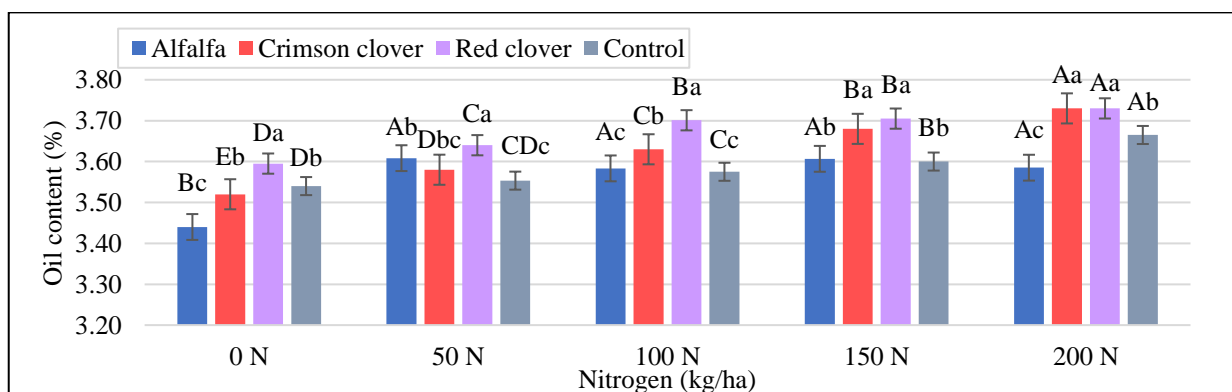


Figure 46. Oil content average in seeds of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rates on oil content. Different letters indicate significant difference at $p<0.05$.

However, the NIR analysis revealed that MA produced significantly higher protein content than the MM treatment at all N levels except 0 kg/ha (Figure 47). Meanwhile, the MCR only

showed a significant protein content difference from MM as the N level increased to 100 - 200 kg/ha. MRC also generated significantly higher protein content than MM at all N levels, and the gap was increased as the N level increased with a 26.9% increment at 200 kg/ha N compared to MM. Besides that, the MRC required 100 kg/ha N, while the MM required 200 kg/ha N to increase the protein content significantly compared to 0 kg/ha N.

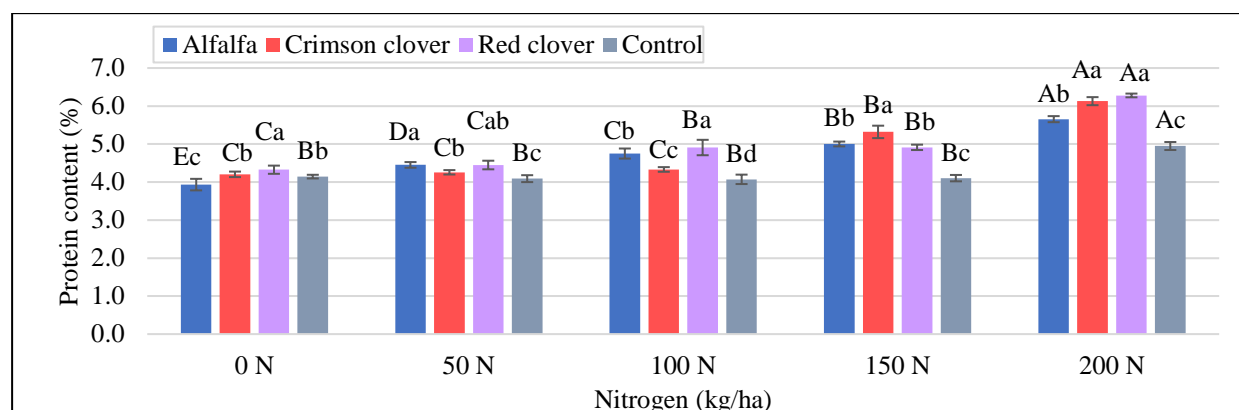


Figure 47. Protein content average in seeds of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rates on protein content. Different letters indicate significant difference at $p < 0.05$.

Moreover, intercropped maize also produced significantly higher starch content at low N levels than the MM (Figure 48). However, alfalfa did not improve the starch content at a higher N rate of 100 to 200 kg/ha. MCR produced the highest starch content at low N levels compared to other treatments, but the increase was stagnant from 50 to 150 kg/ha before significantly increased at 200 kg/ha N. Meanwhile, MRC showed a steadily increasing pattern in starch content as N levels increased and produced the highest percentage at 100 to 200 kg/ha N. Nevertheless, the increment of N levels significantly increased the starch content in all treatments, but higher levels could be achieved with the incorporation of red clover or crimson clover between the maize rows.

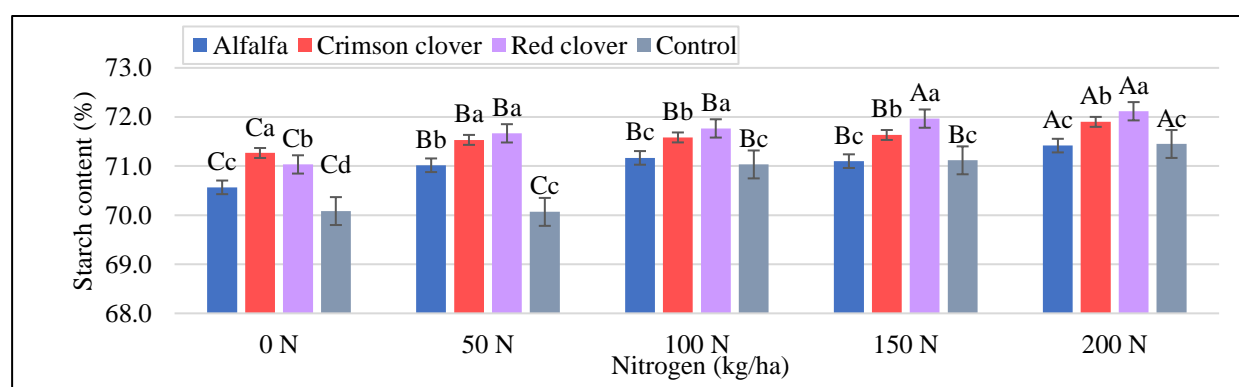


Figure 48. Starch content average in seeds of maize intercropped with different cover crops at various nitrogen levels (2023, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rates on starch content. Different letters indicate significant difference at $p < 0.05$.

4.5.4 Cover crop dry mass

Finally, the ANOVA reveals that the N treatment had a significant effect on the dry mass of the cover crops cultivated between the maize rows with alfalfa $F(4,10) = 2194.7$, $p < 0.001$ partial $\eta^2 = 0.999$, crimson clover $F(4,10) = 4841.54$, $p < 0.001$ partial $\eta^2 = 0.999$ and red clover $F(4,10) = 265.062$, $p < 0.001$ partial $\eta^2 = 0.991$. Table 12 shows that all cover crops achieved the highest dry mass at 200 kg/ha N, with red clover producing the highest value compared to alfalfa and crimson clover. Besides that, red clover accumulated significantly different dry mass at each N level, while alfalfa did not increase the dry mass as the N level increased from 100 to 200 kg/ha. Meanwhile, crimson clover also produced a significantly different dry mass as the N level increased up to 150 kg/ha and no significant increase at 200 kg/ha N.

Table 12. Mean \pm standard deviation of cover crop dry mass at different nitrogen levels harvested within 0.2 m² area between the maize rows (2023, MATE-Gödöllő).

Kg/ha	Alfalfa (g)	Crimson clover (g)	Red clover (g)
0 N	44.5 \pm 1.60E	57.0 \pm 0.80E	71.3 \pm 0.55D
50 N	58.2 \pm 0.85D	66.2 \pm 0.25D	75.2 \pm 1.45D
100 N	92.6 \pm 0.85C	86.1 \pm 0.25C	86.5 \pm 2.60C
150 N	97.6 \pm 0.40B	95.7 \pm 0.20B	98.1 \pm 1.31B
200 N	100.2 \pm 0.53A	97.5 \pm 0.45A	110.9 \pm 2.06A

4.5.5 Discussion

Based on our results, intercropping maize with leguminous cover crops could improve maize performance with the right choice of cover crop species. The results also show that at lower N levels, intercropping maize with alfalfa, red clover or crimson clover generated higher generative growth and yield quantity compared to single maize. Meanwhile, the incorporation of red clover and crimson clover produced a positive effect on maize plants at higher N levels, while alfalfa showed no significant difference in maize growth and yield compared to the single maize system. Our results supported many studies on maize and leguminous cover crop intercropping. Various studies reported that intercropping maize with other leguminous species, including soybean (FAN et al. 2020), peanut (ZHAO, DONG, et al. 2022), cowpeas (LATATI et al. 2014) and faba bean (ZHANG, CHEN, et al. 2012), can also promote the growth and yield of the adjacent maize plant. FU et al. (2023) published that the TKW of maize increased between 4.8 – 7.5% and 2.1 – 10.4% when intercropped with soybean and peanut, respectively, at 0 kg/ha N. Our results found that incorporation of red clover increased up to 7.3% of maize TKW at 0 kg/ha N and up to 4.6% at 200 kg/ha N.

Based on our results, it was visible that the improvement of maize plants by the leguminous cover crops began at the vegetative growth stage, which influenced the yield quantity and quality

harvested at the end of the season. The SPAD value proved that the incorporation of leguminous cover crops could increase the chlorophyll content in the leaves, which increases the photosynthetic rate in the plant. The impact of higher photosynthetic rate was visible in the plant height, leaves area, amount of yield harvested and the grain's chemical composition. The increase in leaf area and leaf area index (LAI) strongly affected the photosynthesis rate and grain filling. Larger maize leaves are able to utilise solar energy more effectively and increase the photosynthate, which affects the grain number and weight (OCHIENG' et al. 2021). A study found that intercropping peanuts with maize improved the light condition on maize ear leaves, which improved sunlight exposure and photosynthate distribution in intercropped maize compared to monocrop maize (LI et al. 2019a)

In intercropping systems, the rhizosphere interaction between the crops is one of the success factors in the system. In leguminous intercropping system, rhizosphere modification can improve enzymatic activities, root exudation, and soil pH, while facilitative root interactions allow nutrient exchange such as fixed N between the species (LI et al. 2019a; NASAR et al. 2023). Studies reported that intercropped maize with leguminous species like faba bean (LI et al. 2006) and alfalfa (ZHANG et al. 2013) developed higher-density roots which penetrated deeper soil profile compared to monocrop maize. In maize-alfalfa intercropping, maize root mass is also distributed in the shallow soil profile and interacts with the neighbouring alfalfa rhizosphere (ZHANG et al. 2013). The root structure adjustment traps the nutrient from leaching and improves nutrient absorption from deeper soil layers, hence directly linked with the increase of photosynthetic activities in intercropped maize leaves compared to monocrop maize (LI et al. 2006).

Besides that, it was discovered that the interspecific competition for N increased in maize-peanut and maize-soybean intercropping. In both systems, it was discovered that maize absorbs more nitrogen (N), phosphorus (P) and potassium (K) than their leguminous companion (FAN et al. 2020; ZHAO, DONG, et al. 2022). It was reported that rapid nutrient absorption by maize root system caused an N deficit in soybean root zone, which increased the nitrogen fixation activity by the leguminous root system. FAN et al. (2020) also highlighted the importance of narrow spacing between the crops to increase the interspecific competition that leads to the N level elevation in soil and higher N uptake in the intercropping system. In conclusion, the root structure adjustment and the rapid N uptake by maize allow the N fertilizer to be applied and fully utilised by the system, mainly by maize crops, which improves the growth and yield performance.

Meanwhile, the soil surface covered by the cover crops also reduced the solar radiation exposure on the soil between the maize rows. Reduced solar radiation exposure will reduce the

soil temperature and moisture lost, which increases the N and P solubilisation and traps them in the rhizosphere. Therefore, higher N and P are available for intercropped maize compared to monocrop maize, hence increasing the whole plant performance (GITARI et al. 2018). Besides that, the soil coverage also improves the population of microorganisms in the rhizosphere, which differentiates the ecosystem in intercropped systems from monocrop systems. ZHAO, DONG, et al. (2022) mentioned that the interaction between maize and peanut increases the bacterial biodiversity with high density in beneficial bacteria, which promotes nitrogen fixation, dehydrogenation and soil nitrogen balance in the rhizosphere. Meanwhile, the rhizobium species in *Arachis hypogaea* L. and *Stylosanthes guianensis* increase the phosphorus, dissolved organic carbon, ammonium and the total nitrogen available for maize to uptake (XIANG et al. 2018).

Furthermore, our results also discovered that increasing N more than 100 kg/ha may not be beneficial for monocrop maize, but in MCR and MRC, significant improvement was visible in the growth and yield performance as the N level increased up to 200 kg/ha. In terms of yield performance, 150 kg/ha may be the optimum rate as increasing up to 200 kg /ha N did not improve the kernel weight and TKW in MCR while significantly decreasing the TKW in MRC. Studies in Hungary also showed that the optimum rate of N fertilizer is around 120 to 180 kg/ha for single maize cultivation, depending on the weather conditions (SZÉLES et al. 2018, 2023). It was discovered that the application of N fertilizer in maize/legumes intercropping system could also improve maize yield by improving leaf chlorophyll content, photosynthetic activities and photosynthetic nitrogen use efficiency (PNUE) compared to mono-cropped maize (NASAR et al. 2021). In the maize/soybean system, the application of 250 kg/ha N increased total N uptake by up to 75% and grain N content by up to 31% (NASAR et al. 2023).

Moreover, a study also found that N fertilizer application increased the nodulation in alfalfa and red clover. Therefore, the application of N fertilizer did not inhibit the nitrogen fixation activities in the leguminous species crop. It was also mentioned that high N application improves the leguminous crops establishment in areas lacking of native rhizobial symbionts (FORRESTER and ASHMAN 2018). Our results also found the positive impact of N on the cover crop dry mass. Besides that, in our study, N application at the maize V6 stage may allow adequate time for the cover crops to establish and form their rhizobium nodules before the presence of chemical N input. An example of successful nodulation formation in crimson clover can be referred to Photo 2.3 in A2 Appendices section. The established cover crops also provide coverage for the fertilizer granules from total sun exposure and reduce evaporation.

In addition, intercropping maize with leguminous cover crops was also found to increase the oil, protein and starch content up to 3.1%, 26.87% and 2.3%, respectively. The starch content

gradually increases with the N rate up to 200 kg/ha for all treatments except for MRC, which showed no significant difference at 150 and 200 kg/ha N. The increase in starch is closely linked with the increase in SPAD and LAI, which increase the photosynthetic rate and photosynthate transfer and are eventually stored in the grain as starch molecules (LI et al. 2019a; NASAR et al. 2021). Meanwhile, a study on silage maize-field bean intercropping found an elevation in protein content up to 21.91% in intercropped maize compared to monocrop maize (SOWINSKI 2024). Improvement in the N and P uptake in the intercropping system is supposed to increase the protein synthesis in maize and increase the content in the grain (LATATI et al. 2016). Meanwhile, the increase in grain moisture content may be due to the bigger seeds that require a longer period to dry than the smaller seeds.

Various studies showed an increment in maize yield when intercropped with alfalfa (ZHANG et al. 2013; SUN, LI, et al. 2018; NASAR et al. 2020). Our study discovered that this is only true at low levels of N but not at higher N levels more than 50 kg/ha N. However, our results found that alfalfa still increased maize protein, but the other growth and yield parameters showed no significant difference from the monocrop maize. It is important to highlight that the cover crop's beneficial effects on maize performances are also highly influenced by various factors such as climatic conditions, soil type, agrotechnical like sowing dates and fertilizer application time, which are different in every publication. Therefore, based on this study, alfalfa may not be the best choice in terms of economic aspects to be incorporated in maize as it increases cost but gives a similar yield volume as the single maize system. Nevertheless, alfalfa was tested again in 2024, and the results will be discussed in section 4.6. Finally, based on our result in 2023, red clover was the best choice to be intercropped with grain maize, followed by crimson clover.

4.6 EXPERIMENTAL RESEARCH 6: Integration of leguminous and Brassicaceae cover crops on maize cultivation under various nitrogen level in 2024

4.6.1 Maize vegetative stage

The MANOVA results in Table A.32 in the Appendices section, reveal the type of cover crops (CC), different N treatment and their interaction showed a significant effect on the vegetative parameters with large effects at $p < 0.001$. Follow-up ANOVA results in Table A.33 in the Appendices section show that N treatments, type of CCs, and their interaction significantly affected maize plant height, leaves area and SPAD value with $p < 0.01$. Based on the partial η^2 , all

factors and their interaction had large effects on maize height and SPAD value but medium effect size on maize leaves area.

Meanwhile, Figure 49 shows that maize intercropped with white mustard (MWM) produced the shortest plant compared to monocropped maize (MM) and other intercropped systems at all N levels. MM produced higher plants than the maize-legume intercropping system at low N levels of 0 and 50 kg/ha, but as the N level increased, the leguminous CCs improved the plant height and produced significantly higher plants than MM. Increasing the N level by more than 100 kg/ha did not increase the height of maize intercropped with mung bean (MMB), while the MA and MM reached constant height at 150 kg/ha N. Photo 3.1 in A2 Appendices section shows the visible height differences in maize subjected to different cover crop and monocropped maize. Besides that, MMB produced the highest plant height at 256.6 m, which is shorter than the maximum height achieved by the intercropped system in 2023 at 267.3 m by MCR. The MM and MA in 2024 also produced shorter plant compared to the 2023 with 24.47 cm and 12.16 cm shorter in MM and MA, respectively.

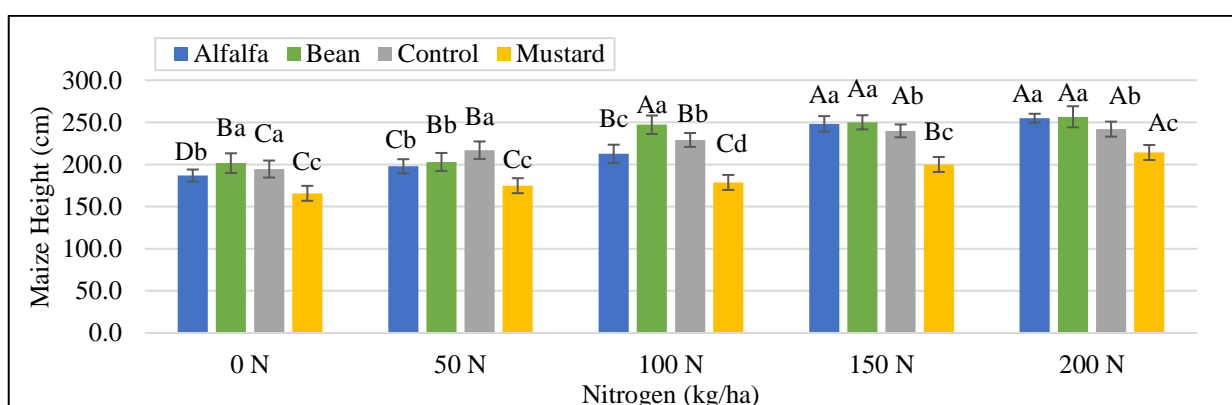


Figure 49. Plant height average at VT of maize plant intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rates on plant height. Different letters indicate significant difference at $p < 0.05$.

Moreover, the integration of mung bean caused a significant improvement in the SPAD values of maize plants compared to MM and the other two CCs at all N levels except 100 kg/ha N (Figure 50). Meanwhile, alfalfa significantly improved maize SPAD value at N levels higher than 100 kg/ha but did not increase SPAD value at low N levels compared to MM. Nevertheless, white mustard significantly decreased maize SPAD values at all N levels compared to MM, with significant improvement at 150 kg/ha and 200 kg/ha N than at lower N rates. At 200 kg/ha N, the

MM and MA in 2024 also produced lower SPAD values with 11.8 % and 2.02% lower compared to 2023.

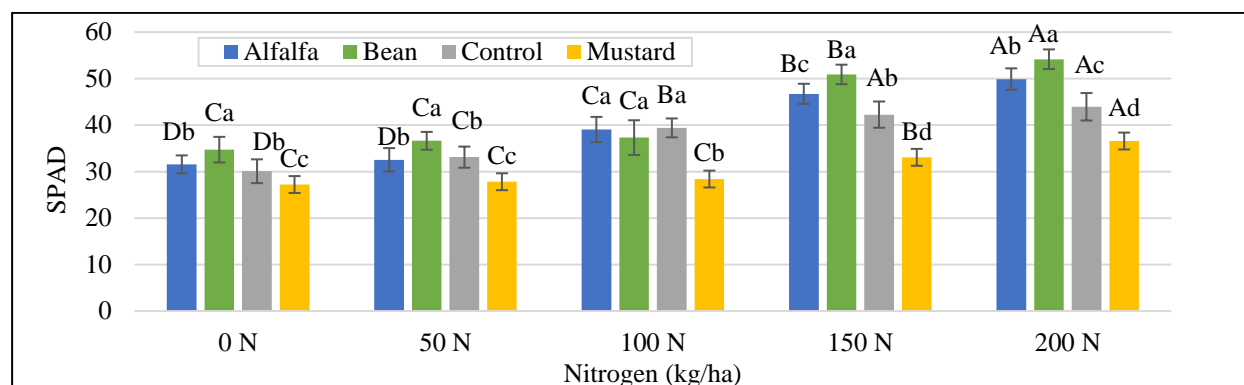


Figure 50. SPAD value average at VT of maize plant intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rate on SPAD value. Different letters indicate significant difference at $p < 0.05$.

Furthermore, Figure 51 presents that the integration of white mustard with maize significantly decreased the maize leaves area (LA) compared to MM and the leguminous intercropping system at all N levels. At low N levels, no significant increase in LA was observed between MA and MM while increasing the N levels to 100 kg/ha and 150 kg/ha produced a significantly higher LA in MA than in MM. At 200 kg/ha N, both MM and MA showed no significant increase in LA than at 150 kg/ha. Our results also showed that mung bean improved the maize leaves area at all N levels compared to MM and was higher than MA at low N levels and 200 kg/ha N.

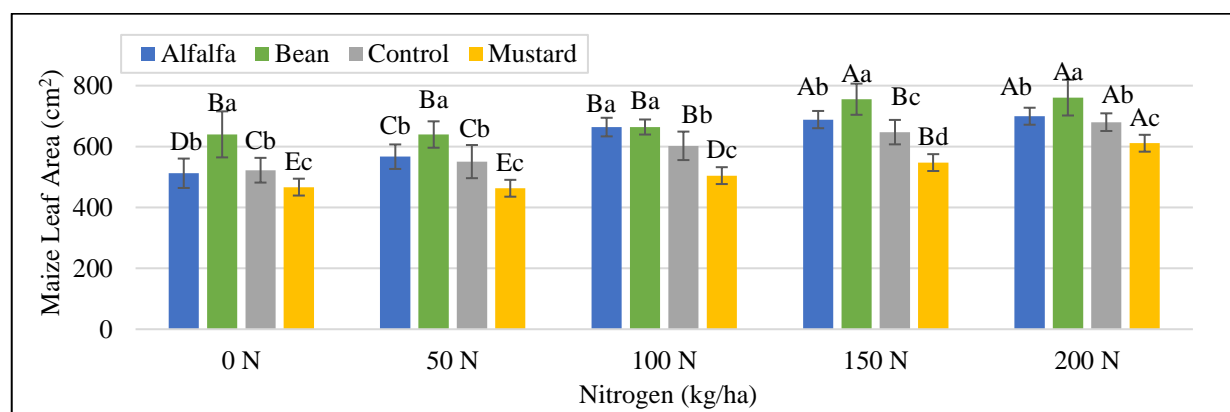


Figure 51. Leave area average at VT of maize plant intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rate on LA. Different letters indicate significant difference at $p < 0.05$.

Meanwhile, the ANOVA results in Table A.34 in the Appendices section, show that the N levels, type of CC and their interaction significantly influenced the maize leaves area index (LAI) with large size effect. Figure 52 illustrates that, maize LAI increased gradually as the N level increased for all CC treatments and MM. Similarly to the other parameters, white mustard also reduced maize LAI compared to the other treatments, while mung bean improved the LAI values at regardless the N rates. MA improved maize LAI only at 100 kg/ha and showed no significant difference at other N levels compared to MM.

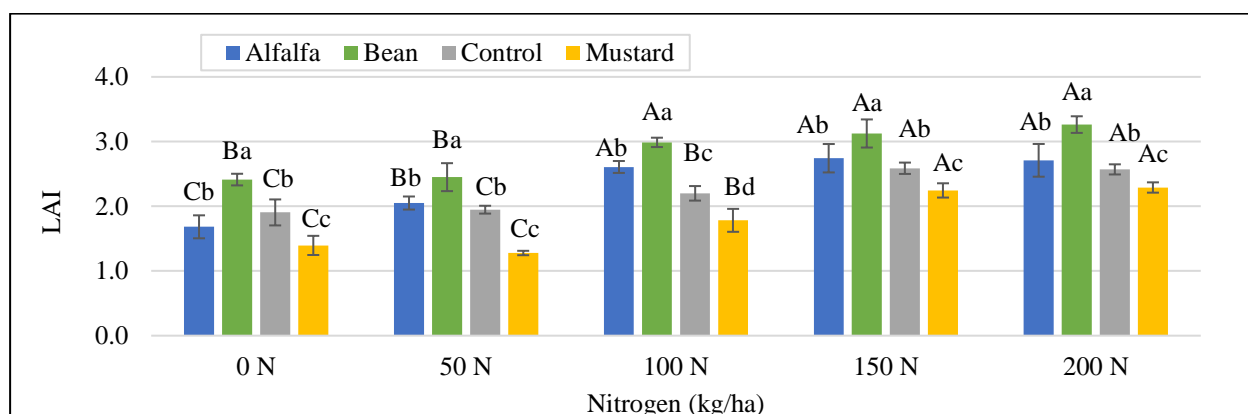


Figure 52. Leave Area Index (LAI) average at VT of maize plant intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rate on LAI. Different letters indicate significant difference at $p < 0.05$.

4.6.2 Maize harvested yield

The MANOVA results in Table A.35 in the Appendices section show that the type of cover crops (CC), different N treatment and their interaction had a significant medium to large effect on yield quantity parameters with $p < 0.001$. Follow-up ANOVA results in Table A.36 in the Appendices section show that all factors and their interaction had significant effects on maize ear weight (EW), ear length (EL), kernel weight (KW) and grain:cob ratio (G:C) with $p < 0.001$. Based on the partial η^2 , the individual factors had bigger effects on the yield variables compared to their interaction.

Furthermore, Figure 53 reveals that the maize EW significantly increased as the N rate increased from 50 to 200 kg/ha in all the treatments. The results also show that MMB produced significantly the highest EW while MWM produced the lowest EW compared to other intercropped combinations and MM at all N levels. MA also improved maize EW at 0, 150 and 200 kg/ha N. Besides that, the MM showed an increment in EW as the N rate increased from 100 to 200 kg/ha compared to 2023, where the EW reached a constant weight at 100 kg/ha N. However, the year 2024 revealed a decline in maize EW compared to 2023, with the MM producing an average of

217.33 g of EW in 2023 compared to 148.67 g 2024. MRC achieved the highest ear weight in 2023 at an average of 246.03 g, while in 2024, the highest EW was achieved by MMB at an average of 188.2 g.

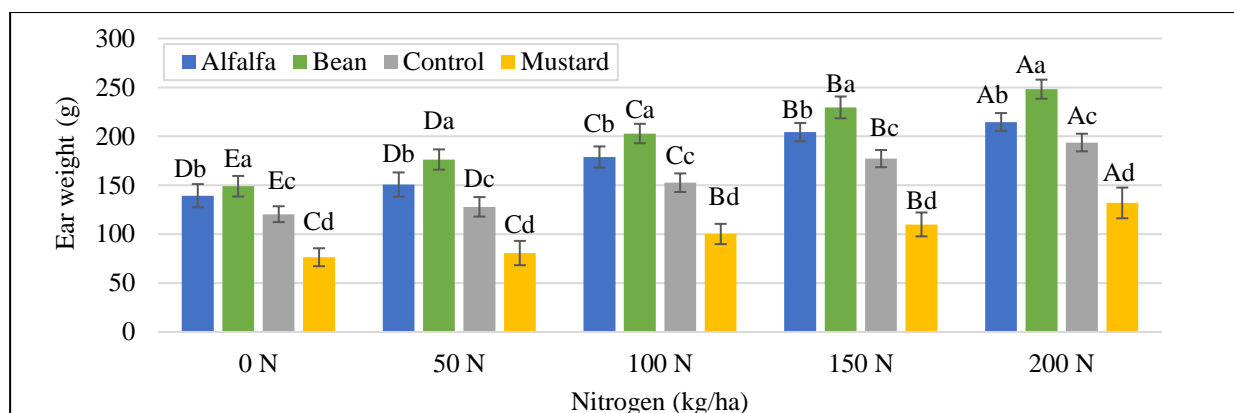


Figure 53. Ear weight average of maize intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rate on EW. Different letters indicate significant difference at $p < 0.05$.

Moreover, the maize EL also produced a gradual increasing pattern as the N rate increased, as shown in Figure 54. Similar to the EW, the MMB produced the longest maize ears, while white mustard reduced the maize ears significantly compared to the other intercropped combination and MM. Meanwhile, alfalfa significantly improved maize ear length at higher N levels between 100 to 200 kg/ha compared to MM. Lastly, only MMB improved the EL as the N level increased to 200 kg/ha, while the other three treatments showed no significant difference with the EL at 150 kg/ha N.

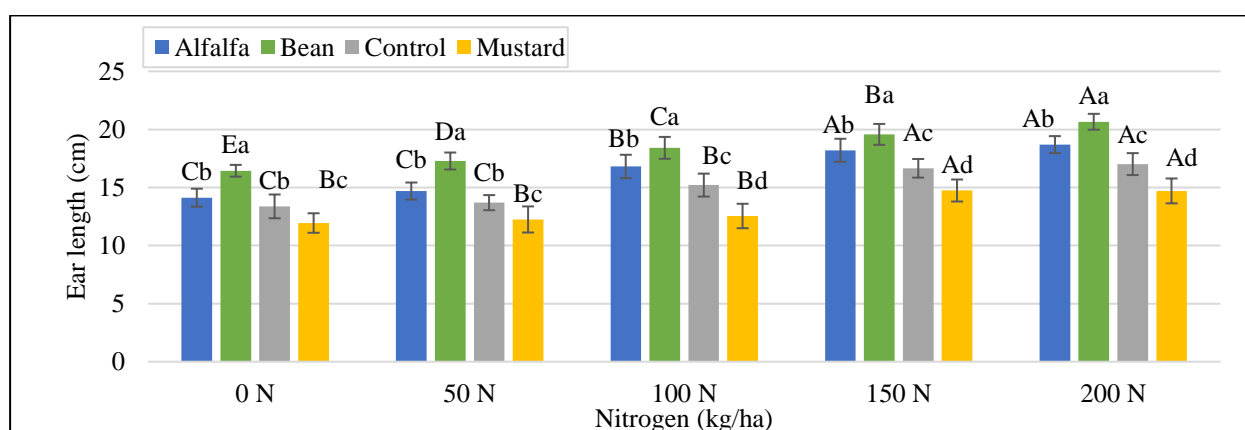


Figure 54. Ear length average of maize intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rate on EL. Different letters indicate significant difference at $p < 0.05$.

Furthermore, maize-legume integration significantly improved the G:C ratio and KW in maize, while the white mustard severely reduced these two parameters compared to the MM (Figure 55 and Figure 56). MMB produced the highest G:C increase at 100 kg/ha with 23%, while the KW showed the highest increment rate of 34.51% at 150 kg/ha N compared to MM. MMB improved the G:C ratio up to 200 kg/ha N, while no significant difference in the ratio between MA and MM at 200 kg/ha N. MM and MWM required an N rate of more than 100 kg/ha to increase the G:C ratio significantly. Mung bean proved to be superior in increasing maize KW compared to alfalfa, especially with the presence of N. Lastly, all intercropped maize and MM showed no significant increment in KW as the N rate increase from 150 kg/ha to 200 kg/ha. The KW of MM in 2024 was also lower than 2023 with 8.26% difference at 200 kg/ha N.

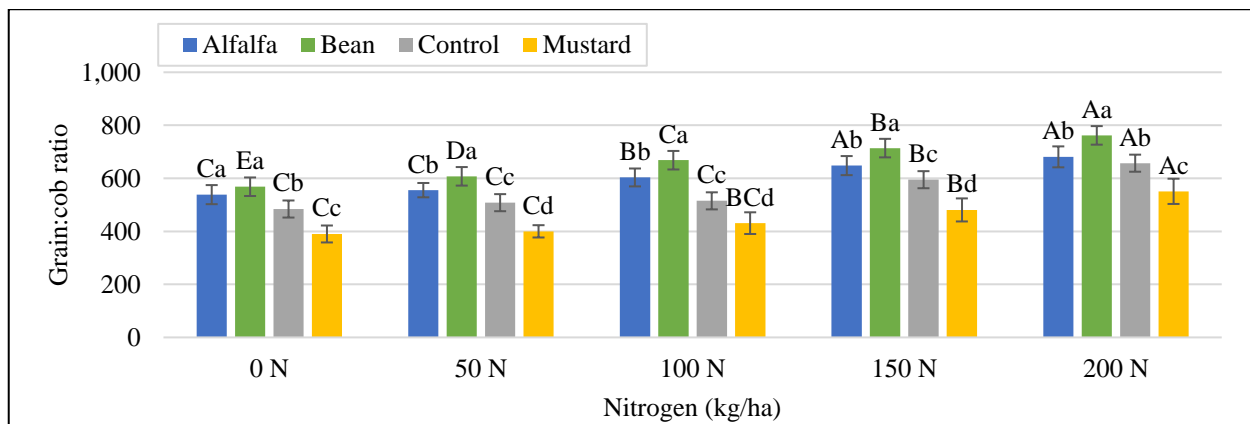


Figure 55. G:C ratio average of maize intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rate on G:C ratio. Different letters indicate significant difference at $p < 0.05$.

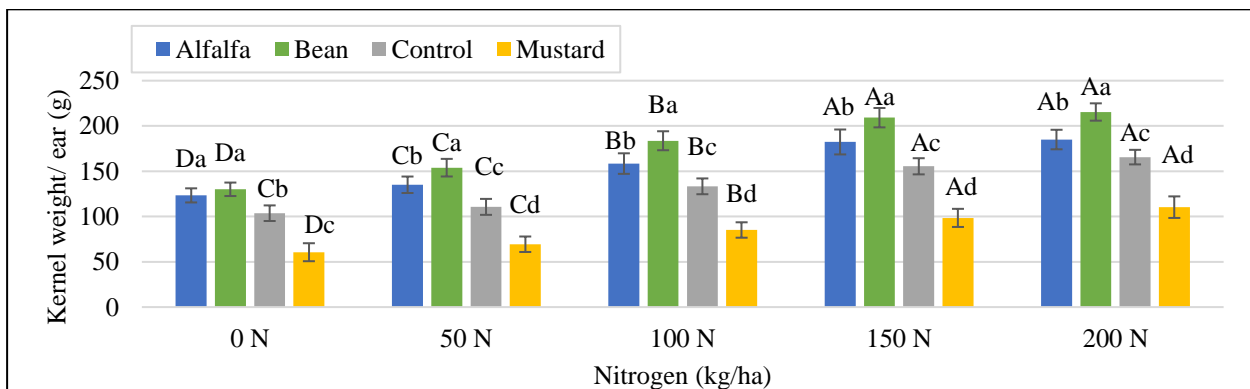


Figure 56. Average kernel weight per ear of maize intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rate on KW. Different letters indicate significant difference at $p < 0.05$.

Furthermore, a univariate ANOVA result in Table A.37 in the Appendices section, showed that the two factors and their interaction significantly affected the thousand kernel weight (TKW) of maize with $p < 0.001$ and with a large effect size.

Figure 57 shows that the TKW of maize-legume intercropped increased as the N level increased to 150 kg/ha. Increasing the N rate to 200 kg/ha significantly decreased the TKW value compared to the 150 kg/ha N. However, both leguminous CCs significantly increased maize TKW compared to the MM at all N levels except at 100 kg/ha, as no significant difference was observed between these three treatments. MMB showed the highest TKW increase, ranging between 6.1 – 16.4% elevation compared to MM. Meanwhile, white mustard severely reduced maize TKW with a significant reduction of around 20.3 – 27.4% depending on the N levels, while increasing the N rate more than 100 kg/ha did not increase the TKW of MWM significantly. Lastly, the TKW value in 2024 was also smaller than the 2023 value. The highest value achieved was 292.9 g in 2024 compared to 317.77 g in 2023, both in intercropped maize. Meanwhile, the MM's highest TKW was at 303.74 g and 261.36 g in 2023 and 2024, respectively.

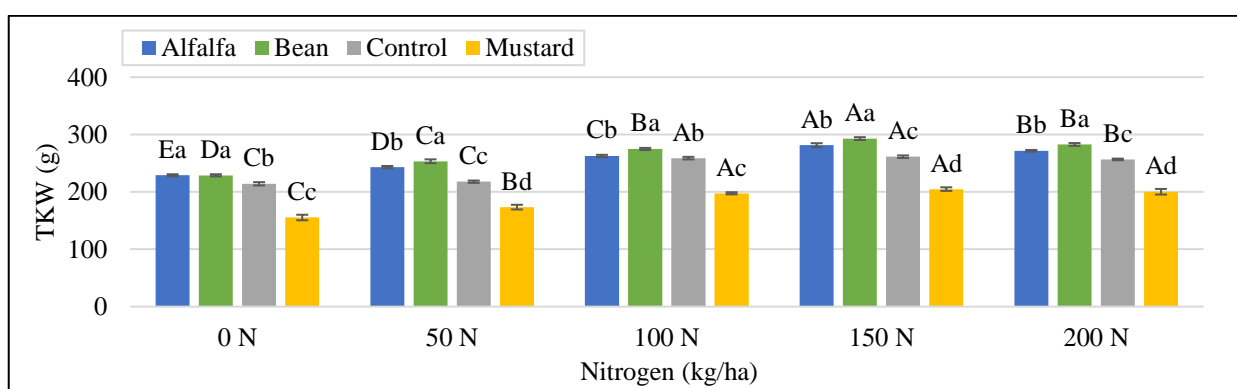


Figure 57. Thousand kernel weight (TKW) average of maize intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rate on TKW. Different letters indicate significant difference at $p < 0.05$.

4.6.3 Yield chemical composition

The MANOVA results in Table A.38 in the Appendices section show that the type of cover crops (CC), different N treatment, and their interaction revealed a significant large effect on yield chemical composition parameters with $p < 0.001$. Meanwhile, the follow-up ANOVA results in Table A.39 in the Appendices section show both factors and their interaction significantly affecting the maize moisture, oil, protein and starch content with $p < 0.01$. Based on the partial η^2 , both factors and their interaction had a large effect on all the chemical composition parameters tested.

Table 13 reveals that the grain's moisture content increased as the N rate increased in all CC treatments and MM. Besides that, all CC treatments and MM produced the highest moisture content at 200 kg/ha N, with MMB showing the highest value. At 0 to 100 N kg/ha, intercropped maize contained significantly less moisture than the MM grains. As the N rate increases to 150 kg/ha and 200 kg/ha, no significant difference in moisture content was observed between MMB and MM, while MA and MWM grains had significantly lower moisture.

Table 13. Mean±standard deviation of moisture content (%) of maize seeds intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).

Nitrogen (kg/ha)	Moisture content (%)			
	Alfalfa	Mung Bean	Mono-maize	Mustard
0 N	10.07±0.1Db	10.02±0.1Db	10.27±0.05Ea	10.27±0.12Ca
50 N	10.22±0.1Cb	10.35±0.1Cb	10.93±0.21Da	10.4±0.24Cb
100 N	10.37±0.1BCc	10.38±0.1Cc	11.23±0.08Ca	10.75±0.05Bb
150 N	10.48±0.2Bb	11.47±0.1Ba	11.58±0.1Ba	10.85±0.19Bb
200 N	10.82±0.1Ad	12.12±0.1Aa	11.8±0.21Ab	11.53±0.16Ac

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rate on moisture content. Different letters indicate significant difference at $p<0.05$.

The oil content analysis in Figure 58 reveals that the oil content in MM and all CCs treatment increased as the N rate increased. Mung bean proved to improve the maize oil content as the N levels increased to 150 and 200 kg/ha N, and no significant difference was observed at N levels below this level. Meanwhile, MA only caused a significant reduction in oil content compared to MM at 50 kg/ha N, but no significant difference was observed at other N levels. However, white mustard significantly reduced maize oil content, especially at low N levels, with the highest percentage difference of 7.6% at 50 kg/ha N, but the reduction percentage was reduced as the N level increased.

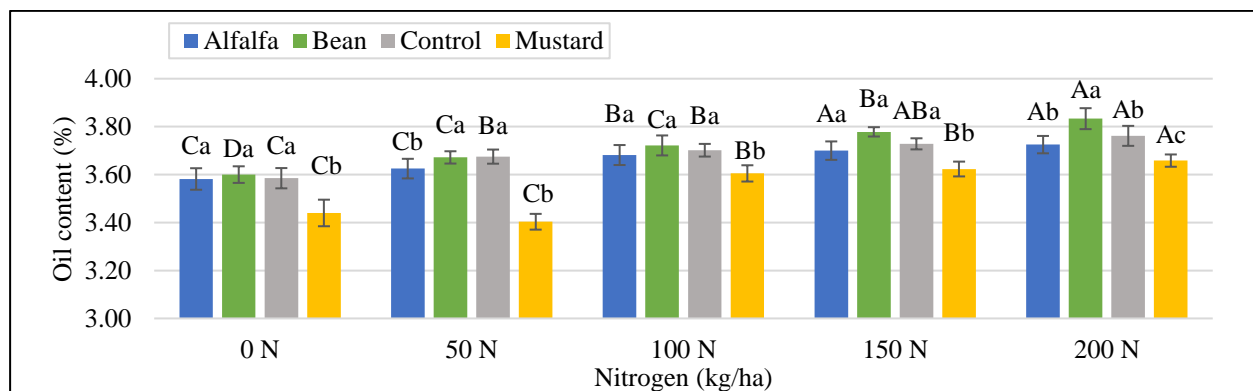


Figure 58. Oil content (%) average in maize seeds intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rate on oil content. Different letters indicate significant difference at $p<0.05$.

In contrast, the NIR analysis shows that mung bean incorporation decreased the maize protein content significantly at all N levels except at 200 kg/ha compared to the MM (Figure 59). Meanwhile, white mustard only reduced maize protein content at N levels 150 – 200 kg/ha and showed higher or no significant difference at lower N levels. MWM also produced significantly higher protein content than MMB at all N levels except 200 kg/ha. Even though MA produced significantly lower protein content than MM with the absence of N, but with the increase of N levels, the MA significantly increased maize protein content, especially at higher N levels. Besides that, the 2024 grains produced higher protein content compared to 2023. The highest value in 2023 was 6.28% in MRC, while in 2024, 8.22% was recorded in the MA system. The MM also showed a 61% protein increase in 2024 compared to 2023.

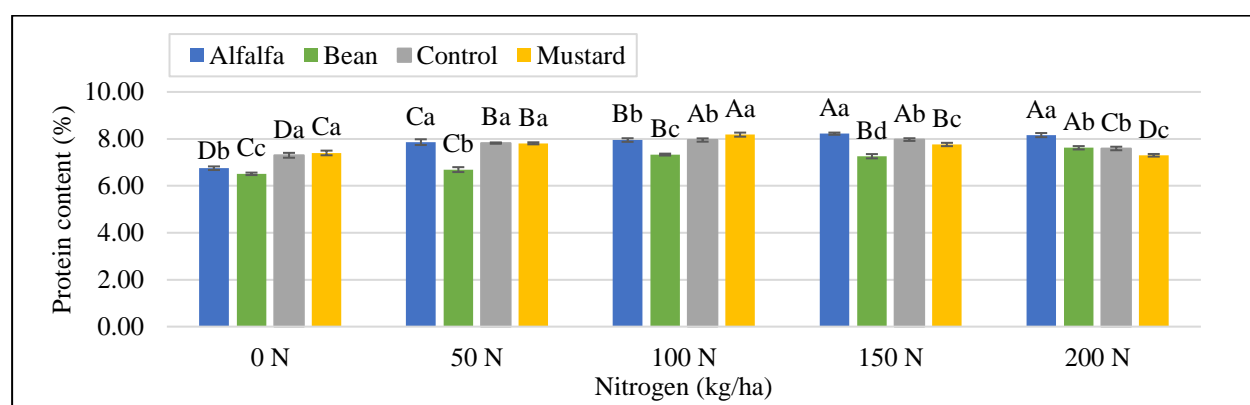


Figure 59. Protein content (%) average in maize seeds intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rate on protein content. Different letters indicate significant difference at $p < 0.05$.

Lastly, mung bean also increased the maize starch contents at all N levels, and the largest increase was at 100 kg/ha, with a 2.2% increase over MM (Figure 60). Alfalfa also increased maize starch content but only with the presence of 50 to 200 kg/ha N, and the percentage was lower than MMB's at a 0.9% increase of 200 kg/ha N. Nevertheless, white mustard significantly reduced maize starch content at all N levels with a percentage reduction of around 1.04 – 2.2% depending on the N levels. In comparison to 2023, MA produced 0.67% higher starch content, while MM produced 0.35% lower in 2024.

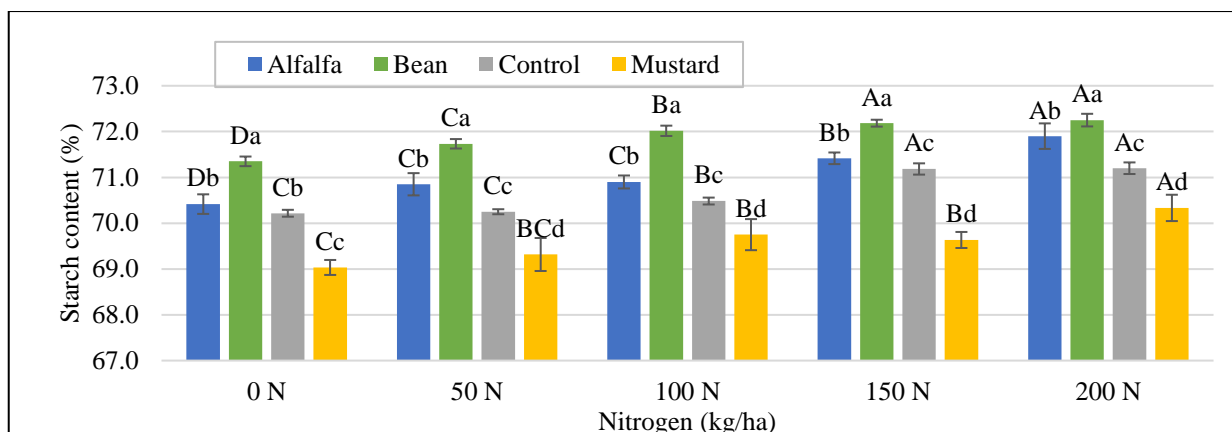


Figure 60. Starch content (%) average in maize seeds intercropped with different cover crops at various nitrogen levels (2024, MATE-Gödöllő).

Capital letters compare the effect of the same CC species under different N rates, while lowercase letters compare the effect of different CC species under the same N rate on starch content. Different letters indicate significant difference at $p < 0.05$.

4.6.4 Cover crop dry mass

Lastly, the ANOVA result shows that the N treatment also significantly affecting the dry mass of the cover crops cultivated between the maize rows with alfalfa $F(4,10) = 463.95, p < 0.001$ partial $\eta^2 = 0.995$, mung bean $F(4,10) = 196.141, p < 0.001$ partial $\eta^2 = 0.987$, white mustard $F(4,10) = 272.473, p < 0.001$ partial $\eta^2 = 0.991$, and mung bean seed $F(4,10) = 459.334, p < 0.001$ partial $\eta^2 = 0.995$. Table 14 summarises that all cover crops achieved the highest dry mass at 200 kg/ha, with mung bean producing the highest value compared to alfalfa and white mustard. Besides that, mung bean also managed to produce seeds, which was also significantly affected by the increase in N rate. Meanwhile, no significant difference was recorded in all three CC dry masses as the N rate increased from 150 to 200 kg/ha. Photo 3.4 in the Appendices section shows the cover crops condition at maize reproductive stage and after harvesting period.

Table 14. Mean \pm standard deviation of cover crop dry mass harvested within 0.2 m² area between the maize rows (2024, MATE-Gödöllő).

Nitrogen (kg/ha)	Dry mass (g)			
	Alfalfa	Mustard	Mung bean	Mung bean seeds
0 N	37.5 \pm 1.80E	53.9 \pm 2.80E	73.5 \pm 0.90E	6.75 \pm 0.70E
50 N	45.3 \pm 2.20D	68.0 \pm 1.60D	81.8 \pm 3.90D	10.1 \pm 0.20D
100 N	74.4 \pm 1.95C	83.3 \pm 3.10C	95.1 \pm 3.80C	14.3 \pm 0.57C
150 N	84.6 \pm 1.35B	96.7 \pm 2.10B	115.3 \pm 2.65B	19.75 \pm 0.45B
200 N	95.8 \pm 2.60A	107.1 \pm 0.90A	126.6 \pm 0.70A	24.15 \pm 0.75A

4.6.5 Discussion

First of all, the different weather patterns in 2023 and 2024 may influence the difference in growth and yield produced by of MA and MM between these two years. The field received 282.3 mm and 160.3 mm of rain between May and September in 2023 and 2024, respectively. The

average temperature in 2023 was also lower compared to 2024, as shown in Figure 4. It was visible that the maize planted in 2023 produced higher vegetative growth and yield production in MM and MA. The maize planted in 2024 showed a reduction in all generative and yield parameters compared to 2023 except for grain: cob ratio, oil and protein content, which were higher in 2024. The yield quantity and quality difference between 2023 and 2024 may be due to the different amounts of precipitation received in both growing seasons. A similar observation was recorded in Experiment 3, where the oil and protein content were higher in dry and hotter conditions in 2022 than in 2023. In rainfed systems, rain is essential to solubilise the chemical fertilizer and make it available for the crops. However, excess water and high temperature will also increase N leaching from the soil (JABLOUN et al. 2015). Besides that, high soil moisture content is also essential for the nodulation formation in leguminous plants (KASPER et al. 2019). Lastly, based on several yield parameters including G:C ratio, kernel weight and starch content, it was visible that alfalfa improved maize yield performance at various N levels in 2024 than only at 0 and 50 kg/ha N in 2023.

Moving on, based on our results, intercropping maize with leguminous cover crops could improve maize performance, while the white mustard from the *Brassicaceae* family was detrimental to the companion maize plant. Similar to the 2023 results, the leguminous species either increased or did not affect the growth of the neighbouring maize, with mung bean having more superior effects than alfalfa on maize growth and yield production. Mung bean incorporation increased by 25.4 – 39.1% maize kernel weight /ears compared to 11.7 – 22.0% by alfalfa. A study in Indonesia found maize and mung bean intercropped increased maize yield between 2.9 – 7% (SYAFRUDDIN and SUWARDI 2020).

Besides that, mung bean also revealed the potential to reduce N requirement in maize. Based on the kernel weight data, MMB required 100 kg/ha N to produce 153.93 g kernel/ear, while the MM required 150 kg/ha N to produce almost the same amount at 155 g kernel/ear (Figure 56). Similarly to the starch content, at 0 kg/ha N, MMB produced higher starch content than MM starch content at 100 kg/ha N (Figure 60). A recent study reported that, intercropping maize and legumes improved N uptake by regulating rhizosphere N transformations and promoted proliferation of rhizosphere N-acquiring microbiome (WANG et al. 2024). Meanwhile, a study on loamy sand soil in Cambodia found no significant difference in MMB intercropping compared to the MM in terms of maize agronomic traits and yield. However, the growth and yield were not reduced, but the weed suppression increased with the increase in the mung bean seeding rate (RO et al. 2023). Besides that, despite increasing maize growth and yield, it was discovered that the mung bean yield was not improved in this MMB system compared to sole cultivation of the crops (KHA et al.

2014). The benefits of leguminous species including alfalfa in improving maize crops in intercropping systems were discussed in section 4.5.5 with references to many successful studies.

On the other hand, many studies have presented the negative effect of intercropping on the main crop, especially with non-leguminous plants. It was mentioned that non-leguminous companions have higher N scavenger capacity compared with leguminous crops (FAGERIA et al. 2005). Meanwhile, it was discovered that grass cover crops reduced leaf N content in grape vines compared to leguminous cover crops (VUKICEVICH et al. 2019). In our study, the incorporation of white mustard reduced maize growth and yield traits, including TKW, by up to 27.4 % reduction. The results showed a robust interspecific competition between these two crops in essential inputs required for generative growth and yield production. In our study, white mustard growth was also affected by maize, as the plant failed to produce significant seeds at the end of the trials.

Interspecific competition usually involved two competition mechanisms that will influence the interaction between different species in an area. Resources competition and interference competitions are the two mechanisms commonly involved in interspecific competition (ASCHEHOUG et al. 2016). Resources competition mainly defined as the capture of essential resources such as light, water, and soil nutrients from a common, finite pool by neighbouring individuals. Meanwhile interference competition is defined by the ability of one plant to directly suppress its neighbour's ability to acquire resources or grow. Plant allelopathy effect is commonly linked as the interference competition mechanism (ASCHEHOUG et al. 2016).

In our study, both type of mechanisms may be involved in the maize-white mustard system. In our study, it was discovered that white mustard had a vigorous growth rate compared to maize and the other cover crops. At four weeks after emergence, inflorescence emergence was already observed in white mustard plant while maize was only at maize V4 stage. Photo 3.2 show white mustard flowering stage six weeks after sowing in comparison to the neighbouring maize. White mustard is known to have short life cycle 85 - 95 days compared to 105 – 120 days of maize life cycle (MITROVIĆ et al. 2020). This rapid growth is most likely depleted the nutrient and water resources around maize root zone and affecting its growth. As the maize canopy were not obstructed by the white mustard, light resources competition was not observed. Furthermore, it was discovered that white mustard generated a water-soluble compound which could inhibit wheat germination and seedlings growth (SAEEDIPOUR 2010).

As evidenced by several studies, the impact of inter-specific competition in an intercropping system was demonstrated in various species combination. A study discovered that intercropping maize with sunflower reduced the yield of 20% of maize grain compared to monocrop maize (COLL et al. 2012). Meanwhile, it was discovered that wheat will inhibit root

and shoot growth in the maize-wheat intercropping system, which consequently affects the P uptake ability in maize (ZHANG, CHEN, et al. 2012). Even though leguminous species produced many positive results but, some combinations did not benefit maize production. As presented in our results in sections 4.5 and 4.6, alfalfa showed inconsistent effects on maize growth and yield production. In 2023, alfalfa improved the grain's chemical composition but not the yield quantity harvested compared to MM. In a study of maize and *Trifolium ambiguum* M. Bieb, it was discovered that the leguminous cover crop also did not increase maize yield nor reduce the N requirement due to the excessive competition by this species (SAWYER et al. 2010). Therefore, it is crucial to choose crop combinations that bring benefits to each other and not act like a weed.

Additionally, the 2024 results also highlight the significant role of N level in promoting maize growth and yield production in both the intercropping system and MM. Significant improvement was observed in various parameters as the N rate increased. The results also supported the latest publication of (ADHIKARI et al. 2021) which show the effect of nitrogen on growth and yield of different hybrid maize varieties. Besides that, the MM reached a maximum TKW at 100 kg/ha while the grain: cob ratio continuously increased as the N rate increased to 200 kg/ha. Meanwhile, maize intercropped with the two legumes reached the highest TKW at 150 kg/ha and reduced at 200 kg/ha, showing it is not economical to increase the N levels by more than 150 kg/ha. The finding also supported the result in 2023 and the results from trials conducted in Hungary which suggested the amount of 120 – 180 kg/ha N for single maize production (SZÉLES et al. 2018, 2023). Similarly, elevation in N levels also significantly increased the MWM performance, yet the levels of increase never reached or surpassed the MM except for the protein content.

Furthermore, our study found that maize integrated with white mustard produced higher protein content than maize with mung bean at various N levels. A 2023 study in Hungary also found similar results where higher protein content was produced by maize interseeded with *Brassicaceae spp* with white mustard produced the highest value while the lowest protein was produced by maize intercropped with leguminous pea and alfalfa (FODOR et al. 2024). It was discovered that under water deficit conditions, protein content in maize kernel increases due to low starch content in the kernel (OTTAIANO et al. 2021; HUANG et al. 2023). Therefore, we can conclude that the incorporation of white mustard triggered water deficit conditions around the maize rhizosphere. While the uncovered soil surface also increased the rate of moisture evaporation and introduced similar dry conditions around the maize root zone.

Therefore, based on our results, farmers should be able to decide which cover crop is more suitable depending on the end target for the maize production. Intercropping maize with white

mustard may bring considerable benefit for forage production with the increase of protein content. However, farmers may require different agro-techniques to ensure higher productivity and yield volume in MWM, such as delayed planting to reduce the competition effects with maize. A study in China reported that delaying watermelon planting significantly reduced competition for maize (HUANG et al. 2019). Meanwhile, intercropping maize with legumes is recommended if a high yield volume with high starch content is targeted and more desired than other chemical components.

In conclusion, based on the two-year trials, the success of the intercropping system was highly influenced by the species selection, nitrogen levels and climatic factors. In the intercropping system, companion species should not compete with the nutrients and water while increasing the yield to compensate for the additional cost invested. Based on our results, *Brassicaceae* species reduced maize yield performance except for protein content, while alfalfa had higher benefits on maize in dry years at all N levels. In relatively wet year, alfalfa improved maize growth at low N levels. While mung bean displayed the potential to reduce N requirement, red clover and crimson clover also proved to increase the yield in maize, especially at low and high N conditions. Based on the Hungarian weather pattern in the past 3 years, we would suggest that the cover crop be planted at the same time as the maize to allow better plant establishment. Besides that, it is important to choose species or varieties that germinate and emerge quickly for better incorporation. Application of N fertilizer to the soil at the maize V6 stage may allow the nodulation process to occur in the leguminous plants. Low nitrogen (N) content can significantly enhance legume nodulation and nitrogen fixation, as evidenced by various studies (FRUNGILLO 2022; ZHAO, SUN, et al. 2022).

5. CONCLUSION AND RECOMMENDATION

5.1 Conclusion

The study performed on the germination test on three different legume species, alfalfa, red clover, and chickpea, revealed the significant effect of temperature and salinity in influencing the germination rate, radicle length, and plumule of the legumes seedlings. The study found that increasing the temperature will improve the germination rate at low salt concentrations in all three species, but increasing salt concentration will reduce the germination rate at higher temperatures for red clover and chickpea. Alfalfa showed a higher tolerance to salt stress with the highest germination rate at 20°C and 1% NaCl and the ability to germinate at 1.5% NaCl. The radicle and plumule growth gradually decreased as the salt concentration increased in all temperatures, but higher temperatures improved the final length of the radicle and plumules compared to low temperatures. Based on all the parameters, chickpea showed to have the lowest tolerance to salt stress compared to alfalfa and red clover.

Besides that, the study performed on several maize variations found the significant effects of incubation days, salinity, temperature, and their interaction on maize germination performance. The results showed that the combination of high temperature and salinity caused more deterioration in germination initiation and seedlings growth compared to the combination of low temperature and salt stress. At low temperatures, germination performance was significantly lowest regardless of the presence of salt stress. Furthermore, within the same species, the varietal effect significantly affected the high temperature and salt tolerance in maize. Under salt stress conditions, several varieties proved to perform better at 35°C than at 20°C. The established Margitta hybrid and several other variations proved to have the higher tolerance to salt stress and high temperature, with an excellent germination percentage and seedling growth compared to the other tested variations. This result provides an essential input for farmers to choose the suitable variety that is most appropriate for their land soil and climate conditions.

Furthermore, the study also discovered the impact of different climatic conditions on nitrogen fertilizer efficiency in maize crops. The study found that maize cultivated in 2023 was more productive compared to maize in 2022 in terms of growth and yield productivity. The meteorological tower in the study site recorded that 2023 received significantly higher precipitation and lower average temperature than 2022. It was shown that in both years, the optimum rate of N fertiliser was between 100 and 150 kg/ha in terms of maximum grain number

and TKW. In terms of grain nutrient content, the drier climate in 2022 produced grains with lower starch but higher protein and oil than grain produced in 2023. Meanwhile, to produce grains with optimum starch content, 150 kg/ha N was the ideal rate in 2022, while in the wetter year of 2023, increasing the N rate up to 200 kg/ha will still be beneficial in influencing the increase in grain starch content.

The pot trial revealed that drought stress was more detrimental than salt stress in maize development. The study found that drought stress stunted maize vegetative growth and increased the anthesis-silking index, which prevented successful pollination compared to salt stress. Intercropping alfalfa or mung bean with maize improved the performance of maize crops in salt and drought stress conditions. However, alfalfa was superior to mung bean in drought conditions. Intercropping white mustard with maize amplified the abiotic stress effects as this species competes for the essential elements with the maize crops. Maize intercropped with white mustards had worse vegetative growth and reproductive development than sole maize without any companion crops.

The intercropped field trials found that the leguminous cover crop promotes maize productivity. In both studies, planting the companion leguminous crop seeds immediately after maize sowing improved the maize yield productivity. It was revealed that mung bean, red clover, and crimson clover improved various vegetative and yield parameters in maize, which was also positively influenced by the N level. Based on the TKW and starch content results, all leguminous cover crops tested have the potential to reduce the N requirement in maize cultivation compared to *Brassicaceae* species. Alfalfa provides better benefits to maize at lower N levels in the wetter year 2023 but did not change maize performance at higher N levels. However, in relatively drier years, alfalfa significantly improved the maize performance at all N levels. The study also revealed that white mustard was not a compatible companion to maize crops in terms of reducing maize vegetative growth, yield quantity, and grain starch content. However, intercropping maize and white mustard significantly increase the protein content in maize compared to intercropping maize-leguminous species.

Therefore, all the data presented from all six studies will provide several guidelines for farmers and producers in selecting the most appropriate crop species and varieties to be cultivated, especially in abiotic stress conditions. The studies also suggested the optimum application of nitrogen level in various intercropping and climate conditions. The final maize yield harvested and chemical composition should be considered by farmers when deciding on the intercropping combination. The results may allow farmers to intensify their land production in a more sustainable direction.

5.2 Recommendation

In this study, we broadcasted the cover crop seeds between the maize rows. Future studies should consider the use of an interseeder machine between the maize rows to plant the cover crops. Drilling the cover crop seeds into the soil allows seeds to be in full contact with the soil particles and allows a better germination rate than the broadcasting technique. Other than it will reduce the amount of cover crops seeds required, it can save more time and energy required for the cover crop integration between the maize.

Future studies should include the land equivalent ratio (LER) and nitrogen use efficiency (NUE) analysis in the intercropping studies. LER study required the cover crop cultivation to be planted separately, thus extra study area will be required. Overall economic benefits analysis should be carried out with consideration of every input required for the intercropping system.

Besides that, future studies should also investigate the effect of the intercrop system on the yield and nutrient composition produced by the cover crop species. As the cover crops have a value in forage industries, the potential value added is necessary to compensate for the additional seed and management cost spent by farmers.

Future studies should also consider the study on impact of mixed cover species intercropped with maize instead on single species cover crop intercropping. However, optimum rate of each species in the mixtures should be carefully calculated to minimise negative implication on main crop development and yield.

In this study, we carried out a pot trials to investigate the effects of intercropping in salt and drought stress condition. This study can be improved by increasing the sample size and usage of bigger pots for maize cultivation. The drought plots should be protected from being exposed to natural precipitation. The nodulation density of the leguminous cover crop can also be analysed throughout the study.

Besides that, future studies should also investigate the effect of the intercrop system on the yield and nutrient composition produced by the cover crop species. As the cover crops have a value in forage industries, the potential value added is necessary to compensate for the additional seed and management cost spent by farmers.

Further studies should also investigate the effects of delayed nitrogen application on the nodulation formation in the intercropping system.

6. NEW SCIENTIFIC RESULTS

1. The study found that alfalfa has the highest salt tolerance in the germination stage compared to red clover and chickpeas.
2. Genetic variability in maize significantly influences maize salt tolerance. High temperature conditions intensified saline stress during the germination phase of maize more significantly than lower temperatures.
3. Drought conditions in 2022 severely decreased maize productivity but increased the protein content regardless of the N levels. Application of nitrogen over 150 kg/ha was not economical as it did not improve the yield quantity and starch content in the dry year. However, in the wet year 2023, increasing N to 200 kg/ha increased starch content but not the yield quantity than 150 kg/ha N.
4. Alfalfa and mung bean species helped in increasing salt and drought stress tolerance in maize cultivation. However, alfalfa was more recommended in drought stress conditions than mung bean.
5. The leguminous intercropping system allows maize to increase yield performance at a higher nitrogen level of 150 kg/ha, which was stagnant in the mono-crop maize system.
6. White mustard was not suitable for intercropping with maize as it heavily competes for nutrients and water, which negatively affected the growth and maize yield. However, white mustard improved maize protein content compared to leguminous companion crops.
7. Based on the experimental field weather conditions, simultaneous maize and cover crop sowing allowed better cover crop establishment. Early cover crop sowing helped the crop to be stronger against warmer and drier weather in the later growing period. The early establishment also allowed the cover crops to utilise maximum solar radiation before being covered by maize canopy. Besides that, early sowing may allow the rhizobium nodule to form before chemical nitrogen application.

7. SUMMARY

Maize is not only used as human and animal food but also has high potential as a fuel alternative in the future. Global maize production is continuously threatened by ongoing climate change and the depletion of arable land. Extreme weather patterns have already affected maize production in Hungary and other part of the world. Growing stress tolerance maize variety is one of the ways to ensure global food security. Besides that, the high dependency on chemical fertilizer input to produce maximum yield has already been proven not to be a sustainable long-term plan. However, intensification of agriculture is unavoidable due to the expanding human population all around the world. Therefore, the agriculture industries need to find a solution to increase land productivity with less dependency on chemical fertiliser and sustaining soil health. Intercropping may allow sustainable intensification in the agriculture sector with higher overall productivity but less chemical input. Therefore, six experiments were carried out to determine the impact of abiotic stress, different nitrogen applications, and intercropping growing systems on maize production. All trials were carried out at the Hungarian University of Agriculture and Life Science (MATE) Gödöllő, Hungary.

The study began with a germination test of three legume species to determine the role of temperature on the germination activity of leguminous crops exposed to saline conditions. The study focuses on the germination performance in salt stress conditions of alfalfa, red clover, and chickpea under different temperatures. The seeds were incubated at two different temperatures and four levels of salt stress. The germination percentage, radicle length, and plumule length were analysed in this study. The results indicated that the increase in temperature reduced the germination performance in high salt stress in two legume species, while alfalfa displayed the highest salt tolerance compared to the other two species.

The second experiment was carried out with the aim of determining the impact of combined temperature and salinity stress on 16 different maize varieties. The maize seeds were incubated at 15°C, 20°C, and 35°C and two sodium chloride (NaCl) levels simultaneously. Germination percentage, root growth, shoot growth, root: shoot length ratio, and seed vigor index (SVI) were recorded and analysed. The results revealed that the presence of salinity reduced maize germination qualities at all three temperatures tested. However, the reduction rate in the germination parameters was more significant at higher temperatures than at lower temperatures with the combination of salt stress. Besides that, several varieties were proven to have higher

tolerance than others, proving that the varietal effect influences the seed tolerance towards a combination of salt and temperature stress.

Meanwhile, the third experiment investigated the impact of different N rate applications on maize growth and yield production in two different climatic conditions in 2022 and 2023. The field study was carried out with five different N rates, 0, 50, 100, 150, and 200 kg/ha, on five 2 x 4 m plots. The results found that maize cultivated in 2023 was more productive compared to maize in 2022 in terms of growth and yield productivity in all N levels. The increase of N levels also significantly increased maize productivity. In both years, the optimum rate of N fertiliser is between 100 to 150 kg/ha to produce the highest grain number and TKW. Meanwhile, the drier climate in 2022 produced grains with lower starch but higher protein and oil than grain produced in 2023. Meanwhile, to produce grains with optimum starch content, 150 kg/ha N was the ideal rate in 2022, while in the wet year of 2023, increasing the N rate up to 200 kg/ha will still be beneficial in influencing the increase in grain starch content.

The fourth experiment was carried out to identify the potential use of cover crops in alleviating abiotic stress in maize cultivation. Alfalfa, mung bean, and white mustard were planted with maize seeds in 26 cm pots to determine the best companion plants for maize in different abiotic stress. The pots were subjected to three different treatments: no stress (NS), non-irrigated to impose drought stress (DS), and salinity stress (SS), which were applied weekly at 100 mM NaCl. The results discovered that drought stress is more detrimental than salt stress for maize growth. The DS treatment caused severe vegetative growth delays and increased anthesis-silking index (ASI), which prevented successful pollination. Intercropping alfalfa or mung bean with maize improved the performance of maize crops in salt and drought stress conditions. However, alfalfa was superior to mung bean in drought conditions. Intercropping white mustard and maize amplified the abiotic stress effects as this species competes for the essential elements required by maize crops. Maize intercropped with white mustards had worse vegetative growth and reproductive development than sole maize without any companion crops.

The fifth experiment identified the benefits of intercropping maize and leguminous cover crops on maize productivity at different N levels. The study was conducted using a randomized complete block design (RCBD) with three replications, with each replicate consisting of 5 plants. Alfalfa, red clover, and crimson clover were planted separately within the 75 cm maize rows. Maize seeds were planted at a density of 75,000 plants per hectare, and the cover crop seeds were applied one day after the sowing process by broadcasting the seeds into the soil between the maize

rows. Application of N ammonium nitrate was carried out at maize V5-V6 stage, at the levels of 0 kg/ha, 50 kg/ha, 100 kg/ ha, 150 kg/ha, and 200 kg/ha N in each of the intercrop and monocrop plots.

Lastly, the sixth experiment investigated the effect of different cover crop families on maize intercropping systems at different N levels. Alfalfa and mung bean from the *Fabaceae* family, while white mustard from *Brassicaceae* were planted separately between maize rows. The trial was conducted using a strip plot design with three replications, with each replicate consisting of 5 plants. The planting and N application methods were similar to the fifth experiment except that mung bean was sowed at 4 seeds per maize plant.

The fifth and sixth experiments found that the leguminous cover crops promoted maize productivity. It was revealed that mung bean, red clover, and crimson clover improved various vegetative and yield parameters in maize, which was also positively influenced by the N level. Alfalfa provided better benefits to maize at lower N levels in the wetter year 2023 but did not change maize performance at higher N levels. However, in relatively drier year, alfalfa significantly improved the maize performance at all N levels. In contrast, it was found that white mustard was not a compatible companion to maize crops due to adverse effects on maize vegetative growth, yield quantity, and grain starch content. However, intercropping maize and white mustard significantly increased the maize protein content at 100 kg/ha N, while alfalfa improved maize protein content at 150 – 200 kg/ha N. Lastly, mung beans reduced maize protein content at all N levels except at 200 kg/ha.

APPENDICES

A1. Bibliography

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A2. Research Activities /Photos

1) Research activities (Experimental Research 4)



1.1 Control, salinity treatment, drought treatment maize height difference



1.2. Irrigation system



1.3. Maize-mung bean (salt stress)



1.4. Maize-alfalfa (VT stage) (No stress)



1.5. Harvesting day

2) Research activities (Experimental research 5)



2.1. 4 weeks after sowing (crimson clover)



2.2. 5 weeks after sowing (alfalfa)



2.3 Rhizobium nodulation in crimson clover



2.4 Maize-red clover



2.5 Maize-alfalfa



2.6 Reproductive stage (maize-alfalfa)

3) Research activities (Experimental research 6)



3.1 Maize height difference



3.2 Maize-white mustard (6 weeks after sowing)



3.3 Early maize vegetative growth and cover crop establishments (alfalfa, mung bean & mustard)



3.4 Maize reproductive stage and CCs condition after maize harvest

A3. List of Statistical Analysis Tables

Table A.1. Three-way ANOVA for germination percentage, radicle length and plumule length of cover crop germinated under different temperature and salinity levels (2021, MATE-Gödöllő)	137
Table A.2. Pillai's trace MANOVA of interaction between salinity and temperature on germination performance of 16 maize varieties (2022, MATE-Gödöllő)	137
Table A.3. ANOVA of day, salinity, temperature, and their interaction on germination percentage, radicle length, and plumule length of 16 maize varieties (2022, MATE-Gödöllő)	138
Table A.4. ANOVA of root: shoot ratio and seed vigor index of different salinity treatments, temperatures, and combination of the factors (2022, MATE-Gödöllő)	138
Table A.5. Pillai's trace MANOVA of interaction between variety and temperature on germination performance in 100mM condition (2022, MATE-Gödöllő)	138
Table A.6. ANOVA of germination percentage, radicle and plumule growth of different maize varieties, temperatures, and combination of the factors in salt stress condition (2022, MATE-Gödöllő)	139
Table A.7. Wilks lambda MANOVA of interaction between nitrogen treatment and growing year on maize vegetative growth cultivated under five different nitrogen levels (2023, MATE-Gödöllő)	139
Table A.8. ANOVA of the effect of N treatment and year on maize vegetative growth (2023, MATE-Gödöllő)	139
Table A.9. Wilks lambda MANOVA of interaction between nitrogen treatment and growing year on maize yield cultivated at five different N levels (2023, MATE-Gödöllő)	139
Table A.10. ANOVA on the effect of N treatment and year on maize harvested yield (2023, MATE-Gödöllő)	140
Table A.11. ANOVA of the effect of N treatment and year on maize thousand kernel weight (TKW) (2023, MATE-Gödöllő)	140
Table A.12. Wilks lambda MANOVA of interaction between N treatment and growing year on the grain chemical composition (2023, MATE-Gödöllő)	140
Table A.13. ANOVA on the effect of N treatment and year on grain chemical composition (2023, MATE-Gödöllő)	140
Table A.14. Wilks' lambda MANOVA of interaction of the cover crop species, different stress and week on pots soil moisture and temperature (2024, MATE-Gödöllő)	141
Table A.15. ANOVA on the effect of cover crop, stress, week and their interaction on soil moisture and temperature (2024, MATE-Gödöllő)	141
Table A.16. Wilks' lambda MANOVA of interaction of the cover crop species, different stress and week on maize height and SPAD value (2024, MATE-Gödöllő)	141
Table A.17. ANOVA on the effect of cover crop, stress, week and the interaction on SPAD value and maize height (2024, MATE-Gödöllő)	142
Table A.18. ANOVA on the effect of cover crop and stress treatments on the leaf area (2024, MATE-Gödöllő)	142
Table A.19. Wilks' Lambda MANOVA of interaction between different stress treatments and types of cover crops on the flowering parameters (2024, MATE-Gödöllő)	142
Table A.20. ANOVA on the effects of dependent variables on the flowering parameters (2024, MATE-Gödöllő)	142

Table A.21. Wilks' Lambda MANOVA of interaction between different stress treatments and type of cover crops on the yield parameters (2024, MATE-Gödöllő).....	143
Table A.22. ANOVA on the effects of dependent cover crops and type of stress on ear weight and ear length (2024, MATE-Gödöllő).	143
Table A.23. ANOVA on the effect of abiotic stress type on cover crop dry mass (2024, MATE-Gödöllő).	143
Table A.24. Wilks' Lambda MANOVA of nitrogen levels and type of cover crops on the dependent variables (2023, MATE-Gödöllő).	143
Table A.25. ANOVA on the effect of cover crop, nitrogen and their interaction on maize height, leaves area and SPAD value (2023, MATE-Gödöllő).	143
Table A.26. ANOVA on the effect of nitrogen, cover crop types and their interaction on maize Leave Area Index (LAI) (2023, MATE-Gödöllő).	144
Table A.27. Wilks' Lambda MANOVA of nitrogen level and cover crop type on yield parameters (2023, MATE-Gödöllő).	144
Table A.28. ANOVA on the effect of cover crop, nitrogen and the interaction on ear weight, kernel weight, ear length and grain:cob ratio (2023, MATE-Gödöllő).	144
Table A.29. ANOVA of nitrogen, cover crops and their interaction on maize thousand kernel weight (TKW) (2023, MATE-Gödöllő)	144
Table A.30. Wilks's lambda MANOVA of the cover crop type and nitrogen level on yield chemical composition parameters (2023, MATE-Gödöllő).	144
Table A.31. ANOVA on the effect of cover crop, nitrogen and the interaction on the kernel moisture, oil, protein and starch content (2023, MATE-Gödöllő).	145
Table A.32. Wilks' Lambda MANOVA of cover crop types and nitrogen levels on the dependent variables (2024, MATE-Gödöllő).	145
Table A.33. ANOVA on the effect of cover crop, nitrogen and the interaction on maize height, leaves area and SPAD value (2024, MATE-Gödöllő).	145
Table A.34. ANOVA on nitrogen, cover crop treatments and their interaction on Leave Area Index (LAI) (2024, MATE-Gödöllő).	146
Table A.35. Wilks' Lambda MANOVA cover crop types and nitrogen levels on yield parameters (2024, MATE-Gödöllő).	146
Table A.36. ANOVA on the effect of cover crop, nitrogen and their interaction on ear weight, kernel weight, ear length and grain:cob ratio (2024, MATE-Gödöllő).	146
Table A.37. ANOVA of the effect of nitrogen, cover crops and their interaction on maize thousand kernel weight (TKW) (2024, MATE-Gödöllő).	146
Table A.38. Wilks' Lambda trace MANOVA of type of cover crop and nitrogen levels on yield chemical composition parameters (2024, MATE-Gödöllő).	146
Table A.39. ANOVA on the effect of cover crop, nitrogen and the interaction on the kernel moisture, oil, protein and starch content (2024, MATE-Gödöllő).	147

RESEARCH 1.

Table A.1. Three-way ANOVA for germination percentage, radicle length and plumule length of cover crop germinated under different temperature and salinity levels (2021, MATE-Gödöllő)

Source		Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Cover crop (C)	GP	5.242	2	2.621	15.087	<0.001	0.023
	RL	86.39	2	43.195	89.242	<0.001	0.124
	PL	62.779	2	31.389	37.078	<0.001	0.055
Temperature (T)	GP	4.669	1	4.669	26.872	<0.001	0.021
	RL	189.883	1	189.883	392.305	<0.001	0.237
	PL	103.355	1	103.355	122.085	<0.001	0.088
Salinity (S)	GP	25.639	3	8.546	49.19	<0.001	0.105
	RL	220.462	3	73.487	151.828	<0.001	0.265
	PL	546.424	3	182.141	215.149	<0.001	0.338
C x T	GP	3.348	2	1.674	9.634	<0.001	0.015
	RL	40.483	2	20.242	41.82	<0.001	0.062
	PL	2.409	2	1.205	1.423	0.241	0.002
C x S	GP	2.762	4	0.691	3.975	0.003	0.012
	RL	25.871	4	6.468	13.363	<0.001	0.041
	PL	19.941	4	4.985	5.889	<0.001	0.018
T x S	GP	1.349	2	0.674	3.881	0.021	0.006
	RL	24.362	2	12.181	25.166	<0.001	0.038
	PL	25.168	2	12.584	14.865	<0.001	0.023
C x T x S	GP	7.283	4	1.821	10.48	<0.001	0.032
	RL	8.313	4	2.078	4.294	0.002	0.013
	PL	60.244	4	15.061	17.79	<0.001	0.053
Error	GP	219.436	1263	0.174			
	RL	611.315	1263	0.484			
	PL	1069.233	1263	0.847			
Total	GP	900	1282				
	RL	2378.946	1282				
	PL	4286.48	1282				

RESEARCH 2.

Table A.2. Pillai's trace MANOVA of interaction between salinity and temperature on germination performance of 16 maize varieties (2022, MATE-Gödöllő).

Effect	Value	F	Hypothesis df	Error df	Sig.	Partial η^2
Day	0.824	190.884	9.0	4536.0	<0.001	0.275
Salinity (S)	0.736	1404.791	3.0	1510.0	<0.001	0.736
Temperature (T)	0.697	269.271	6.0	3022.0	<0.001	0.348
S x T	0.443	143.386	6.0	3022.0	<0.001	0.222
D x S	0.515	104.428	9.0	4536.0	<0.001	0.172
D x T	0.825	95.594	18.0	4536.0	<0.001	0.275
D x S x T	0.325	30.642	18.0	4536.0	<0.001	0.108

df degree of freedom, Sig. Significance, $p < 0.05$

Table A.3. ANOVA of day, salinity, temperature, and their interaction on germination percentage, radicle length, and plumule length of 16 maize varieties (2022, MATE-Gödöllő).

Source		Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Day	GP	562241.234	3	187413.745	354.032	<0.001	0.413
	RL	6291.827	3	2097.276	1031.185	<0.001	0.672
	PL	1661.496	3	553.832	1104.078	<0.001	0.687
Salinity (S)	GP	12590.734	1	12590.734	23.784	<0.001	0.015
	RL	6829.679	1	6829.679	3358.006	<0.001	0.690
	PL	804.441	1	804.441	1603.672	<0.001	0.515
Temperature (T)	GP	69261.572	2	34630.786	65.419	<0.001	0.080
	RL	3920.152	2	1960.076	963.727	<0.001	0.560
	PL	1029.280	2	514.640	1025.947	<0.001	0.576
S x T	GP	5896.854	3	1965.618	3.713	0.011	0.007
	RL	2316.944	3	772.315	379.731	<0.001	0.430
	PL	298.467	3	99.489	198.334	<0.001	0.282
D x S	GP	236270.651	6	39378.442	74.387	<0.001	0.228
	RL	1666.794	6	277.799	136.588	<0.001	0.351
	PL	709.255	6	118.209	235.653	<0.001	0.483
D x T	GP	8887.097	2	4443.548	8.394	<0.001	0.011
	RL	1568.162	2	784.081	385.516	<0.001	0.338
	PL	246.300	2	123.150	245.503	<0.001	0.245
D x S x T	GP	30742.747	6	5123.791	9.679	<0.001	0.037
	RL	636.690	6	106.115	52.174	<0.001	0.172
	PL	193.412	6	32.235	64.262	<0.001	0.203
Error	GP	800406.267	1512	529.369			
	RL	3075.181	1512	2.034			
	PL	758.456	1512	0.502			
Total	GP	6342951.994	1536				
	RL	44784.942	1536				
	PL	9423.367	1536				

Table A.4. ANOVA of root: shoot ratio and seed vigor index of different salinity treatments, temperatures, and combination of the factors (2022, MATE-Gödöllő).

		Sum of Squares	df	Mean Square	F	Sig.*	Partial η^2
Salinity	R:S	103.041	1	103.041	206.724	<0.001	0.358
	SVI	60624256.83	1	60624256.83	427.145	<0.001	0.535
Temperature	R:S	76.61	2	38.305	76.849	<0.001	0.293
	SVI	46702743.49	2	23351371.75	164.529	<0.001	0.470
S x T	R:S	8.534	2	4.267	8.561	<0.001	0.044
	SVI	14239692.98	2	7119846.487	50.165	<0.001	0.213
Error	R:S	184.923	371	0.498			
	SVI	52655647.02	371	141928.968			

df degree of freedom, Sig. Significance, *Statistically significant difference: $p < 0.05$

Table A.5. Pillai's trace MANOVA of interaction between variety and temperature on germination performance in 100mM condition (2022, MATE-Gödöllő).

	Value	F	Hypothesis df	Error df	Sig.*	Partial η^2
Variety (V)	0.467	72.904	6.000	1436.000	<0.001	0.233
Temperature (T)	0.272	4.785	45.000	2157.000	<0.001	0.091
V x T	0.211	1.817	90.000	2157.000	<0.001	0.070

df degree of freedom, Sig. significance, *Statistically significant difference: $p < 0.05$

Table A.6. ANOVA of germination percentage, radicle and plumule growth of different maize varieties, temperatures, and combination of the factors in salt stress condition (2022, MATE-Gödöllő).

Source		Sum of Squares	df	Mean Square	F	Sig.*	Partial η^2
Variety (V)	GP	31.938	15	2.129	11.301	<0.001	0.191
	RL	117.213	15	7.814	7.405	<0.001	0.134
	PL	78.116	15	5.208	5.209	<0.001	0.098
Temperature (T)	GP	6.597	2	3.298	17.507	<0.001	0.046
	RL	316.017	2	158.008	149.729	<0.001	0.294
	PL	304.026	2	152.013	152.063	<0.001	0.297
V x T	GP	17.461	30	0.582	3.089	<0.001	0.114
	RL	75.253	30	2.508	2.377	<0.001	0.090
	PL	56.578	30	1.886	1.887	0.003	0.073
Error	GP	135.459	719	0.188			
	RL	758.756	719	1.055			
	PL	718.764	719	1.000			

df degree of freedom, Sig. significance, *Statistically significant difference: $p < 0.05$

RESEARCH 3.

Table A.7. Wilks lambda MANOVA of interaction between nitrogen treatment and growing year on maize vegetative growth cultivated under five different nitrogen levels (2023, MATE-Gödöllő).

Pillai's trace	Value	F	Hypothesis df	Error df	Sig.*	Partial η^2
Nitrogen (N)	0.456	21.926	12.000	762.268	<0.001	0.230
Year (Y)	0.077	1153.669	3.000	288.000	<0.001	0.923
N x Y	0.818	5.024	12.000	762.268	<0.001	0.065

Table A.8. ANOVA of the effect of N treatment and year on maize vegetative growth (2023, MATE-Gödöllő).

Source		Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Nitrogen (N)	Height	35705.537	4	8926.384	37.480	<0.001	0.341
	LA	720426.112	4	180106.528	42.887	<0.001	0.372
	SPAD	610.445	4	152.611	5.145	<0.001	0.066
Year (Y)	Height	622896.333	1	622896.333	2615.432	<0.001	0.900
	LA	1898638.946	1	1898638.946	452.102	<0.001	0.609
	SPAD	13505.888	1	13505.888	455.324	<0.001	0.611
N x Y	Height	3154.097	4	788.524	3.311	0.011	0.044
	LA	53078.647	4	13269.662	3.160	0.015	0.042
	SPAD	1177.801	4	294.450	9.927	<0.001	0.120
Error	Height	69066.959	290	238.162			
	LA	1217877.738	290	4199.578			
	SPAD	8602.016	290	29.662			

Table A.9. Wilks lambda MANOVA of interaction between nitrogen treatment and growing year on maize yield cultivated at five different N levels (2023, MATE-Gödöllő).

Pillai's trace	Value	F	Hypothesis df	Error df	Sig.*	Partial η^2
Nitrogen (N)	0.131	51.811	16.000	877.437	<0.001	0.398
Year (Y)	0.076	870.424b	4.000	287.000	<0.001	0.924
N x Y	0.640	8.639	16.000	877.437	<0.001	0.106

Table A.10. ANOVA on the effect of N treatment and year on maize harvested yield (2023, MATE-Gödöllő).

Source		Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Nitrogen (N)	EL	528.787	4	132.197	129.475	<0.001	0.641
	EW	143118.562	4	35779.641	144.619	<0.001	0.666
	GW	135158.759	4	33789.690	313.049	<0.001	0.812
	GC	762457.280	4	190614.320	107.250	<0.001	0.597
Year (Y)	EL	1621.223	1	1621.223	1587.838	<0.001	0.846
	EW	318430.404	1	318430.404	1287.072	<0.001	0.816
	GW	231953.203	1	231953.203	2148.963	<0.001	0.881
	GC	1218198.963	1	1218198.963	685.425	<0.001	0.703
N x Y	EL	4.460	4	1.115	1.092	0.361	0.015
	EW	16180.597	4	4045.149	16.350	<0.001	0.184
	GW	13447.833	4	3361.958	31.147	<0.001	0.301
	GC	85111.520	4	21277.880	11.972	<0.001	0.142
Error	EL	296.097	290	1.021			
	EW	71747.986	290	247.407			
	GW	31301.810	290	107.937			
	GC	515413.767	290	1777.289			

EL = ear length; EW = ear weight; GW = Grain weight/cob; GC = Grain:cob ratio

Table A.11. ANOVA of the effect of N treatment and year on maize thousand kernel weight (TKW) (2023, MATE-Gödöllő).

Source	Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Year (Y)	39020.1	1	39020.1	2843.278	<0.001	0.983
Nitrogen (N)	32713.52	4	8178.381	595.934	<0.001	0.979
N x Y	7874.656	4	1968.664	143.451	<0.001	0.920
Error	686.182	50	13.724			

Table A.12. Wilks lambda MANOVA of interaction between N treatment and growing year on the grain chemical composition (2023, MATE-Gödöllő).

Effect	Value	F	Hypothesis df	Error df	Sig.	Partial η^2
Nitrogen (N)	0.002	60.357	16.000	144.225	<0.001	0.790
Year (Y)	0.004	2694.354	4.000	47.000	<0.001	0.996
N x Y	0.181	6.747	16.000	144.225	<0.001	0.347

Table A.13 . ANOVA on the effect of N treatment and year on grain chemical composition (2023, MATE-Gödöllő).

Source		Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Nitrogen (N)	Moisture	0.537	4	0.134	1.675	0.171	0.118
	Oil	1.752	4	0.438	601.261	<0.001	0.980
	Protein	12.406	4	3.101	385.561	<0.001	0.969
	Starch	12.803	4	3.201	135.820	<0.001	0.916
Year (Y)	Moisture	8.513	1	8.513	106.143	<0.001	0.680
	Oil	0.128	1	0.128	175.581	<0.001	0.778
	Protein	61.651	1	61.651	7664.247	<0.001	0.994
	Starch	26.534	1	26.534	1125.891	<0.001	0.957
N x Y	Moisture	0.647	4	0.162	2.018	0.106	0.139
	Oil	0.016	4	0.004	5.398	0.001	0.302
	Protein	0.608	4	0.152	18.886	<0.001	0.602
	Starch	1.137	4	0.284	12.065	<0.001	0.491
Error	Moisture	4.010	50	0.080			
	Oil	0.036	50	0.001			
	Protein	0.402	50	0.008			
	Starch	1.178	50	0.024			

RESEARCH 4

Table A.14. Wilks' lambda MANOVA of interaction of the cover crop species, different stress and week on pots soil moisture and temperature (2024, MATE-Gödöllő).

Effect	Value	F	Hypothesis df	Error df	Sig.	Partial η^2
Cover crop (C)	0.242	73.901	6	430	<0.001	0.508
Stress (S)	0.010	968.934	4	430	<0.001	0.900
Week (W)	0.004	611.504	10	430	<0.001	0.934
C x S	0.390	21.567	12	430	<0.001	0.376
C x W	0.232	15.426	30	430	<0.001	0.518
S x W	0.102	45.970	20	430	<0.001	0.681
C x S x W	0.176	9.900	60	430	<0.001	0.580

Table A.15. ANOVA on the effect of cover crop, stress, week and their interaction on soil moisture and temperature (2024, MATE-Gödöllő).

Source		Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Cover crop (C)	Moisture	1900.750	3	633.583	148.593	<0.001	0.674
	Temperature	22.560	3	7.520	31.017	<0.001	0.301
Stress (S)	Moisture	88936.750	2	44468.375	10429.065	<0.001	0.990
	Temperature	39.451	2	19.725	81.358	<0.001	0.430
Week (W)	Moisture	12473.458	5	2494.692	585.074	<0.001	0.931
	Temperature	949.830	5	189.966	783.514	<0.001	0.948
C x S	Moisture	1169.917	6	194.986	45.730	<0.001	0.560
	Temperature	9.621	6	1.603	6.613	<0.001	0.155
C x W	Moisture	1039.125	15	69.275	16.247	<0.001	0.530
	Temperature	64.634	15	4.309	17.772	<0.001	0.552
S x W	Moisture	5110.667	10	511.067	119.859	<0.001	0.847
	Temperature	26.398	10	2.640	10.888	<0.001	0.335
C x S x W	Moisture	1165.833	30	38.861	9.114	<0.001	0.559
	Temperature	79.942	30	2.665	10.991	<0.001	0.604
Error	Moisture	921.000	216	4.264			
	Temperature	52.370	216	0.242			

Table A.16. Wilks' lambda MANOVA of interaction of the cover crop species, different stress and week on maize height and SPAD value (2024, MATE-Gödöllő).

Effect	Value	F	Hypothesis df	Error df	Sig.	Partial η^2
Cover crop (C)	0.567	73.340	6	1342	<0.001	0.247
Stress (S)	0.349	232.464	4	1342	<0.001	0.409
Week (W)	0.037	399.615	14	1342	<0.001	0.807
C x S	0.861	8.665	12	1342	<0.001	0.072
C x W	0.718	5.760	42	1342	<0.001	0.153
S x W	0.326	36.066	28	1342	<0.001	0.429
C x S x W	0.823	1.631	84	1342	<0.001	0.093

Table A.17. ANOVA on the effect of cover crop, stress, week and the interaction on SPAD value and maize height (2024, MATE-Gödöllő).

Source		Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Cover crop (C)	SPAD	9866.463	3	3288.821	128.518	<0.001	0.365
	Height	16299.832	3	5433.277	43.664	<0.001	0.163
Stress (S)	SPAD	10825.731	2	5412.866	211.520	<0.001	0.386
	Height	106470.152	2	53235.076	427.816	<0.001	0.560
Week (W)	SPAD	19213.837	7	2744.834	107.260	<0.001	0.528
	Height	1605958.342	7	229422.620	1843.722	<0.001	0.951
C x S	SPAD	2097.998	6	349.666	13.664	<0.001	0.109
	Height	3166.470	6	527.745	4.241	<0.001	0.036
C x W	SPAD	3005.181	21	143.104	5.592	<0.001	0.149
	Height	16527.869	21	787.041	6.325	<0.001	0.165
T x W	SPAD	6267.305	14	447.665	17.493	<0.001	0.267
	Height	110307.690	14	7879.121	63.319	<0.001	0.569
C x S x W	SPAD	2245.635	42	53.468	2.089	<0.001	0.116
	Height	6228.931	42	148.308	1.192	0.193	0.069
Error	SPAD	17196.719	672	25.590			
	Height	83619.985	672	124.435			

Table A.18. ANOVA on the effect of cover crop and stress treatments on the leaf area (2024, MATE-Gödöllő).

Source	Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Cover crop (C)	131503.26	3	43834.42	52.419	<0.001	0.652
Irrigation (I)	82929.078	2	41464.539	49.585	<0.001	0.541
C x I	19436.392	6	3239.399	3.874	0.002	0.217
Error	70243.771	84	836.235			
Total	28786802	96				

Table A.19. Wilks' Lambda MANOVA of interaction between different stress treatments and types of cover crops on the flowering parameters (2024, MATE-Gödöllő).

Effect	Value	F	Hypothesis df	Error df	Sig.	Partial η^2
Stress (S)	0.034	182.426	4.000	166.000	<0.001	0.815
Cover crop (C)	0.242	28.535	6.000	166.000	<0.001	0.508
C x S	0.438	7.071	12.000	166.000	<0.001	0.338

Table A.20. ANOVA on the effects of dependent variables on the flowering parameters (2024, MATE-Gödöllő).

Source		Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Stress (S)	Tasseling	7387.771	2	3693.885	572.220	<0.001	0.932
	Silking	14045.271	2	7022.635	1063.604	<0.001	0.962
	ASI	1081.188	2	540.594	227.476	<0.001	0.844
Cover crop (C)	Tasseling	790.125	3	263.375	40.799	<0.001	0.593
	Silking	1586.198	3	528.733	80.079	<0.001	0.741
	ASI	148.281	3	49.427	20.798	<0.001	0.426
C x S	Tasseling	515.813	6	85.969	13.317	<0.001	0.488
	Silking	321.146	6	53.524	8.106	<0.001	0.367
	ASI	61.563	6	10.260	4.317	<0.001	0.236
Error	Tasseling	542.250	84	6.455			
	Silking	554.625	84	6.603			
	ASI	199.625	84	2.376			

ASI = Anthesis- silking index

Table A.21. Wilks' Lambda MANOVA of interaction between different stress treatments and type of cover crops on the yield parameters (2024, MATE-Gödöllő).

Effect	Value	F	Hypothesis df	Error df	Sig.	Partial η^2
Stress (S)	0.200	16.093	4.000	52.000	<0.001	0.553
Cover crop (C)	0.416	4.763	6.000	52.000	0.001	0.355
S x C	0.539	2.356	8.000	52.000	0.030	0.266

Table A.22. ANOVA on the effects of dependent cover crops and type of stress on ear weight and ear length (2024, MATE-Gödöllő).

Source		Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Stress (S)	EW	683.658	3	227.886	8.362	<0.001	0.482
	EL	58.875	3	19.625	9.268	<0.001	0.507
Cover crop (C)	EW	1878.535	2	939.267	34.463	<0.001	0.719
	EL	132.436	2	66.218	31.271	<0.001	0.698
S x C	EW	45.178	4	11.294	0.414	0.797	0.058
	EL	15.175	4	3.794	1.792	0.160	0.210
Error	EW	735.863	27	27.254			
	EL	57.174	27	2.118			

Table A.23. ANOVA on the effect of abiotic stress type on cover crop dry mass (2024, MATE-Gödöllő).

Cover crop	Source	Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Alfalfa	Irrigation	1371.440	2	685.720	405.485	<0.001	0.989
	Error	15.220	9	1.691			
	Total	32966.940	12				
Bean	Irrigation	4641.307	2	2320.653	1427.606	<0.001	0.997
	Error	14.630	9	1.626			
	Total	30957.540	12				

RESEARCH 5

Table A.24. Wilks' Lambda MANOVA of nitrogen levels and type of cover crops on the dependent variables (2023, MATE-Gödöllő).

Effect	Value	F	Hypothesis df	Error df	Sig.	Partial η^2
Nitrogen (N)	0.176	118.090	12.000	1529.536	<0.001	0.439
Cover crop (C)	0.614	34.631	9.000	1406.851	<0.001	0.150
N x C	0.871	2.273	36.000	1708.493	<0.001	0.045

Table A.25. ANOVA on the effect of cover crop, nitrogen and their interaction on maize height, leaves area and SPAD value (2023, MATE-Gödöllő).

Source		Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Nitrogen (N)	Height	47626.283	4	11906.571	229.916	<0.001	0.613
	LA	1648204.431	4	412051.108	101.388	<0.001	0.411
	SPAD	23925.494	4	5981.373	352.154	<0.001	0.708
Cover crop (C)	Height	854.058	3	284.686	5.497	0.001	0.028
	LA	183807.316	3	61269.105	15.076	<0.001	0.072
	SPAD	4964.825	3	1654.942	97.435	<0.001	0.335
N x C	Height	2224.383	12	185.365	3.579	<0.001	0.069
	LA	57126.860	12	4760.572	1.171	0.300	0.024
	SPAD	439.813	12	36.651	2.158	0.012	0.043
Error	Height	30036.233	580	51.787			
	LA	2357179.471	580	4064.103			
	SPAD	9851.372	580	16.985			

Table A.26. ANOVA on the effect of nitrogen, cover crop types and their interaction on maize Leave Area Index (LAI) (2023, MATE-Gödöllő).

Source	Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Nitrogen (N)	21.170	4	5.292	356.555	<0.001	0.960
Cover crop (C)	1.892	3	0.631	42.479	<0.001	0.680
N x C	0.496	12	0.041	2.787	0.004	0.358
Error	0.891	60	0.015			
Total	562.859	80				

Table A.27. Wilks' Lambda MANOVA of nitrogen level and cover crop type on yield parameters (2023, MATE-Gödöllő).

Effect	Value	F	Hypothesis df	Error df	Sig.	Partial η^2
Nitrogen (N)	0.195	77.983	16.000	1763.402	<0.001	0.335
Cover crop (C)	0.717	17.060	12.000	1526.890	<0.001	0.105
N x C	0.699	4.508	48.000	2224.703	<0.001	0.086

Table A.28. ANOVA on the effect of cover crop, nitrogen and the interaction on ear weight, kernel weight, ear length and grain:cob ratio (2023, MATE-Gödöllő).

Source		Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Nitrogen (N)	EW	520152.127	4	130038.032	240.980	<0.001	0.624
	KW	290123.283	4	72530.821	180.504	<0.001	0.555
	EL	1661.642	4	415.411	205.031	<0.001	0.586
	G:C ratio	2181988.200	4	545497.050	60.042	<0.001	0.293
Cover crop (C)	EW	30129.473	3	10043.158	18.611	<0.001	0.088
	KW	30677.907	3	10225.969	25.449	<0.001	0.116
	EL	257.576	3	85.859	42.377	<0.001	0.180
	G:C ratio	283003.885	3	94334.628	10.383	<0.001	0.051
N x C	EW	22081.260	12	1840.105	3.410	<0.001	0.066
	KW	15674.943	12	1306.245	3.251	<0.001	0.063
	EL	39.957	12	3.330	1.643	0.076	0.033
	G:C ratio	124441.507	12	10370.126	1.141	0.323	0.023
Error	EW	312980.733	580	539.622			
	KW	233057.867	580	401.824			
	EL	1175.133	580	2.026			
	G:C ratio	5269457.367	580	9085.271			

EW= ear weight, KW= kernel weight/ear, EL= ear length, G:C ratio= grain:cob ratio

Table A.29. ANOVA of nitrogen, cover crops and their interaction on maize thousand kernel weight (TKW) (2023, MATE-Gödöllő)

TKW	Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Nitrogen (N)	24077.550	4	6019.388	701.016	<0.001	0.966
Cover crop (C)	4250.067	3	1416.689	164.987	<0.001	0.832
N x C	7733.183	12	644.432	75.050	<0.001	0.900
Error	858.667	100	8.587			
Total	9339934.000	120				

Table A.30. Wilks's lambda MANOVA of the cover crop type and nitrogen level on yield chemical composition parameters (2023, MATE-Gödöllő).

Effect	Value	F	Hypothesis df	Error df	Sig.	Partial η^2
Cover crop (C)	0.045	47.491	12.0	256.929	<0.001	0.643
Nitrogen (N)	0.012	61.234	16.0	296.978	<0.001	0.672
C x N	0.063	8.210	48.0	375.693	<0.001	0.499

Table A.31. ANOVA on the effect of cover crop, nitrogen and the interaction on the kernel moisture, oil, protein and starch content (2023, MATE-Gödöllő).

Source		Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Cover crop (C)	Moisture	0.283	3	0.094	3.159	0.028	0.087
	Oil	0.228	3	0.076	30.658	<0.001	0.479
	Protein	8.552	3	2.851	255.556	<0.001	0.885
	Starch	18.272	3	6.091	81.297	<0.001	0.709
Nitrogen (N)	Moisture	6.278	4	1.569	52.578	<0.001	0.678
	Oil	0.360	4	0.090	36.217	<0.001	0.592
	Protein	38.815	4	9.704	869.943	<0.001	0.972
	Starch	13.779	4	3.445	45.980	<0.001	0.648
C x N	Moisture	1.004	12	0.084	2.803	0.002	0.252
	Oil	0.104	12	0.009	3.508	<0.001	0.296
	Protein	6.387	12	0.532	47.713	<0.001	0.851
	Starch	3.628	12	0.302	4.036	<0.001	0.326
Error	Moisture	2.985	100	0.030			
	Oil	0.248	100	0.002			
	Protein	1.115	100	0.011			
	Starch	7.492	100	0.075			
Total	Moisture	15444.290	120				
	Oil	1570.863	120				
	Protein	2720.259	120				
	Starch	609643.990	120				

RESEARCH 6

Table A.32. Wilks' Lambda MANOVA of cover crop types and nitrogen levels on the dependent variables (2024, MATE-Gödöllő).

Effect	Value	F	Hypothesis df	Error df	Sig.	Partial η^2
Cover crop (C)	0.182	76.339	9	676.73	<0.001	0.434
Nitrogen (N)	0.153	63.275	12	735.81	<0.001	0.465
C x N	0.554	5.048	36	822.11	<0.001	0.179

Table A.33. ANOVA on the effect of cover crop, nitrogen and the interaction on maize height, leaves area and SPAD value (2024, MATE-Gödöllő).

Source		Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Cover crop (C)	Height	89476.160	3	29825.387	126.084	<0.001	0.575
	SPAD	6051.563	3	2017.188	124.091	<0.001	0.571
	LA	1157587.423	3	385862.474	137.274	<0.001	0.595
Nitrogen (N)	Height	129900.613	4	32475.153	137.286	<0.001	0.662
	SPAD	10674.440	4	2668.610	164.165	<0.001	0.701
	LA	1027746.793	4	256936.698	91.408	<0.001	0.566
C x N	Height	15285.307	12	1273.776	5.385	<0.001	0.188
	SPAD	1432.716	12	119.393	7.345	<0.001	0.239
	LA	92170.900	12	7680.908	2.733	0.002	0.105
Error	Height	66234.400	280	236.551			
	SPAD	4551.595	280	16.256			
	LA	787049.508	280	2810.891			

Table A.34. ANOVA on nitrogen, cover crop treatments and their interaction on Leave Area Index (LAI) (2024, MATE-Gödöllő).

Source	Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Cover crop (C)	11.160	3	3.720	173.197	<0.001	0.896
Nitrogen (N)	10.481	4	2.620	121.986	<0.001	0.890
C x N	0.742	12	0.062	2.879	0.003	0.365
Error	1.289	60	0.021			
Total	451.114	80				

Table A.35. Wilks' Lambda MANOVA cover crop types and nitrogen levels on yield parameters (2024, MATE-Gödöllő).

Effect	Value	F	Hypothesis df	Error df	Sig.	Partial η^2
Cover crop (C)	0.075	101.832	12.000	733.165	<0.001	0.579
Nitrogen (N)	0.119	53.311	16.000	846.887	<0.001	0.413
C x N	0.563	3.583	48.000	1069.072	<0.001	0.134

Table A.36. ANOVA on the effect of cover crop, nitrogen and their interaction on ear weight, kernel weight, ear length and grain:cob ratio (2024, MATE-Gödöllő).

Source		Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Cover crop (C)	EW	423454.746	3	141151.582	522.028	<0.001	0.848
	EL	1091.808	3	363.936	497.851	<0.001	0.842
	KW	363933.326	3	121311.109	521.414	<0.001	0.848
	G:C	1845825.120	3	615275.040	643.868	<0.001	0.873
Nitrogen (N)	EW	237307.926	4	59326.982	219.412	<0.001	0.758
	EL	661.026	4	165.257	226.065	<0.001	0.764
	KW	183907.912	4	45976.978	197.616	<0.001	0.738
	G:C	1122947.947	4	280736.987	293.783	<0.001	0.808
C x N	EW	11880.436	12	990.036	3.662	<0.001	0.136
	EL	27.905	12	2.325	3.181	<0.001	0.120
	KW	9382.684	12	781.890	3.361	<0.001	0.126
	G:C	38656.747	12	3221.396	3.371	<0.001	0.126
Error	EW	75709.445	280	270.391			
	EL	204.684	280	0.731			
	KW	65144.244	280	232.658			
	G:C	267565.867	280	955.592			

EW: ear weight, KW: kernel weight/ear, EL: ear length, G:C ratio: grain:cob ratio

Table A.37. ANOVA of the effect of nitrogen, cover crops and their interaction on maize thousand kernel weight (TKW) (2024, MATE-Gödöllő).

Source	Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Cover crop (C)	116528.069	3	38842.690	11424.376	<0.001	0.997
Nitrogen (N)	49123.498	4	12280.874	3612.040	<0.001	0.993
C x N	1605.471	12	133.789	39.350	<0.001	0.825
Error	339.998	100	3.400			
Total	6966638.350	120				

Table A.38. Wilks' Lambda trace MANOVA of type of cover crop and nitrogen levels on yield chemical composition parameters (2024, MATE-Gödöllő).

Effect	Value	F	Hypothesis df	Error df	Sig.	Partial η^2
Cover crop (C)	0.0002	512.190	12.0	256.929	<0.001	0.941
Nitrogen (N)	0.0003	236.592	16.0	296.978	<0.001	0.865
C x N	0.001	40.423	48.0	375.693	<0.001	0.826

Table A.39. ANOVA on the effect of cover crop, nitrogen and the interaction on the kernel moisture, oil, protein and starch content (2024, MATE-Gödöllő).

Source		Sum of Squares	df	Mean Square	F	Sig.	Partial η^2
Cover crop (C)	Moisture	6.455	3	2.152	114.754	<0.001	0.775
	Oil	0.513	3	0.171	101.611	<0.001	0.753
	Protein	9.681	3	3.227	2800.290	<0.001	0.988
	Starch	82.108	3	27.369	652.429	<0.001	0.951
Nitrogen (N)	Moisture	26.324	4	6.581	350.993	<0.001	0.934
	Oil	0.620	4	0.155	91.995	<0.001	0.786
	Protein	11.586	4	2.896	2513.591	<0.001	0.990
	Starch	20.227	4	5.057	120.540	<0.001	0.828
C x N	Moisture	11.996	12	1.000	53.313	<0.001	0.865
	Oil	0.100	12	0.008	4.946	<0.001	0.372
	Protein	7.147	12	0.596	516.873	<0.001	0.984
	Starch	2.448	12	0.204	4.863	<0.001	0.369
Error	Moisture	1.875	100	0.019			
	Oil	0.168	100	0.002			
	Protein	0.115	100	0.001			
	Starch	4.195	100	0.042			

LIST OF PUBLICATIONS OF THE AUTHOR IN THE RESEARCH FIELD

1. **Khalid, N.**, Tarnawa, Á., Balla, I., Omar, S., Abd Ghani, R., Jolánkai, M. and Kende, Z., (2023). Combination Effect of Temperature and Salinity Stress on Germination of Different Maize (*Zea mays* L.) Varieties. *Agriculture*, 13(10), p.1932. <https://doi.org/10.3390/agriculture13091836>.
2. **Khalid, N.**, Sghaier, A.H., Jolánkai, M. and Tarnawa, Á. (2023). The role of temperature on the germination activity of leguminous crops exposed to saline conditions. *Időjárás*, 127(2), pp.253–265. <https://doi.org/10.28974/idojaras.2023.2.6>.
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Conference Presentation

1. **Khalid, N.**, Sghaier, A.H., Jolánkai, M., Balla, I. (2023) The role of temperature on the germination activity of leguminous crops exposed to saline conditions. Oral presented in *The Youth Science Forum*, 8 June 2023, Keszthely, Hungary.
2. Omar, S., Abd Ghani, R., **Khalid, N.**, & Jolánkai, M. (2023). Evaluation of maize inbred lines and hybrids for agronomic characteristics, yield, and grain quality. Oral presented in *Youth Science Forum*, 8 June 2023, Keszthely, Hungary.

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1. Abd Ghani, R., Omar, S., Jolánkai, M., Tarnawa, Á., Kende, Z., **Khalid, N.**, Gyuricza, C. and Kassai, M.K. (2023). Soilless Culture Applications for Early Development of Soybean

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1. Sghaier, A.H., **Khalid, N.**, Omar, S., Varga, A., and Kende, Z. Methodological approaches to the germination of sunflower and oilseed rape in vitro. Oral presented in Youth Science Forum, 8 June 2023, Keszthely, Hungary.
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ACKNOWLEDGEMENT

First and foremost, praises to Allah, the Almighty, for giving me strength and blessings throughout my study, especially during the field research and writing process.

I wish to express my deepest and most sincere gratitude to my supervisor, Dr. Ákos Tarnawa, for his invaluable guidance and unwavering support throughout my PhD journey. I am also profoundly grateful for his dedicated efforts, particularly in the fieldwork, and for his guidance during the writing process. May you be blessed with enduring happiness and strength.

I extend my heartfelt thanks to all the members of the Crop Production Institute at the Hungarian University of Agricultural and Life Sciences, Gödöllő, including Professor Jolánkai Márton and Dr. Zoltán Kende, for their advices and invaluable assistance, as well as the lab assistants, Ms. Szikoráné Nagy Diána and Ms. Kakucska Csilla, for their dedicated support in the laboratory. Special thanks go to my friends and colleagues who shared this journey with me, particularly Suhana Omar, Rosnani Abd Ghani, Asma Haj Sghaier, and Huynh Ahn Kiet. I am equally grateful to the entire community at the Hungarian University of Agricultural and Life Sciences, including my lecturers and the Doctoral and Habilitation Centre.

I also wish to express my heartfelt appreciation for the boundless love and encouragement of my family and friends in Malaysia and Hungary, especially Zuraini Jamil Osman, Nor Amira Abdul Rahman, and Siti Halijah Dzahir. My PhD journey would not have been possible without the unwavering support, love, and sacrifices from Beáta Mucsi, and the entire Mucsi family.

Finally, I am deeply thankful to the Hungarian government for granting me the Stipendium Hungaricum Scholarship, which enabled me to pursue my studies in this remarkable country and at this outstanding university.