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**ADAPTING MAIZE BREEDING TO ORGANIC AGRICULTURE**

Thesis to obtain the Degree of  
Master of Science

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## PREFACE

This thesis takes the experiences learnt after including organic trials in the maize Value for Cultivation and Use (VCU) Test in Germany and apply them to conventional breeding programs, which try to meet the needs of the organic sector, in order to adapt their breeding outlines for selection of organic maize varieties. The idea of this project appeared after receiving a data set of grain and silage maize VCU Test from the company KWS SAAT AG, with help from both of my supervisors, plus my continuous and incessant interest in organic agriculture. I wrote this thesis as part of my MSc. Plant Sciences Programme. Its main purpose is to learn whether testing conventional bred varieties under organic conditions is appropriate to designate varieties for organic farming. I choose and feel comfortable with this subject since it offered me the opportunity to research about the two aspects I am most interested about: plant breeding and organic farming. Additionally, it also offered me the opportunity to study and learn the varietal testing for maize in Europe and its position towards organic agriculture.

After one complete year (2008/09) inside a sustainable community in Winsconsin, USA working for an organic plant breeding program, I became fascinated about organic and biodynamic agriculture and their ways of assessing food production; this is by far my main source of inspiration. For the elaboration of this thesis project most of the resources were offered by Wageningen University and Research Center (WUR), where I was able to review different literature about comparative studies between conventional and organic production in different crops, VCU Test and breeding; also the company KWS SAAT AG in Einbeck offered useful and appreciable resources while contributing with the data for maize VCU Test. Moreover, the company offered us a visit tour to their installations and field plots in order to have a better insight about its activities and goals. In order to develop the research, analyze the data and write this report it took a total of 7 months from August 2011 to February 2012.

Once finished this work, I feel academically satisfied, it makes me evoke the propose of my MSc. studies and the research in organic agriculture, moreover during this period I learnt new methodologies and statistical analysis. In order to read this document a basic knowledge in plant breeding, varietal testing and statistics is needed. During the writing process, different literature was reviewed, consultations with maize researchers and experts was made via mail or in person, and the data set were analyzed with different methodologies in order to select at the end the most suitable to be presented.

Finally, to accomplish this challenge I have to thank and express my gratitude to the people who with a lot of generosity, gave me their time, encouraged me in the process of writing this thesis, and support with this study: my supervisors Edith Lammerts van Bueren and Oene Dolstra from WUR who orientated me during the whole thesis period, Walter Schmidt and Henriette Burger from KWS SAAT AG who offered the maize data set and took the time to show us the company in a visit to Einbeck during the last summer, Walter Goldstein from Mandaamin Institute and Nick van Eekeren from Louis Bolk Institute for sharing their opinions without any problem when they were consulted.

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## ABSTRACT

Organic agriculture has gained significance since the 1990s and nowadays exist a growing demand for varieties specifically adapted to organic or low input conditions. However, breeding programs specific for organic farming would entail major time, resources, and labor investment and to breed organic varieties would require specific testing conditions and different breeding strategies, In Germany, the Federal Office of Plant Varieties decided to include organic trials in the variety testing of conventional maize in order to recommend which of these varieties are adapted to the organic farming. In the present study, data sets from grain and silage maize VCU Test, including conventional bred varieties tested in parallel under both conventional and organic conditions, were analysed to obtain quantitative genetic parameters, correlations, stability performances and similarities in cluster analysis for 3 traits for grain maize: grain fresh matter (GFM), grain yield, grain dry matter content (GDCn1); and 3 traits in silage maize: total fresh matter (TFM), silage yield, total dry matter content (TDCn2). Results showed in general high repeatability (with lower values under organic conditions) and very high correlation between genotypic means of both farming systems for 3 of the traits: GDCn1, TFM and TDCn2. Trait silage yield showed high repeatability under conventional conditions and much lower under organic conditions and high correlation between genotypic means of both farming systems. However GFM and grain yield registered low repeatability in both farming conditions and medium to low genotypic correlation between both farming systems, respectively. These results combined with literature review and consultations with maize experts from USA, Germany and the Netherlands were used to assess whether it is enough to rely on the conventionally selected maize varieties and varietal testing under organic conditions to supply the seed for organic agriculture. It was concluded that for traits with high correlated response between both farming systems, testing and/or breeding under conventional, organic or both farming conditions produce reliable and predictable performances between both farming systems. For silage yield, the organic agricultural management is relevant when determining its value. For traits with low correlated response between both farming systems, the parallel testing under conventional and organic environments and the inclusion of standard varieties that perform adequately in low-input conditions, are convenient below the cross-over point of the GxE interaction. However, the evaluation of only conventionally bred genotypes, and the lack of characteristics required to cope organic environments in the evaluation, are limiting factor for the accurate recommendation of organic varieties. Finally, based on the lessons learnt from these experiences, a breeding procedure for organic maize varieties was designed by adapting a conventional breeding program. For such breeding scheme it is necessary to include and characterize organic environments during the stage of general and specific combinatory ability, and evaluate key traits required for the crop to deal with organic environments, in order to identify genotypes adapted to organic farming.

## I. INTRODUCTION

At the end of century 20<sup>th</sup> an increasing number of European farmers became interested in low-input farming systems, due mainly to a rising environmental concern. Organic agriculture has gained significance since the 1990s and nowadays there is a growing demand for varieties specifically adapted to organic or low input conditions (Lorenzana & Bernardo, 2008; Burger *et al.*, 2008; Löschenberger *et al.*, 2008; Messmer *et al.*, 2010). Since varieties used to be bred to give high yields in conventional systems, they may not be well-adapted to organic conditions (Lammerts van Bueren *et al.*, 2003; Lammerts van Bueren & Verhoog, 2006); therefore, there might be a need of cultivars specifically bred for organic production (Colley & Dillon, 2004; Lammerts van Bueren, 2006; Wolfe *et al.*, 2008). In June 1991, the European Commission issued the Council Regulation (EEC) No 2092/91 on organic production of agricultural products and indications referring thereto on agricultural products and foodstuffs. The legislation obliges the organic farmers to use organically produced seeds and vegetative multiplication material (Löschenberger *et al.*, 2008). Nevertheless, nowadays, organic farmers frequently employ conventional varieties in their production systems (Lammerts van Bueren & Verhoog, 2006; Löschenberger *et al.*, 2008; Wolfe *et al.*, 2008). Varieties cultivated in organic farming can basically originate from three different types of breeding approaches (Wolfe *et al.*, 2008):

- a) Breeding programs for conventional agriculture (BFCA): the entire breeding process takes place under conventional conditions and the farmers select the best varieties which perform good enough under organic agriculture.
- b) Breeding program for organic agriculture (BFOA): the breeding process is initially done under conventional conditions and thereafter advanced promising lines are tested and selected in organic environments.
- c) Organic breeding programs (OPB): the entire breeding process takes place under organic conditions, additionally the selection and propagation techniques fulfil the organic principles.

However, breeding programs specific for organic farming would entail time, resources, and labour investment (Lorenzana & Bernardo, 2008) and to breed organic varieties would require specific testing conditions and different breeding strategies (Burger *et al.*, 2008). For these reasons advantages of selection under organic conditions need to be determined (Lammerts van Bueren & Verhoog, 2006), especially since the organic market is still unprofitable for large breeding companies and capital for public breeding is limited (Lorenzana & Bernardo, 2008).

For varietal traits and characterization in EU and many countries around the world the International Union for the Protection of New Varieties of Plants (UPOV) developed its own set of regulations through the Community Plant Variety Office (CPVO). In the countries under UPOV regulation trade is only permitted for registered varieties, for the EU it means varieties registered in the Common EU Catalogue of Varieties based on the National List of Varieties of each member state. To be admitted a variety should prove to have distinctness, uniformity and stability (DUS) and it is evaluated the DUS field tests during 2 years for several characteristics by crop-specific DUS protocols which can be obtained from the Community Plant Variety Office (CPVO) website (<http://www.cpvo.europa.eu>). Another element of the EU legislation for registration of new varieties for certain arable crops, such as maize, is the Value of Cultivation and Use (VCU) test. The aim of VCU testing is to only admit into the market new varieties that are a clear improvement compared with the existing varieties. This test is normally run under conventional conditions and evaluates production and processing performance of the candidate variety. However, such test may not pay attention to characteristics that may be relevant to organic farming systems and there is a risk to exclude varieties suitable for the organic sector due to a lack of information. For that reason, in order to assess if new varieties that are suitable for organic farming, some EU member states study the necessity to adapt the VCU protocol with requirements specifically relevant for the organic sector. In general, for cereals there are many initiatives and projects in this respect and valuable guidelines for organic variety testing have been produced. On the other hand, arable and fodder crops mixtures have no research or guidelines for organic testing available (Micheloni & Plakolm, 2007; Osman *et al.*, 2008a).

## 1.1. Maize Breeding

Maize breeding has features that are quite different from those used in self-pollinating cereal crops, being one of the most successful examples of a man-made crop with tremendous breeding achievements in the last century in particular. Breeding techniques for both self- and cross-pollinated crops can be applied in maize allowing handling large sample sizes easily and application of a diversity of breeding methods. Since maize is used for economically important uses, such as feed, fiber, fuel, and food; breeding methods have evolved to rise the effectiveness and efficiency of selection for several quantitative traits in multiple stages and environments. While the basic breeding methods for development of maize were described in 1910, significant contributions have been made to optimize their use. For instance, information in maize genetics grew in an extensive way during the past 50 years until the unravelling of the genome sequence in 2008. In the recent years, the large-scale use of doubled haploids and application of genome-wide selection enabled by the enormous and fast developments in molecular genetics and the generation of genotypic data at a cheaper cost have drastically changed how maize breeders design and conduct breeding programs. On one hand quantitative genetic studies are collected on the phenotypes of families and progenies replicated within and across environments. On the other hand molecular geneticists study gene and allele effects at the level of DNA. However, both approaches are converging, with the constant advancements in molecular genetics and the need for phenotyping (Hallauer *et al*, 2010); and generating an integrated activity, where molecular geneticist assist with respect to choice of parents and support the selection of DH lines of interest (Dolstra, 2012 personal communication).

Grain maize yields have increased significantly since the past century in the United States (1930s), and other parts of the world (1950s and 1960s), especially in countries where this crop is grown in a commercial way. The main factors for improving the yield were both cultural practices (use of synthetic nitrogen fertilizers, increasing of plant density, weed and pest control, timeliness of planting, and improved efficiency of harvest equipment), and the plant breeding process (farmer breeding developed thousands of landraces and open pollinated cultivars, the production of hybrids from crosses of inbred lines taking advantage of the heterosis, the production of synthetics by intercrossing the best inbred lines, and the establishing of recurrent selection in breeding programs). Consequently, breeders had consistently increased the hybrid yield ability since 1935 (Duvick, 2005). According to Duvick (2005), the main trait changes that support the genetic yield gains in hybrid maize in the US are listed in two categories:

- A. Traits that promote efficiently grain-yield:
  - a) Leaf angle significantly more upright.
  - b) Tassel size markedly reduced.
  - c) Longer period of grain fill but faster dry down
  - d) Kernel weight is greater (except under drought stress at terminal periods of grain fill)
  - e) Lower percentage grain protein.
  - f) Higher percentage grain starch.
  - g) More responsive to favourable environments (they make more efficient use of abundant inputs), although results are not consistent in this regard.
  
- B. Traits that increase tolerance to biotic and abiotic stress:
  - a) Grain yield increased in linear fashion, increases are greatest at high plant density and exhibited in all environments.
  - b) Leaf rolling during drought stress is increased.
  - c) Stay-green markedly improved.
  - d) Anthesis-silking interval is shortened, especially under drought or high plant density conditions.
  - e) Increased resistance to sterility under drought at flowering time or higher plant density conditions.
  - f) Higher harvest index under biotic stresses that induce sterility.
  - g) Linear improvements in resistance to root lodging.
  - h) Linear improvements in resistance to stalk lodging.
  - i) Linear improvement in yield in seasons with high temperature during the growing stage.
  - j) Linear improvement in yield in seasons with low temperature during the growing stage.
  - k) More drought tolerance.
  - l) More tolerant of excessive soil moisture (water-logged soils).

- m) More tolerant to soil nitrogen deficiency.
- n) More tolerant to unspecified abiotic stresses ("low-yield" sites).
- o) More tolerant to second-generation European corn borer (*Ostrinia nubilalis*), and, recently, transgenic hybrids have expressed a sharply increased level of resistance to both generations of European corn borer and (separately) to two species of rootworm.
- p) New kinds of disease resistance, however the contributions to the yield of sequential changes in disease resistance are not documented.
- q) More tolerant to the stresses of higher plant density.
- r) Increased tolerance to specific herbicides
- s) More efficient photosynthesis under stress, and improved capacity to recover the photosynthetic rate after stress.
- t) More efficient canopy gas exchange, stem water potential, transpiration, and respiration under water stress.
- u) Canopy temperature under drought stress is decreased.

Despite that morphological and physiological changes are relevant for yield enhancement in maize, it is clear that, by far the majority of the improvements in maize breeding are about stress tolerance, indicating that this development has been the primary driving force for the higher yield ability of the modern hybrids (Duvick, 2005). In the European situation, breeding achievements are at least as or even more impressive than those in US, especially considering the climatic and environmental differences (Dolstra, 2012 personal communication); a clear example is the introduction of the first hybrids more tolerant to low temperature, especially in the cooler conditions of northern Europe. At the beginning, the usage of maize in Europe was for human feeding, though the value of forage maize was quickly recognized since the exploration and conquest of America, or shortly after its introduction in Europe. Maize turned certainly into fodder crop due to the possibilities of chemical weed control, the mechanization of plant cropping, the easy and reliable conservation method, and the very efficient management of high quality seed production. Nevertheless, maize breeding efforts and financial inputs were only focused on the improvements of grain traits over a long time in USA, France and Germany; though, the main objective in the Netherlands was the breeding for fodder maize able to yield a large quantity of high-energy silage maize. Breeding for forage maize plant started actually in Europe during 1980s and most of the available European fodder maize are still greatly based on grain maize germplasm. For this reason, genetic resources allowing further quality improvements in silage maize need to be re-discovered (Barrière *et al.*, 2006). According to Barrière *et al.* (2006), the main traits improved by maize breeding in Europe are:

- a) Grain yield.
- b) Cold tolerance: better plant growth, better root growth, better tolerance to chilling injury grain filling and ripening, and early vigour during spring.
- c) Stay-green: higher leaf area duration, which allow a longer photosynthesis duration.
- d) More erected leaves, which increase the interception of photoactive radiations.
- e) Later flowering
- f) Higher leaf number and leaf area index, which allows faster grain filling and ripening.
- g) Higher stress tolerance.
- h) Better recovery from stress.
- i) Better root/shoot ratio during the grain filling period, which allows a higher nutrient and water uptake.
- j) Higher source/sink ratio, which may imply higher supply of assimilates to the roots.
- k) Earliness
- l) Better stalk standability
- m) Better stalk rot resistance
- n) Increased lodging resistance
- o) Increased whole plant yield (forage maize)
- p) Feeding value: higher plant cell wall digestibility and intake, especially since genetic variation for highly digestible cell walls were eliminated or lost during grain maize breeding for stalk standability, stalk breakage resistance and grain yield (Barrière *et al.*, 2005).

Future improvement in maize agronomic value (water and N uptake, root growth, standability, and stress tolerance) depends on a better knowledge of maize physiological process related to yield and yield stability; and must be balanced with more sustainable cropping managements, and with

improvement of the plant feeding value, which implies a better understanding of cell wall biogenesis (Barrière *et al.*, 2006).

## 1.2. Maize requirements in Organic Agriculture

Organic agriculture differs from conventional agriculture since it embraces the concept of naturalness, which includes three approaches: non-chemical, agro-ecological and integrity of life. Organic agriculture looks for prevention strategies in the whole system through a multilevel approach, and it is not only replacing of chemical pesticides for natural ones. For plant breeding, varieties suitable for organic agriculture do not aim maximum yield level and not only direct pest and disease resistance, but optimal yield stability and general adaptation to low-input conditions, including the robustness, flexibility, and reliability of varieties which leads to less yield reduction. It implies several extra plant features and aspects that can contribute to yield stability and decrease the risks of quality and yield loss. Unfortunately, the majority of these traits have few or no relevance from conventional breeding programs or do not have the priority they have in organic farming systems (Lammerts van Bueren *et al.*, 2002, 2004 and 2006). In 1991, the Environment European Commission issued the Nitrates Directive (91/676/EEC) concerning the protection of waters against pollution caused by nitrates from agricultural sources. The aim of the legislation is to safeguard water quality across Europe by preventing nitrates from agricultural sources polluting ground and surface waters and by encouraging the use of good farming practices. Since then, implementations of the Nitrates Directive (Box 1) have been reached (European Commission, 2010). Additionally, the enlarged use of pesticides and herbicides with broader spectra resulted in adverse effects on human and animal health, water and soil pollution, side effects on beneficial organisms, impoverishment of the flora and indirectly of the weed-associated fauna in agricultural landscapes (Meissle *et al.*, 2010).

### **BOX 1. Implementations of the Nitrates Directive**

- Identification of polluted or threatened surface freshwaters, groundwater (containing a concentration of more than 50 mg/L of nitrates), freshwater bodies, estuaries, coastal waters and marine waters (found to be eutrophic or that could become eutrophic).
- Designation of areas of land which drain into polluted or threatened waters and which contribute to N pollution, so called "vulnerable zones" (NVZs). The whole country has been designated as a NVZ in Austria, Denmark, Finland, Germany, The Netherlands and Luxemburg (Jakobsson *et al.*, 2002).
- Establishment of Code(s) of good agricultural practice (CGAP) to be voluntarily implemented by farmers: time when fertilizers can be applied on land, conditions for fertilizer application, requirement for a minimum storage capacity for livestock manure, crop rotations, soil winter cover, catch crops.
- Establishment of Action Programmes to be compulsorily implemented by farmers within NVZs: the Code(s) of good agricultural practice, and limitation of fertilizers to be applied taking into account crops needs, all N inputs and soil supply. The maximum amount of animal manure to be applied is 170 kg N organic/hectare/year.
- National monitoring and reporting every 4 years on: nitrates concentrations, eutrophication, assessment of Action Programmes impact, and revision of NVZs and Action Programmes.

Consequently, reduction of pesticide use and risk is one of the key issues of the EU agenda for agriculture. The so-called "Pesticide Package" includes the Directive 2009/128/EC, which objective is to reduce dependence on, as well as the risks and adverse impact of, pesticide use on human health and the environment (Vasileiadis *et al.*, 2011). Therefore, due to the current regulations in EU, a need for adaptation to low-input conditions and adoption of more sustainable practices actually also exists for conventional agriculture, since it remains a major source of environmental related problems

(European Commission, 2010). The main requirements for maize varieties to be suitable for organic agriculture can be summarized in five points (Schmidt & Burger, 2008 and 2010):

- a) Seed: high genetic vigour, germination, and “thousand-grain weight” (TGW).
- b) Seedling: ability to suppress weeds and high competitive ability against weeds, which implicate high cold tolerance and rapid juvenile development compared with weeds.
- d) High nutrient uptake efficiency through a widespread root system and ability to actively obtain nutrients through excretion of root exudates or through symbiosis with mycorrhizal fungi.
- e) Temporary nutrient deficiency stress tolerance, especially in dry periods (tolerance to drought stress). This requirement is also important for conventional varieties.
- f) Resistance to ear and foliar diseases, plague and abiotic stress. This requirement is also relevant for conventional varieties.

These requirements and consequent breeding goals for organic varieties must be in addition to the conventional breeding objectives such as yield, quality, stability and resistance abilities. Even though, conventional and organic varieties may have different priority of each objective (Lammerts van Bueren *et al.*, 2003; Schmidt & Burger, 2008 and 2010). Additionally, it should be considered that the desired traits for organic maize vary depending on the conditions and management of the farm, the farmer and whether it is feeding maize for livestock or a cash crop for selling (Goldstein, 2011 personal communication). Table 1.1 shows the top 5 traits for different maize purposes in conventional and organic farming systems.

**Table 1.1. Main traits and priorities for maize in conventional and organic farming**

Priority	Grain Maize <sup>a</sup>	Silage Maize <sup>a</sup>	Energy Maize <sup>a</sup>	Organic Feeding Maize <sup>b</sup>	Organic Silage Maize <sup>c</sup>
1	Maturity corrected grain yield	Maturity corrected digestible yield	Maturity corrected methan yield	High reliable yields under organic conditions where N is obtained from organic matter	High stabile yields under organic conditions in rotation with grass-clover
2	Yield stability	Yield stability	Yield stability	High nutritional value (protein content and quality, more minerals, vitamins and better palatability)	Good ability to compete with weeds and fast development to cope with bird damage
3	Standability (green snap, root lodging, and stalk rot resistance)	Standability (green snap and root lodging resistance)	Standability (green snap and root lodging resistance)	Good ability to compete with weeds	Feeding value (digestibility and starch content)
4	Disease resistance ( <i>Helminthosporium</i> , and ear rot)	Disease resistance ( <i>Helminthosporium</i> )	Disease resistance ( <i>Helminthosporium</i> )	Good resistance to abiotic (drought, heat during anthesis, waterlogged conditions, cold, wet cold conditions during grain filling) and biotic stress	Disease resistance
5		Digestibility (starch content, DNDF value)	Methan production rate	Good standability	Root development for nutrient efficiency and organic matter production in the soil

<sup>a</sup> According to PhD. Walter Schmidt. Chief of Germany Maize Breeding KWS SAAT AG, Einbeck, Lower Saxony -Germany; <sup>b</sup> According to PhD. Walter A. Goldstein. Program Director of Mandaamin Institute, Elkhorn, Winsconsin - USA; <sup>c</sup> According to Dr. Nick J.M. van Eekeren. Senior Researcher Agrobiodiversity and Sustainable Animal Husbandry, Louis Bolk Institute, Driebergen, Utrecht - The Netherlands

Due to incidences of contamination of non-GMO and GMO seed because of pollen flow, most of the organic farmers in USA want ‘resistance’ to transgenic pollen as well. But cultivars with gametophytic incompatibility are only now coming onto the market (Goldstein, 2011 personal communication). Overall, farmers want to continue using hybrids and only a maximum of 10% reduction in yield is accepted (Goldstein *et al*, 2012).

A current limitation is the relatively small percentage of organic agriculture area and consequently the limited economic interest of the conventional breeding companies to elaborate breeding programmes specialized in organic varieties, unless the conventional market can also profit from such programs. An important factor for future development of organic plant breeding programs is the societal pressure and governmental policy with respect to the level of chemical inputs in agricultural systems. However,

for ideal results some traits, for instance adaptation to organic soil fertility management, nutrient efficiency, weed suppression, a unique product quality, and root development; require direct selection under organic soil conditions (Lammerts van Bueren *et al.*, 2002). This situation leads toward the questioning of the necessity for breeding and selection under organic conditions. In the future, it is expected that organic breeding programs may be beneficial for both organic and conventional farming systems especially when they are moving away from high input of synthetic agrochemicals (Lammerts van Bueren *et al.*, 2002).

### 1.3. Maize Husbandry

In order to establish the differences between both conventional and organic farming systems, only these management practices that differ between both maize systems will be described.

- A. Seed Treatment: maize seeds may be infected by several seed-borne fungi causing seed rot, and maize seedlings may be infected by soil-borne fungi causing seedling blight. These fungi can produce significant stand loss in poorly drained, excessively compacted, cold and/or wet soils (Cartwright *et al.*, 2006). The most important fungal diseases causing root rot are several species of the genera *Fusarium*, *Pythium*, *Rhizoctonia* and *Acremonium* (Meissle *et al.*, 2010). Therefore, each commercial hybrid maize seed is treated with one or more prevention fungicides. More than 95% of maize seeds planted in Europe are treated with fungicides, with amide, dithiocarbamate and pyrole being the most common active ingredients. Additionally, seeds can also be treated with insecticides against arthropod pest, being wireworms (*Agriotes* spp.) the main targets for these seed treatments in Europe. On a regional level during 2008, Southwest Germany applied seed treatments against Western rootworm larvae (*Diabrotica virgifera virgifera*), and Spain against sap sucking pests in order to prevent transmission of virus. Thiamethoxam, tefluthrin, clothianidin are the most used active ingredients against insect in pesticides. The total area in Europe where seeds were treated with insecticide in 2007 varies from 20% in Hungary and Southwest Poland to 100% in Spain (Meissle *et al.*, 2010). Cultural control practices include planting depth, soil type, seed age, seed viability, seed coat condition (Cartwright *et al.*, 2006):
- Plant high quality, wound-free and disease-free seed into a warm, moist, and firm seedbed.
  - Assure proper placement of seed, pesticides and fertilizers to promote sound and vigorous seedlings.
- B. Nutrient Management: several tools are useful to elaborate a good fertility program, for example, soil tests, local research information, on-the farm research trials, crop nutrient removal, plant analysis, experience from past, or a combination of those. However, the most reliable tool in order to provide guidelines for the optimal use of lime and fertilizers is soil testing. The desirable soil pH for maize ranges from 5.8 to 7.0. Lime is generally recommended at pH values below 5.7 (Espinoza & Ross, 2006).

#### USA

Usually, based on the yield level, 0.45 Kg to 0.68 Kg of pure nitrogen (N) are required for each 25.4 Kg of grain produced. Less N is normally required on sandy loams and silt loam (Table 1.2) than on silty clays and clays soils (Table 1.3). It should be noted that time and methods of nitrogen application are as essential to efficient nitrogen use as nitrogen application rate (Figure 1.1). In this sense only half of the total recommended N should be applied at planting, since the root system is not yet fully developed and there is a possibility of salt damage for the young seedlings. The remaining N should be side-dressed or top-dressed after establishment but before the crop gets the "stage of rapid growth" (6 leaves). All recommended P and K should be applied at or before planting, along with at least 33 Kg of N per hectare. Potash is recommended at soil test levels below 308 Kg/ha. Recommended rates vary from 56 to 224 Kg of K<sub>2</sub>O per hectare, based on needs. Potash applications superior to 100 Kg/ha must be fragmented to avoid salt damage (Espinoza & Ross, 2006).

**Table 1.2. Fertilizer recommendations (N-P<sub>2</sub>O<sub>5</sub>-K<sub>2</sub>O) in Kg/ha for grain maize in sandy loams or silt loams according to yield, P and K levels in the soil**

Soil Test P (Kg/ha)	Soil Test K (Kg/ha)			Yield (Kg/ha)
	Above 308	168-308	Below 168	
Above 112	134-0-0	134-0-67	134-0-100	7 846
67-112	134-56-0	134-56-56	134-67-100	
Below 67	134-67-0	134-67-67	134-67-100	
Above 112	168-0-0	168-0-67	168-0-112	9 416
67-112	168-56-0	168-67-67	168-56-112	
Below 67	168-78-0	168-78-78	168-78-112	
Above 112	202-0-0	202-0-78	202-0-134	10 985
67-112	202-67-0	202-78-78	202-67-134	
Below 67	202-90-0	202-90-90	202-90-134	
Above 112	235-0-0	235-0-90	235-0-168	12 554
67-112	235-67-0	235-67-90	235-67-168	
Below 67	235-100-0	235-100-100	235-100-168	
Above 112	269-0-67	240-0-112	240-0-224	14 123
67-112	269-67-67	240-56-112	240-56-224	
Below 67	269-112-67	269-112-112	269-112-224	

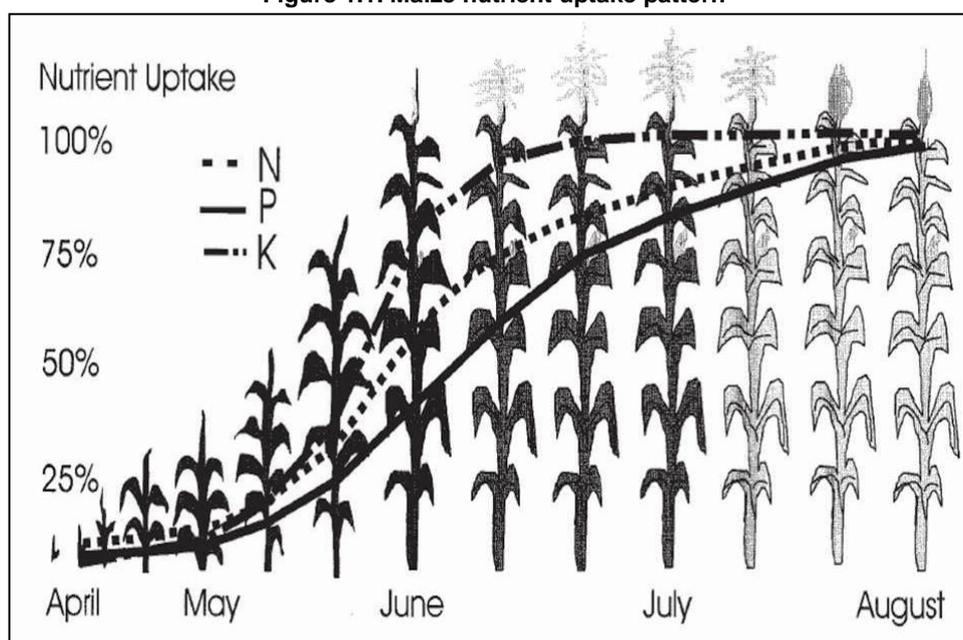
Adapted from Corn Production Handbook, University of Arkansas, 2006

**Table 1.3. Fertilizer recommendations (N-P<sub>2</sub>O<sub>5</sub>-K<sub>2</sub>O) in Kg/ha for grain in silty clays, silty clay loams and clays according to yield, P and K levels in the soil**

Soil Test P (Kg/ha)	Soil Test K (Kg/ha)			Yield (Kg/ha)
	Above 308	168-308	Below 168	
Above 112	140-0-0	140-0-67	140-0-100	6 277
67-112	140-56-0	140-56-56	140-56-100	
Below 67	140-67-0	140-67-67	140-67-100	
Above 112	196-0-0	196-0-67	196-0-112	7 532
67-112	196-56-0	196-67-67	196-56-112	
Below 67	196-78-0	196-78-78	196-78-112	
Above 112	252-0-0	252-0-67	252-0-134	8 788
67-112	252-56-0	252-67-67	252-67-134	
Below 67	252-90-0	252-90-90	252-90-134	
Above 112	336-0-0	336-0-90	336-0-168	10 043
67-112	336-67-0	336-67-90	336-67-168	
Below 67	336-90-0	336-90-90	336-90-168	

Adapted from Corn Production Handbook, University of Arkansas, 2006

Figure 1.1. Maize nutrient uptake pattern



Source: Corn Production Handbook, University of Arkansas, 2006

## EU

The situation in Europe is distinct since the Nitrate Directive limits the maximum amount of manure that can be spread. As an example, in the Netherlands the limits (application standards), the N fertilizer replacement values of various organic manure types, and periods of the year in which the use of fertilizer and manures is forbidden are defined in the Netherlands Action Programme, and published by the Ministry of Economic Affairs, Agriculture and Innovation (Ministerie van Economische Zaken, Landbouw en Innovatie) (Schröder & Neeteson, 2008). Table 1.4 shows the application standards of Nitrogen usage in maize for the period 2010-2013 and Table 1.5 shows the Phosphate application in arable lands for the period 2010-2013.

Table 1.4. Nitrogen application standards for maize (Kg plant available N per ha)<sup>a</sup> in four soils types for the period 2010-2013

Soil type	Clay		Sand		Loess		Peat	
	2010/11	2012/13	2010/11	2012/13	2010/11	2012/13	2010/11	2012/13
Maize with derogation	160	160	150	140	150	140	150	150
Maize without derogation	185	185	150	140	150	140	150	150

<sup>a</sup> Sum of mineral fertilizer and N becoming available during the first season after application of a manure. (Source: [www.minlnv.nl/loket](http://www.minlnv.nl/loket))

Table 1.5. Phosphate application standards (Kg P<sub>2</sub>O<sub>5</sub>/ha-year)<sup>b</sup> for arable lands in three categories according to P level in the soil (Kg/ha) for the period 2010-2013

Soil Test P	Category	2010	2011	2012	2013
<36	Low	85	85	85	85
36-55	Neutral	80	75	70	62
55<	High	75	70	65	55

<sup>b</sup> Sum of mineral fertilizer and organic manure. (Source: [www.minlnv.nl/loket](http://www.minlnv.nl/loket))

Additionally, the Netherlands Action Programme also instructs about storage capacity, places where the use of fertilizers and manure is prohibited (slopes, frozen and snow-covered soil, along water courses). Finally, in the case of maize, it obligates to grow a cover crop (*Brassica rapa*, *Raphanus sativus*, grass, triticale, winter barley, winter rye or winter wheat) after cultivation on sand and loess soil (Schröder & Neeteson, 2008).

- C. Weed Management: weeds should be controlled throughout the season to avoid yield reductions, and an increase of the soil seed bank. Light infestations may reduce 10% to 15% of the yield. Heavy infestations can reduce yields in 50%. The most critical period is the 6 weeks after planting. However, late season infestations might interfere with harvesting, decrease yields, and are very efficient at refilling the seed bank in the soil (Smith & Scott, 2006). In Europe, more than 50 taxa are economically important in maize production. Table 1.6 shows the most important weeds in European maize production (Meissle *et al.*, 2010):

**Table 1.6. Most important weeds in European maize production**

Class	Family	Species
Monocotyledonae	Poaceae	<i>Digitaria sanguinalis</i>
		<i>Echinochloa crus-galli</i>
		<i>Elymus repens</i>
		<i>Panicum</i> spp.
		<i>Poa annua</i>
		<i>Setaria viridis</i>
		<i>Sorghum halepense</i>
Dicotyledonae	Amaranthaceae	<i>Amaranthus</i> spp.
	Asteraceae	<i>Ambrosia artemisiifolia</i>
		<i>Anthemis</i> spp.
		<i>Cirsium</i> spp.
		<i>Tripleurospermum inodorum</i>
		<i>Xanthium</i> spp.
	Caryophyllaceae	<i>Stellaria media</i>
	Chenopodiaceae	<i>Chenopodium album</i>
	Convolvulaceae	<i>Calystegia sepium</i>
		<i>Convolvulus arvensis</i>
	Geraniaceae	<i>Geranium</i> spp.
	Malvaceae	<i>Abutilon theophrasti</i>
	Plantaginaceae	<i>Veronica</i> spp.
	Polygonaceae	<i>Fallopia convolvulus</i>
		<i>Polygonum aviculare</i>
		<i>Polygonum persicaria</i>
	Portulacaceae	<i>Portulaca oleracea</i>
Rubiaceae	<i>Galium aparine</i>	
Solanaceae	<i>Datura</i> spp.	
	<i>Solanum nigrum</i>	
Violaceae	<i>Viola</i> spp.	

Adapted from Meissle *et al.*, 2010

Effective and economical weed control in maize requires an integrated program including crop rotation, water management, cautious mechanical practices, correct herbicide selection, and accurate weed identification (Smith & Scott, 2006). However, in Europe weeds are controlled with herbicides in more than 90% of the maize production area. Pre-sowing application are not common, the pre-emergence number of applications per season range from 0.1 (Southwest Poland and Denmark) to 1.1 (Southwest France) and post-emergence application from 0.4 (Southwest France) to 2.3 (Denmark). The most employed active ingredients are ureas, triazine, pyridine, benzoylcyclohexanedione, amide, oxazole, aromatic acid and nitrile (Meissle *et al.*, 2010). In USA no other crop has as many chemical weed control options as maize. Currently there are over 130 different herbicide brand names (many of them are simply different mixtures of a much smaller number of active ingredients) labelled for weed control (Smith & Scott, 2006). In some European member states, for instance Italy, France, Spain and Hungary, mechanical weed control is applied. In the Netherlands, mechanical weed control is applied in 90% of the conventional maize area from 2000 to 2005 due to a political initiative. Some cultural practices for controlling weeds and reducing of weeds proliferation are listed below (Meissle *et al.*, 2010):

- Stale seedbed (pre-emergent mechanical control).
- Cultivation between rows and within rows (post-emergence mechanical control).

- Flame weeding (pre- and post-emergent control) and ridging.
- Fertilizer application in bands instead of broadcast application.
- Higher plant density if water and nutrients are not limiting factors.
- Cover crops.
- Cleaning of machinery to avoid seed transfer between fields.
- Crop rotations.

While, in the American market exists herbicide tolerant maize varieties, which allow the use of glyphosate (Roundup Ready® varieties) or imazethapyr/imazapyr (Clearfield® varieties) and their worldwide cultivation was more than 30 million hectares in 2008, in Europe such genetically modified (GM), herbicide tolerant varieties are in process of authorization (EFSA, 2009). However, GM crops are indicated as tools of less acceptance for implementation in European farming due to concerns about the social impact and the their acceptance by society (Vasileiadis *et al.*, 2011).

- D. Arthropod Management in Europe: currently, the most important arthropod plague for maize in Europe is the European corn borer (*Ostrinia nubilalis*) with estimated yield losses between 5% to 30% without control measures. This plague together with the Mediterranean corn borer (*Sesamia nonagrioides*) cause economic damage in a range of 2 to 4 million maize hectares in Europe. Other arthropods of economic importance are listed in Table 1.7 (Meissle *et al.*, 2010).

**Table 1.7. Most important arthropod pests in European maize production**

Category	Order	Family	Species	Common Name
Main pest	Lepidoptera	Crambidae	<i>Ostrinia nubilalis</i>	European corn borer
		Noctuidae	<i>Sesamia nonagrioides</i>	Mediterranean corn borer
		Noctuidae	<i>Agrotis spp.</i>	Cutworms
		Noctuidae	<i>Helicoverpa armigera</i>	Cotton bollworm
	Coleoptera	Chrysomelidae	<i>Diabrotica virgifera virgifera</i>	Western corn rootworm
		Elateridae	<i>Agriotes spp.</i>	Wireworms
	Sternorrhyncha	Aphididae	Various	Aphids
	Hemiptera	Cicadellidae	Various	Leafhoppers
Diptera	Chloropidae	<i>Oscinella frit</i>	Frit fly	
Regional pest	Lepidoptera	Noctuidae	<i>Pseudaletia unipuncta</i>	Amyworm
	Diptera	Anthomyiidae	<i>Delia platura</i>	Seed corn maggot
		Opomyzidae	<i>Geomyza spp.</i>	Frit flies
		Tipulidae	<i>Tipula spp.</i>	Crane flies
	Coleoptera	Chrysomelidae	<i>Oulema melanopus</i>	Cereal leaf beetle
		Nitidulidae	<i>Glischrochilus quadrisignatus</i>	Four-spotted sap beetle
		Curculionidae	<i>Tanymecus dilaticollis</i>	Maize leaf weevil
		Scarabaeidae	<i>Melolontha melolontha</i>	May bug
	Thysanoptera	Various	Various	Thrips
	Trombidiformes	Tetranychidae	<i>Tetranychus spp.</i>	Spider mites

Adapted from Meissle *et al.*, 2010

Insecticide applications are employed in all European maize production regions. The main active ingredients for soil insecticide applications include tefluthrin and cypermethrin, and wireworms (*Agriotes spp.*) are the main plague targets. In Hungary and Italy other pest targets are Western corn rootworm larvae (*Diabrotica virgifera virgifera*). In foliar treatments usually one application is done, except in Spain where normally two applications are made. Active ingredients in spray insecticides are mostly pyrethroids and organophosphates, nonetheless oxadiazine, nicotinoid, carbamate and diflubenzuron are used as well. The main targets are the corn borers (*Ostrinia nubilalis* and *Sesamia nonagrioides*). Other plague targets include Western corn rootworm adults, Cotton bollworm larvae and other pest listed in Table 1.7. Cultural practices to reduce arthropod pest pressure are (Meissle *et al.*, 2010):

- Corn borers and Cotton bollworm: mowing stalks and ploughing to diminish the quantity of overwintering larvae, trap crops, mating disruption through synthetic sex pheromones, scouting systems to determinate the main fly and egg-lay periods.

- Western corn rootworm: crop rotation, opportune irrigation and fertilization to speed root regeneration after larvae damage, ridging to prevent lodging, early sowing to develop a robust root system prior the larvae feed, late planting to avoid coincidence with larvae, insecticide baits.
- Cutworms and wireworms: ploughing.

Biological control (for example, corn borers with *Trichogramma* spp. and Western corn rootworm with entomopathogenic nematodes) has also been proved in Europe and has potential to decrease the use of chemical insecticides in the future; however it is not an important method for maize production at the moment. On the other hand, GM *Bacillus thuringiensis* (*Bt*) maize, capable to produce Cry proteins (Cry 1Ab against corn borers and Cry3 against Western corn rootworm), are available for more than 10 years and commercialized in USA since 2003. However, in Europe these GM varieties are still in the authorization process in some member states and some other ones, like Italy, France, Germany and Austria, impose national bans to *Bt* maize. In 2008, Cry1Ab maize was planted in 7 EU countries on a total area of 107 thousands hectares, with 74% of this area only in Spain (Meissle *et al.*, 2010).

- E. Diseases Management in Europe: diseases limit the yield in many maize production areas (Cartwright *et al.*, 2006). The most important fungal diseases in Europe are listed in Table 1.8 (Meissle *et al.*, 2010):

**Table 1.8. Most important fungal diseases in European maize production**

Species	Common Name
<i>F.graminearum</i> , <i>F. verticillioides</i> , <i>F. proliferatum</i> , and <i>F. culmorum</i>	Stalk and ear rot
<i>Pythium</i> spp., <i>Rhizoctonia</i> spp., <i>Acremonium</i> spp.	Stalk rot
<i>Sclerophthora macrospora</i>	Downy mildew
<i>Ustilago maydis</i>	Common smut
<i>Sphaerotheca reiliana</i>	Head smut
<i>Helminthosporium turcicum</i>	Northern leaf blight
<i>Puccinia sorghi</i>	Common corn rust

Adapted from Meissle *et al.*, 2010

In general, foliar fungicide sprays are rarely used in Europe, except in seed production in the Southern part of France against *Helminthosporium* spp., *Fusarium* spp. and *Puccinia* spp. The most recommended cultural management practices are (Meissle *et al.*, 2010):

- Use of varieties with genetic resistance.
- Early planting or use of early varieties to reduce exposure to humid conditions.
- Avoidance of drought stress or poor drainage stressed plants, since they are often more susceptible to diseases.
- Crop rotation with non-host crops (no cereals).
- Ploughing of infested residuals.
- Chopping of residuals before mulching in conservation tillage systems to speed decomposition.
- Accurate identification of the disease and its cause.

A major problem in Europe is the production of mycotoxins by *Fusarium*, which leads to the contamination of both human food and animal feed. This has resulted in regulations for the maximum levels of certain mycotoxins (deoxynivalenol, zearalenone and fumonisins) in foodstuffs and animal feed in EU. Some cultural control practices to avoid the proliferation of mycotoxins in kernels are (Meissle *et al.*, 2010):

- Early planting to reduce feeding damage.
- Low density planting to reduce the humid microclimate.
- Type and amount of N-fertilizers applied.
- Biological control, using *Bacillus subtilis* to reduce mycotoxin accumulation of *F. verticillioides* by competitive exclusion.
- Use of survey system to predict disease damage and mycotoxin production.

F. Maize Cropping Systems in Europe: grain maize production dominates in more dry and warm conditions of central and southern Europe, whereas maize in the cooler and wetter conditions from north and western Europe is typically cultivated for silage. It is grown either as continuous maize or in rotation with other crops (Meissle *et al.*, 2010; Vasileiadis *et al.*, 2011). Wheat or barley are the most common crops in the two-year cycle rotation system. Nevertheless rotation systems of 2 to 5 crops also exist and include maize, wheat, alfalfa, sunflower, temporary grassland, soybean, beets, oilseed rape, rice and potato (Meissle *et al.*, 2010). Vasileiadis *et al.* (2011) described four representative European regions of cropping systems based on maize, including continuous maize and rotation. Table 1.9 shows the main characteristics of these systems:

**Table 1.9. Typical maize based cropping systems in Europe**

Region	Location	Cropping System	Description	Area (ha)	% of total area
Northern	Denmark	Non-irrigated continuous silage maize	When possible, maize usually irrigated only once before anthesis	90 000	50%
		Non-irrigated rotated silage maize	Cereals with undersown grass or a grass and clover ( <i>Trifolium</i> spp.) mixture	90 000	50%
	The Netherlands	Non-irrigated continuous silage maize	When possible, maize usually irrigated only once before anthesis	120 000	50%
		Non-irrigated rotated, silage maize	Grass, cereals, potato ( <i>Solanum tuberosum</i> ) and sugar beet ( <i>Beta vulgaris</i> )	120 000	50%
Central-eastern	Tolna County (Hungary)	Non-irrigated continuous grain maize		84 000	60%
	Békés County (Hungary)	Non-irrigated rotated, grain maize	Winter wheat ( <i>Triticum aestivum</i> ), or oilseed rape ( <i>Brassica napus</i> ) and sunflower ( <i>Helianthus annuus</i> )	42.5 000	85%
South-western	Ebro Valley (Spain)	Irrigated rotated, grain maize	Winter wheat	83 000	83%
		Irrigated rotated silage maize			
		Irrigated continuous grain maize		17 000	17%
Southern	Po Valley (Italy)	Irrigated rotated grain maize	Winter wheat, or barley ( <i>Hordeum vulgare</i> ), or soybean ( <i>Glycine max</i> ), sugar beet, grass or alfalfa ( <i>Medicago sativa</i> )	500 000	50%
		Irrigated rotated silage maize			
		Irrigated continuous grain maize	Associated with livestock producing farms and/or organic highly fertile soils	300 000	30%
		Irrigated continuous silage maize			
		Non-irrigated rotated grain maize	Confined to high rainfall areas (>1000 mm per year) and/or shallow water-table areas	200 000	20%

Adapted from Vasileiadis *et al.*, 2011

From the Table 1.9, it can be concluded that three important factors characterising the system are: (1) type of maize production (grain or silage), (2) cropping sequence (continuous maize or crop rotation), and (3) irrigation (irrigated or non-irrigated). Taking into account these three factors, eight main cropping systems based on maize can be identified in Europe: grain maize rotated irrigated, grain maize rotated non-irrigated, grain maize continuous irrigated, grain maize continuous non-irrigated, silage maize rotated irrigated, silage maize rotated non-irrigated, silage maize continuous irrigated, and silage maize continuous non-irrigated (Vasileiadis *et al.*, 2011).

#### 1.4. The Need for Direct or Indirect Selection

Indirect selection, in the context of the studies below, is defined as the selection for traits under conventional conditions with the purpose to obtain a positive (correlated) selection response for traits under organic conditions. The necessity of direct or indirect selection in maize breeding programs will be discussed, first for programs for low- and high-N input conditions, and then for organic and conventional environments. Below a short and chronologic review of the main investigations into this subject:

Brun & Dudley (1989), to determine the relevance variation in N response in germplasm from USA (high-N) and Argentina (low-N), crossed 50 S<sub>2</sub> (from maize populations with 100, 75, 50, and 25% of Argentine flint germplasm) to a flint and a dent inbred tester. The testcrosses were evaluated under low- and high-N level at 2 locations in Illinois during 1985 and at 3 locations in Argentina during 1986. From their observations, they concluded that selection resulted in higher predicted gains in N response with testing under high-N conditions and, in average, performance over both N levels than with testing at low N environments. They suggested use of high-N levels for variety selection even for low-N conditions (indirect selection).

Presterl *et al.* (2003), to estimate quantitative genetic parameters in order to develop optimal breeding strategies for improving productivity under low-N conditions, presented results of 21 field experiments with hybrids from crosses between European dent and flint inbred lines. The experiments were run under high- and low-N conditions during 1989 and 1999 at several locations in Germany and France. The effectiveness of indirect selection for grain yield was 70% compared with direct selection under low-N conditions. They showed an increased efficiency of direct selection with decrease in grain yield at low-N environments.

Lorenzana & Bernardo (2008), to determine if separate maize breeding programs are needed for organic and conventional production systems, evaluated 119 intermated B73 × Mo17 recombinant inbreds under organic and conventional conditions in 2 locations in Minnesota during 2006. The predicted ratio between the indirect and direct response to selection in the organic system was close to 1 for grain yield and moisture and greatly less than 1 for the other traits. They suggested that high-yielding cultivars for organic farming can be obtained basically by testing inbreds and hybrids under conventional conditions (indirect selection).

Burger *et al.* (2008), to estimate quantitative genetic parameters in order to investigate the perspectives of selection for adaptation to organic conditions, conducted comparative field experiments under organic and conventional farming conditions. From two sources of elite material, sets of 11 flint and 11 dent lines were selected based on their performance under organic and conventional conditions. The study evaluated testcross performance in 3 regions of Germany and selection of superior lines was practiced across 2 stages in 2004 and 2005. In 2006, single-cross hybrids (factorial crosses) from the 2 pools were tested in fields at organic and conventional systems in two regions of Germany. In 2004 and 2005 there was no evidence of adaptation to either organic or conventional conditions, but in 2006 some advantages were slightly displayed if each material is evaluated under the respective target farming system. They concluded that including organic test sites in the regular breeding program increases the possibility of detecting hybrids which perform well in both organic and conventional systems.

Messmer *et al.* (2010), to get conclusions for breeding new varieties optimally adapted to organic agriculture, crossed 4 different material groups of preselected lines derived from early European breeding material. The study in 2008 evaluated testcross performance under organic and conventional conditions in 3 regions in Germany. The phenotypic correlations between organic and conventional agriculture were small or moderate for grain yield in each of the 4 material groups, and strong and highly significant for dry matter content. The top grain yield hybrids for organic agriculture often did not show this superiority under conventional agriculture and vice versa. Heritability for grain yield was similar for both agricultural systems. They concluded that test sites under organic conditions are indispensable for optimizing the development of maize varieties for organic systems.

Brandes (2011), to determinate if maize improvement for organic agriculture requires a separated breeding program in his MSc Thesis, examined in 2009 four sites under organic and conventional conditions each one employing 4 material groups and measuring the grain yield. In 2010 two sites

were evaluated for performance of the crop for silage production (dry matter content). In his results intermediate correlations between organic and conventional farming were observed and the traits under study showed different interactions in both farming systems even in the same location. One of his main conclusions was that selection under conventional conditions does not certainly produce stable, high-yielding varieties for both organic and conventional farming.

From the above studies, it can be observed that genetic correlations are decisive whether indirect selection gives greater response to selection for a determinate trait than direct selection. Response to selection depends on estimates of heritability for both of the environments and the genetic correlation between them. Hence, indirect selection for organic agriculture will be effective if:

- Heritability of the conventional indirect environment is significantly greater than that of the organic direct one.
- The genetic correlation between both of the environments is substantially high.

Additionally, the most usual justification for indirect selection for organic agriculture is the high GxE interaction due to lower trait heritability in low yielding environments. However, this hypothesis is not supported by experimental evidence and the suggested explanation also is not accepted unanimously (Ceccarelli, 1994).

### 1.5. Varietal testing for maize in Europe

The Council Directive 2002/53/EC on the common catalogue of varieties of agricultural plant species, specifies the requirements for the admission of varieties to the Common Catalogue which is needed before they can be commercialized within the EU. This Common Catalogue comprises all National Variety Lists (varieties legally accepted for certification and marketing) of the EU member states. Once admitted to the European Catalogue a variety can be traded in the EU. In order to be included in the National List of an EU member state, a variety should satisfy the criteria below:

- A. Be clearly **distinguishable** at least in one characteristic from any other variety known in the community.
- B. Be sufficiently **uniform** taking into account the distinctive features of the reproductive system.
- C. Be **stable** in such a way that the variety keeps true the description of its native characteristics.
- D. Has **Value for Cultivation and Use (VCU)**, showing an obvious improvement with respect to varieties already in the National List or having a special characteristic that compensates inferior characteristic. Additionally, a suitable **variety denomination** should be approved.

The **Distinctiveness-Uniformity-Stability (DUS)** evaluation is very well-harmonised through the EU member states and follows international guidelines, according to the CD 2003/90/EC and CD 2003/91/EC, such as the International Union for the Protection of New Varieties of Plants (UPOV) guidelines and the Community Plant Variety Office (CPVO) protocol (Schnock, 2006; Van Waes, 2009). In addition, the regulations for the suitability of **variety denomination** are stated in the CD 930/2000/EC (Schnock, 2006). On the other hand, the **VCU** evaluation of agricultural crops differs significantly among the EU member states because the directives are not so detailed in this aspect and the definition of VCU (given in the CD 2002/53/EC) is neither so precise. According to CD 2003/90/EC, the traits to be assessed are:

- a) Yield
- b) Resistance against harmful organisms
- c) Environmental behaviour
- d) Quality characteristics

However, there are no clear guidelines to follow or number of locations, repetitions or duration of the evaluations. This situation gives a lot of freedom to operate to member states with respect to testing the agricultural value of varieties, which leads to dissimilarity in VCU trials among countries (Schnock, 2006; Van Waes, 2009). The Council Directive 2002/55/EC on the marketing of vegetable seed meets the requirements for seed commercialisation and national listing (Schnock, 2006; Van Waes, 2009). Both directives, CD 2002/53/EC and CD 2002/55/EC, benefit the farmers and ensure that they are supplied with high quality seed from good varieties (Schnock, 2006).

## 1.6. Varietal testing for maize in Germany

Since the data to be analysed are from Germany, the German testing system will be summarized here for the case of maize. In Germany, the Federal Office of Plant Varieties (Bundessortenamt) is the responsible entity for the variety testing and registration in the National List according to the German Seed Act. This act, as the ones in place in other EU member states, is based on both directives CD 2002/53/EC and CD 2002/55/EC, and states that a variety has a VCU whether it as a whole improves the crop cultivation or its utilization, or any product derived from such a crop. The improvement should be at least relevant to one defined region of Germany; inferior performance traits will be disregarded in that situation. Applications for variety registration in the German National List can be made for a person or a company within the EU. Since maize is a very important agricultural crop, the number of applications for registration in the German National List is large and amounts annually about 150 new varieties, from which only 10% to 20% is registered. After admission, the variety is registered for a 10 years period with a possibility of prolongation (Schnock, 2006). Both DUS and VCU tests will be described according to Schnock (2006):

### 1. DUS Test:

Commonly needs 2 years. For the evaluation the hybrid and its genealogical components (inbred lines, single crosses) are tested. The trials are conducted at 2 testing stations of the Federal Office of Plant Varieties. The evaluation protocol follows the guidelines established by the CPVO.

### 2. VCU Test:

Normally needs 2 years. Each maize variety is tested for silage and/or grain utilization according to the information given in the application for the breeder. Material is divided in three maturity classes:

- early ( $\leq 220$ ),
- mid-early (230–250)
- mid-late to late ( $\geq 260$ )

The breeder has to indicate the maturity class of a new variety sent in for testing. When it does not fit in this maturity class, it either has to be withdrawn and retested in the correct series the next year. Due to the large number of varieties for testing, new candidates and varieties in the second year of testing have to be tested in separate series:

- 1<sup>st</sup> year series are run at 14 trial sites at breeding stations.
- 2<sup>nd</sup> year varieties are tested at 18 trial sites at institutions of the German Federal States.

Locations are distributed over the three climatic zones corresponding with the three maturity classes. Depending on the number of varieties the VCU tests have a randomised block or a lattice design (if there are more than 40 varieties). Trial series have 6 to 7 standard varieties (controls). The trial is one-factorial with 3 replications per variety. Planting is done with a higher density and the final number of plants per plot is realized by thinning. The plot size must be at least 18 m<sup>2</sup>, with 4 rows from which only the 2 inner rows (9 m<sup>2</sup>) are evaluated and harvested in order to avoid the border effects. The traits evaluated are:

- a) Chilling sensitivity (1–9)
- b) Tillering ability (count, 1–9)
- c) Silking date
- d) Plant height (cm)
- e) European corn borer (*Ostrinia nubilalis*) (count)
- f) Maize smut (*Ustilago maydis*) (count)
- g) Stalk rot (*Fusarium*) (count)
- h) *Helminthosporium turcicum* (1–9)
- i) Lodging (count)
- j) Number of plants before harvest (count)
- k) Ripeness of leaves (only silage maize, 1–9)
- l) Other characters if applicable

For silage maize, harvest starts when the standard varieties have reached 30 to 35% dry matter in the whole plant. The harvested plants are evaluated for dry matter content (basis for judgement and the later description of the maturity: early, mid-early, mid-early to late) and for the dry matter yield of the whole plant (1-9). Samples are taken to evaluate digestibility (1-9) by using ELOS, and starch content (1-9) employing the NIRS method in the laboratory. For grain maize, harvest starts when the standard varieties have reached 60% dry matter in the grain. The harvested grain is evaluated for dry matter content (basis for judgement and the later description of the maturity: early, mid-early, mid-early to late), a trait which also is necessary to determine the grain yield (1-9). Samples are taken to evaluate “thousand-grain weight” (1-9) and “share of broken grains” (1-9).

Yearly detailed agricultural value test reports are elaborated after each VCU test evaluation, where the results are calculated using electronic data processing equipment. This report is given to all breeders concerned over the Internet and a password is needed. In this way, breeders can compare the performance of their varieties with the standard varieties and with the other candidate varieties. Usually, poor performing varieties are withdrawn by the breeders. After the two years of agricultural value testing a summarized report is issued and breeders are invited for discussion with the variety committee (formed by 3 members of the Federal Office of Plant Varieties). This committee is which finally admits or rejects the registration of the variety in the National List. After that, the variety is published in the Common Catalogue and certified seed can be marketed throughout the EU.

### **1.7. Variety testing for Organic Farming**

Recently, the Federal Office of Plant Varieties in Germany decided to include organic trials in the variety testing of conventional maize in order to recommend which of these varieties are specifically adapted to the organic farming (Schmidt, 2011, personal communication). However, since all these tested varieties are selected for conventional agriculture and varieties suitable for organic and/or conventional low input agriculture may have to meet other requirements than the ones for conventional agriculture (Lammerts van Bueren *et al.*, 2003; Lammerts van Bueren & Verhoog, 2006; Löschenberger *et al.*, 2008), there is an important question: Is it enough to rely on the conventionally selected maize varieties and varietal testing under organic conditions for the seed supply for organic agriculture? The alternative is to setup dedicated maize breeding programs for organic agriculture and/or varietal testing systems. This necessity does implicate the implementation of specific VCU protocols considering those traits that are of key relevance for organic agriculture. Micheloni & Plakolm (2007) and Osman *et al.* (2008a) mentioned that such traits are related to plant characteristics such as:

- a) Weed competitiveness and suppression ability
- b) Resistance to seed borne diseases
- c) Nitrogen use efficiency
- d) Yield stability
- e) Tolerance to drought
- f) Final product quality (different specifications for each species)

In cereals, some experience in this subject has been gathered in different EU member states. To test the suitability of new varieties for organic farming, countries like Austria, Germany, the Netherlands, Latvia and Switzerland have started to develop specific VCU protocols for the organic sector (Osman *et al.*, 2008a).

In Germany, in order to know if varieties for organic farming need to be trialed under organic conditions and with specific varietal testing protocols, the Federal Office of Plant Varieties started in 1999 a series of organic trials for winter wheat, and in 2005 and 2006 similar research was made in spring barley and potato. The results showed that the relation of variety characteristics for cultivation, diseases resistance, yield and quality remains similar in both production systems, except for the baking quality of winter wheat (Schnock, 2008). Some shortcomings in the organic VCU testing system are: (1) The organic varieties only have to be better under organic growing conditions; however the conventional testing is paid and made for describing the organic variety in comparison with the conventional released varieties, which means additional costs for breeders. (2) The seed used for organic VCU is not necessarily organically produced seed, however almost no farmer is allowed to

use conventional seed for organic production. For these reasons, organic cereal breeders in Germany demand, instead of combined conventional and organic VCU trials, that separate VCU trials should be done under organic farming conditions with solely organic produced and untreated seed. Another option would be to skip the VCU tests and replace them by regional variety trials with similar conditions as in the area where the variety is expected to be grown (Müller, 2008).

In Austria, the credibility of results from conventional variety trials for organic agriculture was in the eyes of the organic farmers extremely low. For that reason the Austrian Official VCU testing implemented in 2001 and 2002 a separate testing system under organic conditions for winter wheat and spring barley. Later, all other cereals were included in the organic varietal testing system (except maize and durum wheat) and during 2007 and 2008 also field peas and potatoes. As a consequence of this initiative, there are nowadays special breeding programs for winter wheat. The breeders run organic fields to select genotypes suitable for organic farming, and there are now on the market varieties registered based on results exclusively from organic varietal testing systems (Flamm, 2008)

In Latvia, official organic VCU test started in 2004 and was subsidized by the Ministry of Agriculture until 2008, The breeders cover the expenses for the testing themselves. Same methods for conventional VCU were employed in organic certified field. Rotation were included, no seed treatment was used, and weeds were controlled by harrowing. The spring barley variety 'Rubiola', well-adapted to organic farming was selected in this way. Contradictory, the breeder was asked to enter the variety in the conventional VCU test, however the Plant Protection Service was unable to perform exclusive organic VCU testing. Since the national regulation was not ready for such situation, changes were made in the Latvian Catalogue of Plant Varieties and nowadays it is possible to evaluate new varieties under organic, conventional or both conditions depending on the desire of the breeder. Traits relevant to organic agriculture were included in the VCU protocols to enable this (Legzdina, 2008).

In the Netherlands, to compare VCU testing of spring wheat under organic and conventional conditions, variety testing trials were conducted from 2001 to 2004 adding three organic locations to the usual conventional sites, using a new organic VCU protocol (including relevant characteristics for the organic farmers and traders) authorized by the Official Plant Variety Board. Varieties to be tested were selected using criteria relevant in relation to the organic spring wheat ideotype as elaborated in 2000. The results showed a high genetic correlation between farming conditions in most of the traits, however few varieties did not show this tendency. Additionally, traits like baking quality, leafiness and ground cover were easier to assess in organic conditions. The project concluded that for a reliable VCU test system able to select the best varieties for the organic system: conventional and few organic trials should be combined during the evaluation, and key traits for organic agriculture should be included in the protocols. They also remarked that the future of both conventional and organic VCU test systems depends on the financing (Osman *et al.*, 2008b).

In Switzerland, conventional and organic VCU tests were run at the same time on winter wheat from 2002 to 2004. The organic variety tests were declared as an official variety testing program, meaning that varieties were accepted for the Swiss Variety List by passing successfully the organic variety testing. However, results indicated that few differences were found between both farming systems in the variety performance (Micheloni & Plakolm, 2007).

From the experiences described above, it is clear that the current VCU test protocol for varieties for conventional agriculture could restrict the admission of varieties specially adapted to wishes of the organic sector (Lammerts van Bueren & Osman, 2002). Fortunately, the EU regulation is sufficiently flexible to permit specific organic VCU testing. Consequently, in order to develop the organic sector, a VCU testing system including the needs of organic farming is essential. In absence of such a system for organic varieties, breeders are not encouraged to select for the organic market segment. In addition, the main concern is the costs for organic variety testing systems. Since the organic area still is relatively small, testing expenses are not in proportion to the seed market size (Osman *et al.*, 2008a).

## II. OVERALL AIM

The overall aim of this thesis research is to design a breeding procedure for organic maize varieties. Lessons learned to adapt the breeding scheme to develop varieties suitable for organic agriculture and the role of testing under organic conditions to point out varieties for organic agriculture.

## III. RESEARCH QUESTIONS

- 3.1. How useful are the results of testing conventional varieties under organic conditions for recommendation of organic varieties with respect to: means, components of variance, repeatability (heritability), and genotypic mean correlation between conventional and organic trials? (Quantitative genetic parameters).
- 3.2. Is the ranking for performance of varieties the same under both conventional and organic conditions for grain yield and dry matter content? (GxE interaction, genotypic means correlation between conventional and organic trials and Spearman's rank correlation coefficient).
- 3.3. Which traits are important for organic farming systems? Which of those can be evaluated under conventional conditions and which ones under organic or low-input conditions? (Literature review, coefficient of genotypic and phenotypic correlation between farming systems)
- 3.4. How suitable is the current VCU testing protocol for evaluating organic maize varieties? (Literature review for different traits).
- 3.5. What is the impact of the incorporation of an organic testing field in conventional breeding program on its efficiency? (Genotype vs environmental grain yield plot and stability analysis).

With respect to design a breeding procedure for organic maize varieties

- 3.6. Adaptation of maize breeding program to low input farming conditions. (Breeding Program)
- 3.7. In which phase of the program is it better to test under organic conditions? (Breeding Program)

## IV. MATERIALS AND METHODS

The data sets used in this study are from two sources. One is the performance test No. 102 for KWS SAAT AG which provides information on grain and silage yield for both organic and conventional variety testing trials for the German Federal Office of Plant Varieties (Bundessortenamt) during years 2009 and 2010. The other data source is the literature regarding maize studies in quantitative genetic parameters and Genotype x Environment (GxE) interaction and consultations with maize experts in USA, Germany and the Netherlands.

### 4.1. Field Experiments

During 2009 and 2010 KWS SAAT AG conducted the maize performance test No. 102 at 4 locations under organic and conventional farming conditions. All the organic research plots and farms were certified according to EU Regulation No 834/2007 and corresponding regulations as organic farms. The geographic location of the regions in which the 4 sites: Einbeck-Wiebrechtshausen (blue), Niederalteich (pink), Kaufering (green) and Grucking (yellow) are placed, is shown in Figure 4.1.

Figure 4.1. Geographic location of the 4 regions for the field experiments in performance test No. 102 during 2009 and 2010



The organic environment in Wiebrechtshausen was about 13 Km away from the conventional one (Einbeck) and both are treated as one single location, in the other three locations both conventional and organic experimental areas were chosen with the least possible distance from each other in order to minimize the weather related differences between the two testing environments. The geographic location details are described in Table 4.1.

Table 4.1. Location description for the performance test No 102

Location	State	District	Coordinates
Einbeck- Wiebrechtshausen	Lower Saxony	Northeim	51°49'N 9°52'E- 51°42'24"N 10°00'4"E
Niederalteich	Bavaria	Deggendorf	48°46'N 13°01'E
Kaufering	Bavaria	Landsberg am Lech	48°05'N 10°53'E
Grucking	Bavaria	Erding	48°22'N 12°0'E

Tables 4.2 to 4.4 show the set of data for the performance test No. 102 provided by KWS SAAT AG for grain maize and silage maize during 2009 and 2010:

Table 4.2. Locations, farming condition and statistical design for grain maize data for performance test No. 102 during 2009

Location	Farming System		Lattice Design 8*8		
	Conventional (CON)	Organic (ORG)	Entries	Repetitions	Blocks
Einbeck	Einbeck (EIK)	Wiebrechtshausen (WIO)	64	3	24
Niederalteich	Niederalteich (NAK)	Niederalteich (NAO)	64	3	24
Kaufering	Kaufering (KAK)	Kaufering (KAO)	64	3	24

In 2009 (Table 4.2), 64 entries were evaluated under 2 farming systems: conventional and organic; at 3 locations: Einbeck-Wiebrechtshausen, Niederalteich and Kaufering; resulting in 6 environments: 3 conventional (EIK, NAK and KAK) and 3 organic (WIO, NAO and KAO). The performance test had a 8\*8 lattice design with 3 repetitions in each environment. The trials in Einbeck and Wiebrechtshausen were treated as one location named Einbeck.

**Table 4.3. Locations, farming condition and statistical design for grain maize data for performance test No. 102 during 2010**

Location	Farming System		Lattice Design 7*7		
	Conventional (CON)	Organic (ORG)	Entries	Repetitions	Blocks
Grucking	Grucking (GR1) [low density]	-	49	2	14
	Grucking (GR2) [normal density]	Grucking (GRO) [normal density]	49	2 (GR2) 3 (GRO)	14 (GR2) 21 (GRO)
Niederalteich	Niederalteich (NAK)	-	49	3	21

In 2010 (Table 4.3), 49 entries were evaluated under 2 farming systems: conventional and organic; at 2 locations: Grucking and Niederalteich. Grucking under conventional conditions had two planting densities: low density (GR1) and normal density (GR2). Under organic conditions only Grucking with normal planting density (GRO) was obtained. Data from the organic environment in Niederalteich (NAO) was discarded. The performance test had a 7\*7 lattice design with different number of repetitions and blocks per environment.

**Table 4.4. Locations, farming condition and statistical design for silage maize data for performance test No. 102 during 2010**

Location	Farming System		Lattice Design 7*7		
	Conventional (CON)	Organic (ORG)	Entries	Repetitions	Blocks
Einbeck	Einbeck (EIS)	Wiebrechtshausen (WSO)	49	3	21
Kaufering	Kaufering (KAS)	Kaufering (KSO)	49	3	21

In 2010 (Table 4.4), 49 entries were evaluated under 2 farming systems: conventional and organic; at 2 locations: Einbeck-Wiebrechtshausen and Kaufering; resulting in 4 environments: 2 conventional (EIS and KAS) and 2 organic (WSO and KSO). The performance test had a 7\*7 lattice design with 3 repetitions in each environment. The trials in Einbeck and Wiebrechtshausen were treated as one location named Einbeck.

The plot size in all the experiments was 18 m<sup>2</sup> with 4 rows per plot (experimental unit). For the evaluation the two central rows were considered. Lattices were randomized in such a way that in each location both conventional and organic plots had the same order of entries in each replication. This choice was made to improve within locations the comparison between both farming systems (Schmidt, personal communication). Only the data from the grain maize trials in 2009 (Table 4.2) and the silage maize trials in 2010 (Table 4.4) were employed in this study. The data from the grain maize trials in 2010 (Table 4.3) were not used, since they do not allow a proper comparison between the two farming systems. Field management, cultural practices and climatic data of each environment and location are summarized in the Annex 1 and 2.

## 4.2. Genetic Materials

**Grain Maize Test:** 64 maize entries (Annex 3) were employed in the 2009 grain maize experiments. These entries represent a mixture of maize varieties and experimental varieties, selected for conventional agriculture, which were under variety testing including organic environments. The entries were bred for either grain production, silage production or both purposes. The set of the entries can roughly be separated in 5 classes:

- a. Checks of the Federal Office of Plant Varieties: AMANATIDIS, NK FALCONE and RICARDINIO (A) (3 entries).
- b. Hybrids from different breeding companies: any entry already placed on the National List (26 entries).

- c. Hybrids tested for first year: entries with code KXA 9XXX (20 entries)
- d. Hybrids tested for second year: entries with code KXA 8XXX (13 entries)
- e. Hybrids tested for third year: entries with code KXA 7XXX (2 entries)

All entries were evaluated for three traits: grain fresh matter (GFM), grain dry matter content (GDCn1) and grain yield. The mean of each genotype per environment and per trait are presented in Annex 3.

**Silage Maize Test:** 49 maize entries (Annex 4) were employed in the 2010 silage maize experiments. These entries include maize varieties and experimental varieties selected for conventional agriculture. They are included in variety testing for conventional and organic farming and were bred for grain production, silage production or both purposes. The set of the entries comprises 4 classes:

- a. Checks for the Federal Office of Plant Varieties: FABREGAS and NK FALCONE (2 entries).
- b. Hybrids from different breeding companies: any entry already placed on the National List (28 entries).
- c. Hybrids tested for first year: entries with code KXA 0XXX (11 entries)
- d. Hybrids tested for second year: entries with code KXA 9XXX (8 entries)

All entries were evaluated for three traits: total fresh matter (TFM), total dry matter content (TDCn2) and silage yield. Their mean performance in each trial for these traits is summarized in Annex 4.

### 4.3. Statistical Analyses

The statistical analysis was performed employing the software GenStat for Windows 13th Edition and Microsoft Office Excel 2010.

#### 4.3.1. Mean Values and Variance Components

Since the incomplete block design (lattice design) was balanced, the usual analysis of variance (ANOVA) was used to perform all test. Firstly, in order to check the effect of the genotypes in each environment, ANOVAs were performed individually per environment, according to Cochran & Cox (1957), employing the next model:

$$Y_{ij} = \mu + \underline{G}_i + B_j + \underline{e}_{ij} \quad \begin{array}{ll} \underline{G}_i & i.i.d. \sim N(0, \sigma^2_G) \\ \underline{e}_{ij} & i.i.d. \sim N(0, \sigma^2_e) \end{array} \quad \text{where:}$$

$Y_{ij}$  Value for the  $i^{th}$  genotype  
 $\mu$  Grand mean  
 $\underline{G}_i$  Effect of the  $i^{th}$  genotype  
 $B_j$  Effect of the  $j^{th}$  block  
 $\underline{e}_{ij}$  Residual error

Here, the genotypes were defined as random factors and the block structure was formed by the repetitions and plots in each environment. Secondly, in order to analyse the Genotype\*Environment (GxE) interaction and obtain variances components, combined ANOVAs were performed across the environments, farming systems and locations, employing the next model:

$$Y_{ij} = \mu + \underline{G}_i + E_j + B(E)_k + \underline{GE}_{ij} + \underline{e}_{ijk} \quad \begin{array}{ll} \underline{G}_i & i.i.d. \sim N(0, \sigma^2_G) \\ \underline{GE}_{ij} & i.i.d. \sim N(0, \sigma^2_{GE}) \\ \underline{e}_{ijk} & i.i.d. \sim N(0, \sigma^2_e) \end{array} \quad \text{where:}$$

$Y_{ij}$  Value for the  $i^{th}$  genotype in the  $j^{th}$  environment  
 $\mu$  Grand mean  
 $\underline{G}_i$  Effect of the  $i^{th}$  genotype  
 $E_j$  Effect of the environment in the  $j^{th}$   
 $B(E)_k$  Effect of the  $k^{th}$  block  
 $\underline{GE}_{ij}$  Interaction effect of the  $i^{th}$  genotype and the  $j^{th}$  environment  
 $\underline{e}_{ijk}$  Residual error

Since the main interest was in the GxE interaction, the genotypes were defined as random factors, then the environments were defined as fixed factors and nested in the block structure together with repetitions and plots. The original grain maize dataset contained 20 missing values for each trait. In addition, the data from plots with less than 70 plants were omitted and treated as missing value in the ANOVAs. For silage maize data, values from plots with less than 60 plants were replaced by asterisks for the ANOVAs. Thirdly, variance components per farming system were calculated based on the combined ANOVA across farming systems according to Snedecor & Cochran (1980):

$$\begin{aligned}\sigma_e^2 &= MS_{Residual} \\ \sigma_G^2 &= (MS_G - MS_{GE})/ER \\ \sigma_{GE}^2 &= (MS_{GE} - MS_{Residual})/R\end{aligned}\quad \text{where:}$$

$\sigma_e^2$	Error variance
$\sigma_G^2$	Genetic variance
$\sigma_{GE}^2$	Variance of GxE interaction
$MS_{Residual}$	Mean square of the residual error
$MS_G$	Mean square of the genotype
$MS_{GE}$	Mean square of the GxE interaction
E	Number of environments
R	Number of repetitions

#### 4.3.2. Repeatability Coefficient (*Rep%*)

In these experiments, the same genetic material was evaluated in replicated trials in two different farming systems: conventional and organic. Moreover, there was not information through years. Therefore, in order to assess how consistent the measurement is in each farming system: repeatability, instead of heritability, was determined for each farming system based on the variance components of their combined ANOVAs by the following formula (Hallauer *et al.*, 2010):

$$Rep\% = V_G/V_P = \sigma_G^2 / (\sigma_G^2 + \sigma_{GE}^2/E + \sigma_e^2/ER) \quad \text{where:}$$

<i>Rep%</i>	Repeatability
$V_G (= \sigma_G^2)$	Genetic Variance
$V_P$	Phenotypic Variance
$\sigma_{GE}^2$	Variance of GxE interaction
$\sigma_e^2$	Error variance
E	Number of environments
R	Number of repetitions

#### 4.3.3. Pearson's Correlation Coefficients

Pearson's Coefficient determines to which extent the values of two variables are linear related to each other. Next formula according to Mode & Robinson (1959), was used:

$$r_{xy} = \text{COV}_{(x;y)} / \sqrt{\sigma_x^2 \sigma_y^2} \quad \text{where:}$$

$r_{xy}$	Coefficient of correlation between variables x and y
$\text{COV}_{(x;y)}$	Covariance between variables x and y
$\sigma_x^2$	Variance of variable x
$\sigma_y^2$	Variance of variable y

This coefficient was employed with two purposes in this study:

- To measure the linear correlation (*r*) between the traits under study in each farming system and overall environments.
- To calculate the genotypic correlation ( $r_{GCON-ORG}$ ) between the means of the entries under organic and conventional conditions per trait.

#### 4.3.4. Spearman's Rank Correlation Coefficient

After ranking the genotype means for the traits grain yield and GDCn1 in each farming system, the Spearman's rank coefficient ( $R_s$ ) measured the level of correlation (linear dependence) of the ranked means between the two farming conditions, giving a value between +1 and -1, following the formula (Murphy *et al.*, 2007):

$$R_s = 1 - 6(Sd^2) / (n^3 - n)$$

where:

$Sd^2$                     Difference in rank change of each genotype squared and summed for all genotypes  
 $n$                         Number of genotypes

#### 4.3.5. Genotype vs Environmental Grain Yield Plot

The top 3 entries of each of the 6 environments were selected for grain yield. There were 3 environments under conventional and 3 environments under organic conditions. With those entries a scatterplot facing the grain yield of each genotype (Y-axis) vs the environmental mean grain yield (X-axis) was made in order to observe the behavior of each entry in the six different environments and later determinate and localize, if exist, the cross-over point of the GxE interaction for the trait grain yield, as it was explained in Ceccarelli (1994) using barley.

#### 4.3.6. Stability Analysis

The stability analysis was performed employing the Francis & Kannenberg (1978) mean-CV method. The mean-CV method assesses performance and stability at the same time in a simple graphical way. For this method, a stable genotype was defined as the genotype that perform the best and consistently through the environments in a group basis analysis, which implicate a high mean performance (for example, yield) and a small variation (coefficient of variation = CV). The mean performances of each genotype, in the X-axis, were plotted against their CV(%) in the Y-axis. The grand means for performance and CV, across all genotypes (dotted lines in the graphic), divided the scatterplot in four quadrants named groups (Francis & Kannenberg, 1978):

Group I:                high performance, small variation (stable genotypes)  
Group II:              high performance, large variation  
Group III:             low performance, small variation  
Group IV:             low performance, large variation

Before performing this analysis, plot data with a residual larger than 5 times its standard error were omitted and replaced by the fitted values obtained by GenStat.

#### 4.3.7. Cluster Analysis

A hierarchical cluster analysis was performed in order to group the genotypes into classes according to their similarity. For this analysis a similarity matrix was formed with the mean performances of the traits GFM, grain yield and GDCn1 for grain maize (annex 3), and with the mean performances of the traits TFM, silage yield and TDCn2 for silage maize (annex 4). For this and due to the differences in scaling of the traits in study, the data were normalized in order to obtain the same units for all the traits. For normalization the next formula was employed:

$$(X - \mu) / s$$

Where:

$X$                     Individual value (repetition) for each trait  
 $\mu$                     Average for each repetition  
 $s$                     Standard deviation for each repetition

Similarities between genotypes were calculated using the Euclidean algorithm, which uses the geometric distance between the points representing each pair of observations employing the formula (Gower, 1985; Gower & Legendre, 1986):

$$1 - \{(X_i - X_j)/r\}^2 \text{ Where:}$$

$X_i$  Value for observation i  
 $X_j$  Value for observation j  
 $r$  Range of the variate

The method for clustering the genotypes was the Group Average Agglomerative Clustering. This method defines similarity, between a group and a new group formed by combining two groups, as the average of the similarities with each of the original clusters. It should be noticed that this average is taken overall the genotypes in the two merging groups (Gower, 1971).

## V. RESULTS

In order to organize the results of this thesis research, first, the data set for grain maize test were analyzed, summarized and explained. Afterwards, in a similar way the data set for silage maize test were analysed and summarized.

### 5.1. Grain Maize

#### 5.1.1. Varietal Performance

To answer the research question 1 (the utility of testing conventional varieties under organic conditions for recommendation of organic varieties), individual ANOVAs per environment and combined ANOVAs overall environments, per farming systems and per location were performed to observe the significance of the genotype and GxE interaction effects, analyse the mean differences among farming systems and environments, and calculate genetic parameters under organic and conventional farming systems per each trait in study. Individual ANOVAs per environment (Table 5.1) show that there were highly significant ( $p < 0.001$ ) differences between the entries in almost each environment for the three traits under study. In the environment KAO, the traits GFM and grain yield were significant at  $p < 0.05$ . For a detailed list of mean performances of genotypes in each environment consult Annex 3.

**Table 5.1. Summary of statistical analysis per environment for 3 traits in grain maize during 2009. N=64 genotypes**

Environment	Trait	GFM					Grain Yield					GDCn1				
		d.f.	m.v.	s.s.	m.s.	F pr.	d.f.	m.v.	s.s.	m.s.	F pr.	d.f.	m.v.	s.s.	m.s.	F pr.
EIK	Genotype	63		82.9	1.3	<.001	63		29.2	0.5	<.001	63		248.0	3.9	<.001
	Residual	126		66.4	0.5		126		30.0	0.2		126		19.0	0.2	
	Total	191		151.9			191		60.8			191		269.9		
WIO	Genotype	63		271.5	4.3	<.001	63		132.0	2.1	<.001	63		299.0	4.7	<.001
	Residual	95	-31	103.5	1.1		95	-31	44.4	0.5		106	-20	41.3	0.4	
	Total	160	-31	318.7			160	-31	146.1			171	-20	315.8		
NAK	Genotype	63		285.4	4.5	<.001	63		134.6	2.1	<.001	63		442.4	7.0	<.001
	Residual	126		171.7	1.4		126		99.2	0.8		126		55.7	0.4	
	Total	191		472.2			191		240.3			191		506.3		
NAO	Genotype	63		302.5	4.8	<.001	63		133.0	2.1	<.001	63		705.0	11.2	<.001
	Residual	124	-2	213.4	1.7		124	-2	116.0	0.9		126		61.1	0.5	
	Total	189	-2	584.4			189	-2	292.3			191		788.8		
KAK	Genotype	63		229.4	3.6	<.001	63		81.6	1.3	<.001	63		588.4	9.3	<.001
	Residual	125	-1	123.6	1.0		125	-1	55.4	0.4		126		57.6	0.5	
	Total	190	-1	361.7			190	-1	140.1			191		647.7		
KAO	Genotype	56	-7	530.5	9.5	0.014	56	-7	216.3	3.9	0.025	63		624.5	9.9	<.001
	Residual	94	-32	532.5	5.7		94	-32	229.7	2.4		126		182.4	1.4	
	Total	152	-39	970.1			152	-39	407.3			191		812.7		

d.f. = degrees of freedom, m.v. = missing values, s.s. = sum of squares, m.s. = mean square, F pr = probability value for the F test

The combined ANOVAs over all environments (Table 5.2) indicate that the main factor Genotype and GxE interaction are highly significant ( $p < 0.001$ ) for the three traits under study.

**Table 5.2. Summary of statistical analysis across 6 environments for 3 traits in grain maize during 2009. N=64 genotypes**

Trait	GFM					Grain Yield					GDCn1				
	d.f.	m.v.	s.s.	m.s.	F pr.	d.f.	m.v.	s.s.	m.s.	F pr.	d.f.	m.v.	s.s.	m.s.	F pr.
Genotype	63		614.6	9.8	<.001	63		208.5	3.3	<.001	63		2262.2	35.9	<.001
GxE	308	-7	1099.4	3.6	<.001	308	-7	523.9	1.7	<.001	315		645.2	2.0	<.001
Residual	690	-66	1211.2	1.8		690	-66	574.7	0.8		736	-20	417.1	0.6	
Total	1078	-73	8658.9			1078	-73	4247.0			1131	-20	26461.9		

d.f. = degrees of freedom, m.v. = missing values, s.s. = sum of squares, m.s. = mean square, F pr = probability value for the F test

Tables 5.3 and 5.4 summarize the overall means as well as the mean performance of entries for the three grain maize traits per environment and farming system and the corresponding statistical analyses. The first table also shows the highest and lowest entry means. The performance for grain maize traits were on average high, considering both farming systems and the significant effect of GxE interaction, with a substantial differences between environments and farming systems. Table 5.3 shows that the trait means under conventional conditions were significantly ( $p < 0.05$ ) higher than the corresponding trait means under organic conditions. For GFM and grain yield 3.2 Kg/plot and 2.5 Kg/plot superior respectively. For GDCn1 the difference was 2.7%. The mean range, expressed as the difference between maximum and minimum means, was larger in the organic farming system in all of the three traits.

**Table 5.3. Comparison of the trait means of conventional and organic farming systems for 3 traits in grain maize during 2009. N=64 genotypes**

Trait		Overall	Farming System		Environment					
			CON	ORG	EIK	WIO	NAK	NAO	KAK	KAO
GFM (Kg/plot)	<b>Mean</b>	<b>14.5</b>	<b>16.1</b>	<b>12.9</b> *	<b>15.7</b>	<b>16.2</b>	<b>15.5</b>	<b>12.8</b> *	<b>17.1</b>	<b>9.8</b> *
	Max	16.2	17.9	15.2	17.0	18.7	18.9	15.4	19.3	13.8
	Min	12.9	14.2	10.2	14.2	13.2	11.8	9.4	13.1	4.6
Grain Yield (Kg/plot)	<b>Mean</b>	<b>9.9</b>	<b>11.1</b>	<b>8.6</b> *	<b>10.4</b>	<b>10.2</b>	<b>11.7</b>	<b>9.3</b> *	<b>11.3</b>	<b>6.2</b> *
	Max	10.9	12.1	9.9	11.2	11.7	13.6	11.0	12.5	9.1
	Min	8.9	10.0	6.8	9.4	8.2	9.0	6.8	8.9	3.0
GDCn1 (%)	<b>Mean</b>	<b>68.0</b>	<b>69.3</b>	<b>66.6</b> *	<b>66.1</b>	<b>63.2</b> *	<b>75.4</b>	<b>72.7</b> *	<b>66.3</b>	<b>64.0</b> *
	Max	70.8	72.2	70.2	68.8	66.1	78.0	77.1	70.3	68.5
	Min	64.9	66.2	63.6	62.8	60.4	71.9	68.6	63.0	61.2

Max = maximum performance, Min = minimum performance, \* Significant difference at  $p = 0.05$

Since GxE interactions were significant for the three traits in the combined ANOVA, in order to count with a more detailed analysis and determinate genetic parameters, ANOVAs per farming system and locations were run. Table 5.4 shows that all GxE interaction effects were highly significant ( $p < 0.001$ ) per farming system and per location for the three traits under study. Genotype main effect for GFM and GDCn1 were still highly significant ( $p < 0.001$ ) per farming system, however, genotype main effect for grain yield was significant at  $p = 0.01$ . Per location, the main effect of genotype were not significant in locations Einbeck and Kaufering for GFM and grain yield. In Niederalteich GFM showed highly significant ( $p < 0.001$ ) main effect of genotype, and the trait grain yield was significant at  $p = 0.05$ . Trait GDCn1 presented significant genotype main effect in the three the locations.

**Table 5.4. Summary of statistical analysis of conventional and organic farming systems and per location for 3 traits in grain maize during 2009. N=64 genotypes**

Farming System or Location	Trait	GFM					Grain Yield					GDCn1				
		Source of variation	d.f.	m.v.	s.s.	m.s.	F pr.	d.f.	m.v.	s.s.	m.s.	F pr.	d.f.	m.v.	s.s.	m.s.
CON	Genotype	63		338.1	5.4	<.001	63		112.3	1.8	0.006	63		1091.5	17.3	<.001
	GxE	126		259.6	2.1	<.001	126		133.1	1.1	<.001	126		187.3	1.5	<.001
	Residual	377	-1	361.8	1.0		377	-1	184.6	0.5		378		132.3	0.4	
	Total	574	-1	1249.6			574	-1	610.4			575		12314.7		
ORG	Genotype	63		595.6	9.5	<.001	63		257.7	4.1	0.001	63		1282.3	20.4	<.001
	GxE	119	-7	568.1	4.8	<.001	119	-7	254.8	2.1	<.001	126		346.2	2.7	<.001
	Residual	313	-65	849.4	2.7		313	-65	390.1	1.2		358	-20	284.8	0.8	
	Total	503	-72	4956.1			503	-72	2119.0			555	-20	12346.2		
Einbeck	Genotype	63		189.8	3.0	0.236	63		81.4	1.3	0.5	63		486.7	7.7	<.001
	GxE	63		164.6	2.6	<.001	63		79.8	1.3	<.001	63		60.3	1.0	<.001
	Residual	221	-31	170.0	0.8		221	-31	74.4	0.3		232	-20	60.3	0.3	
	Total	352	-31	497.3			352	-31	207.5			363	-20	1365.7		
Niederaltlach	Genotype	63		411.3	6.5	<.001	63		166.9	2.6	0.019	63		1067.7	16.9	<.001
	GxE	63		176.6	2.8	<.001	63		100.7	1.6	<.001	63		79.6	1.3	<.001
	Residual	250	-2	385.1	1.5		250	-2	215.2	0.9		252		116.8	0.5	
	Total	381	-2	1759.4			381	-2	1079.4			383		2008.1		
Kaufering	Genotype	63		447.7	7.1	0.159	63		153.9	2.4	0.659	63		1105.9	17.6	<.001
	GxE	56	-7	316.5	5.7	<.001	56	-7	145.6	2.6	<.001	63		107.0	1.7	<.001
	Residual	219	-33	656.1	3.0		219	-33	285.1	1.3		252		240.0	1.0	
	Total	343	-40	5511.1			343	-40	2557.3			383		1960.4		

d.f. = degrees of freedom, m.v. = missing values, s.s. = sum of squares, m.s. = mean square, F pr = probability value for the F test

### 5.1.2. Genetic Parameters

Based on the combined ANOVAs across farming systems the variance components were calculated for comparison of parameters under conventional and organic conditions. In Table 5.5, for the trait GFM, the estimate for  $\sigma^2_G$  was slightly higher, for  $\sigma^2_{GE}$  was almost the double, and for  $\sigma^2_e$  was much higher under organic than under conventional conditions. Under conventional conditions,  $\sigma^2_{GE}$  was similar to  $\sigma^2_G$ ; under organic conditions  $\sigma^2_{GE}$  was slightly higher than  $\sigma^2_G$ . The  $\sigma^2_e$  was higher than  $\sigma^2_{GE}$  under conventional conditions and very much higher under organic conditions. *Rep%* under conventional conditions was 12% higher than under organic ones.

**Table 5.5. Estimates of genetic parameters under conventional and organic conditions for 3 traits in grain maize during 2009. N=64 genotypes**

Parameter	GFM		Grain Yield		GDCn1	
	CON	ORG	CON	ORG	CON	ORG
$\sigma^2_G$	0.37	0.52	0.08	0.22	1.76	1.96
$\sigma^2_{GE}$	0.37	0.69	0.19	0.30	0.38	0.65
$\sigma^2_e$	0.96	2.71	0.49	1.25	0.35	0.80
<i>Rep%</i>	61.6	49.5	40.7	47.7	91.4	86.5

$\sigma^2_G$  = genetic variance,  $\sigma^2_{GE}$  = GxE interaction variance,  $\sigma^2_e$  = error variance, *Rep%* = repeatability

In the case of grain yield, the estimate for  $\sigma^2_G$  was higher, for  $\sigma^2_{GE}$  was slightly higher and for  $\sigma^2_e$  was higher under organic farming than the corresponding ones for conventional farming. Under conventional conditions,  $\sigma^2_{GE}$  was higher than  $\sigma^2_G$ , and  $\sigma^2_e$  was higher than  $\sigma^2_{GE}$ . Under organic conditions,  $\sigma^2_{GE}$  was slightly higher than  $\sigma^2_G$ , and  $\sigma^2_e$  was very much higher than  $\sigma^2_{GE}$ . *Rep%* under organic conditions was 7% higher than under conventional conditions. For the trait GDCn1, the estimate for  $\sigma^2_G$  was similar for both farming systems. The estimate for  $\sigma^2_{GE}$  was almost the double and for  $\sigma^2_e$  was higher under organic farming than the corresponding ones for conventional farming. Both farming systems presented lower  $\sigma^2_{GE}$  than  $\sigma^2_G$ , and similar  $\sigma^2_e$  and  $\sigma^2_{GE}$  for this trait. *Rep%* under conventional conditions was 5% higher than under organic ones.

### 5.1.3. Correlation Coefficients

After the means, variances and genetic parameters were described for each trait, the linear relationship ( $r$ ) between the three traits under study was assessed in order to support the discussion about these results. Table 5.6 shows that the traits GFM and grain yield were highly significant and positively correlated in both farming systems and over all the environments. On the other hand, traits GFM and GDCn1 showed in general a highly significant negative correlation. The traits grain yield and GDCn1 were not significantly correlated.

**Table 5.6. Estimates of linear correlations between 3 traits in grain maize during 2009. N=64 genotypes**

Farming System	Pearson's Correlation	Traits		
		GFM-Grain Yield	GFM-GDCn1	Grain Yield-GDCn1
CON	$r$	0.91	-0.59	-0.22
	$t$ pr.	<.001	<.001	0.08
ORG	$r$	0.96	-0.20	0.07
	$t$ pr.	<.001	0.11	0.58
Overall	$r$	0.92	-0.52	-0.15
	$t$ pr.	<.001	<.001	0.24

$t$  pr. = t-approximation test, with approximately a t-distribution on  $n-2$  degrees of freedom

Additionally, in order to answer the research questions 1 (the utility of testing conventional varieties under organic conditions for recommendation of organic varieties) and 2 (the performance of varieties under both conventional and organic conditions), genotypic correlation between the means of the genotypes under conventional and organic conditions ( $r_{\text{GCON-ORG}}$ ) was evaluated per trait. Table 5.7 shows that trait GFM presented a highly significant medium correlation between farming systems. For grain yield the correlation between farming systems is low and highly significant. In the case of GDCn1, there was a highly significant total correlation between farming systems.

**Table 5.7. Estimates of correlations between genotypic means with conventional and organic farming conditions for 3 traits in grain maize during 2009. N=64 genotypes**

Trait	Pearson's Correlation	Farming System	Einbeck	Niederalteich	Kaufering
GFM	$r_{\text{GCON-ORG}}$	0.70	0.12	0.59	0.31
	$t$ pr.	<.001	0.35	<.001	0.01
Grain Yield	$r_{\text{GCON-ORG}}$	0.51	0.02	0.41	0.03
	$t$ pr.	<.001	0.88	<.001	0.81
GDCn1	$r_{\text{GCON-ORG}}$	1.01	0.82	0.92	0.90
	$t$ pr.	<.001	<.001	<.001	<.001

$t$  pr. = t-approximation test, with approximately a t-distribution on  $n-2$  degrees of freedom

### 5.1.4. Rank Correlations

The  $r_{\text{GCON-ORG}}$  already measured the correlation between the genotypic means of farming systems per trait. However, since the hybrids mean performance were significantly different between farming systems, there is an interest to know if the best performing hybrids in the conventional system are also the best performing hybrids in the organic system (research question 2). To achieve this answer the linear relationship of ranks per farming systems have to be evaluated. For that, the Spearman's Rank Correlation Coefficient ( $R_s$ ) is employed.

From Table 5.8, the analysis for t-approximation test indicated that the three traits had significant correlation between conventional and organic ranks; however trait grain yield had slight significant ( $p=0.016$ ) correlation. The estimates for the coefficients were in general positive for each of the three traits studied; those for grain yield and GFM were low correlated, and those for GDCn1 very high, indicating an almost perfect monotone increasing relationship between conventional and organic ranks for this trait. In Einbeck, the traits GFM and grain yield did not show a significant correlation between

farming system ranks. GDCn1 showed a highly significant correlation between farming system ranks. In Niederalteich, the coefficients for GFM and grain yield were just low to very poor, respectively; whereas the one for GDCn1 was high and highly significant. In Kaufering, GFM and grain yield showed not a significant correlation between farming system ranks, GDCn1 showed a high and significant correlation. These results shows that performance testing under conventional and organic conditions does not produce the same ranking for GFM and grain yield, whereas the farming system is irrelevant for GDCn1 ranks.

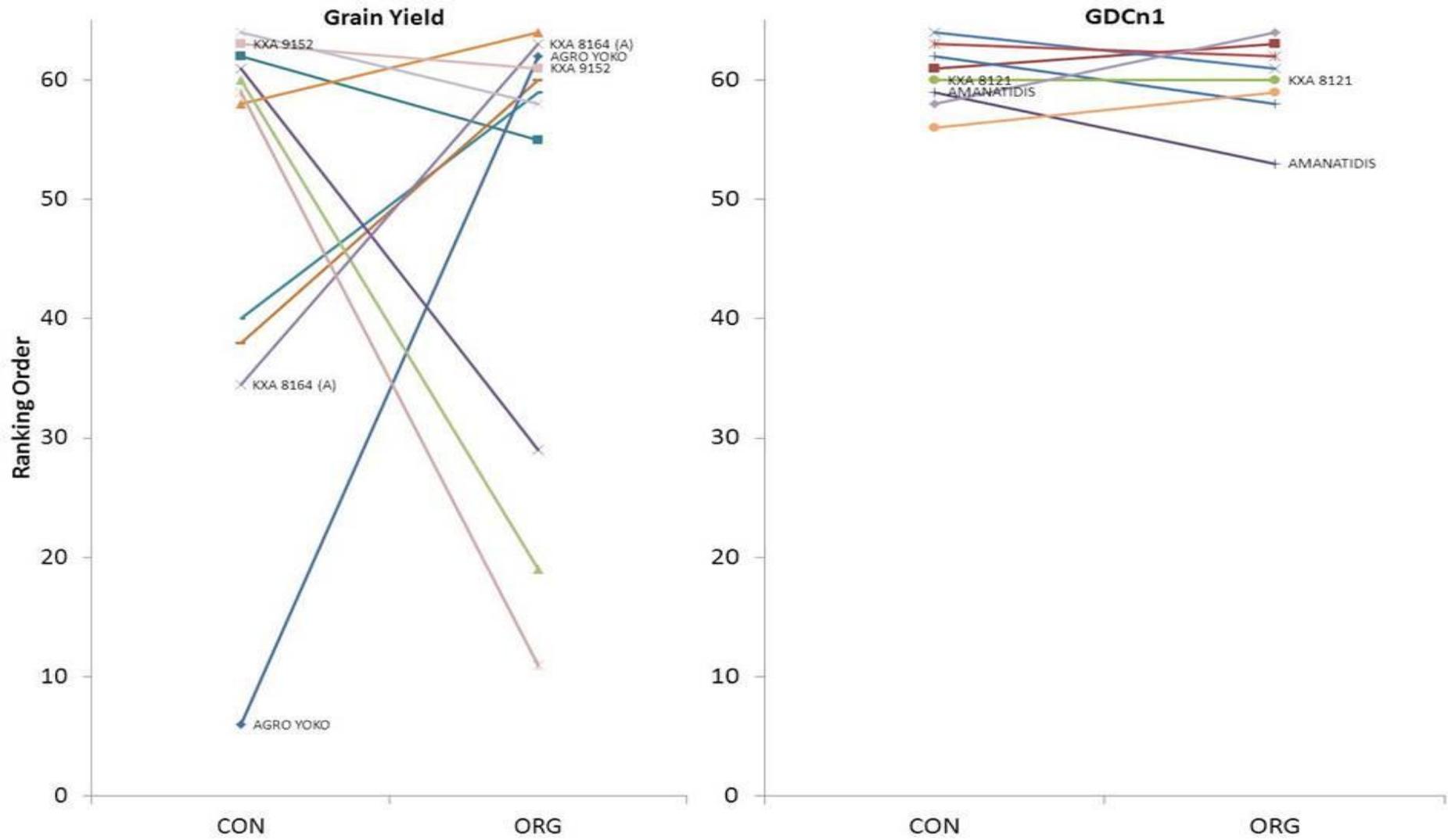
**Table 5.8. Estimates of rank correlations with conventional and organic farming for 3 traits in grain maize during 2009. N=64 genotypes**

Trait	Spearman's Rank Correlation	Farming System	Einbeck	Niederalteich	Kaufering
GFM	$R_s$	0.41	0.08	0.40	0.20
	t pr.	<.001	0.521	0.001	0.118
Grain Yield	$R_s$	0.30	0.00	0.26	0.04
	t pr.	0.016	0.984	0.04	0.74
GDCn1	$R_s$	0.91	0.74	0.88	0.82
	t pr.	<.001	<.001	<.001	<.001

t pr. = t-approximation test, with approximately a t-distribution on n-2 degrees of freedom

Plotting ranks offers an insight of the rank change patterns. Since the interest of VCU testing is to identify the best varieties, only the top 6 entries per farming system were traced for the traits grain yield and GDCn1. For a complete trait-specific list of varietal rankings, see Annex 5. Figure 5.1 shows for grain yield that only one genotype was ranked in the top 6 of both farming systems: KXA 9152. Entry KXA 8164 (A) is an example of genotype with medium rank change. Moreover, entry AGRO YOKO represented a genotype with extreme rank change in grain yield. On the other hand, trait GDCn1 showed relatively small rank changes between conventional and organic farming. This is illustrated by the entry KXA 8121 without rank change and AMANATIDIS showing a moderate change in ranks. Most of the genotypes ranking first under conventional conditions, also rank first under organic conditions, the top 6 for this trait have four genotypes in common. As a remark, trait GFM (figure not presented) showed a similar pattern as grain yield, since both traits are highly correlated (Table 5.6).

Figure 5.1. Comparison of rank change patterns for the traits grain yield and GDCn1. Genotypes are ranked from 64 = highest to 1 = lowest



### 5.1.5. GxE interaction: Genotype vs Environmental Grain Yield Plot

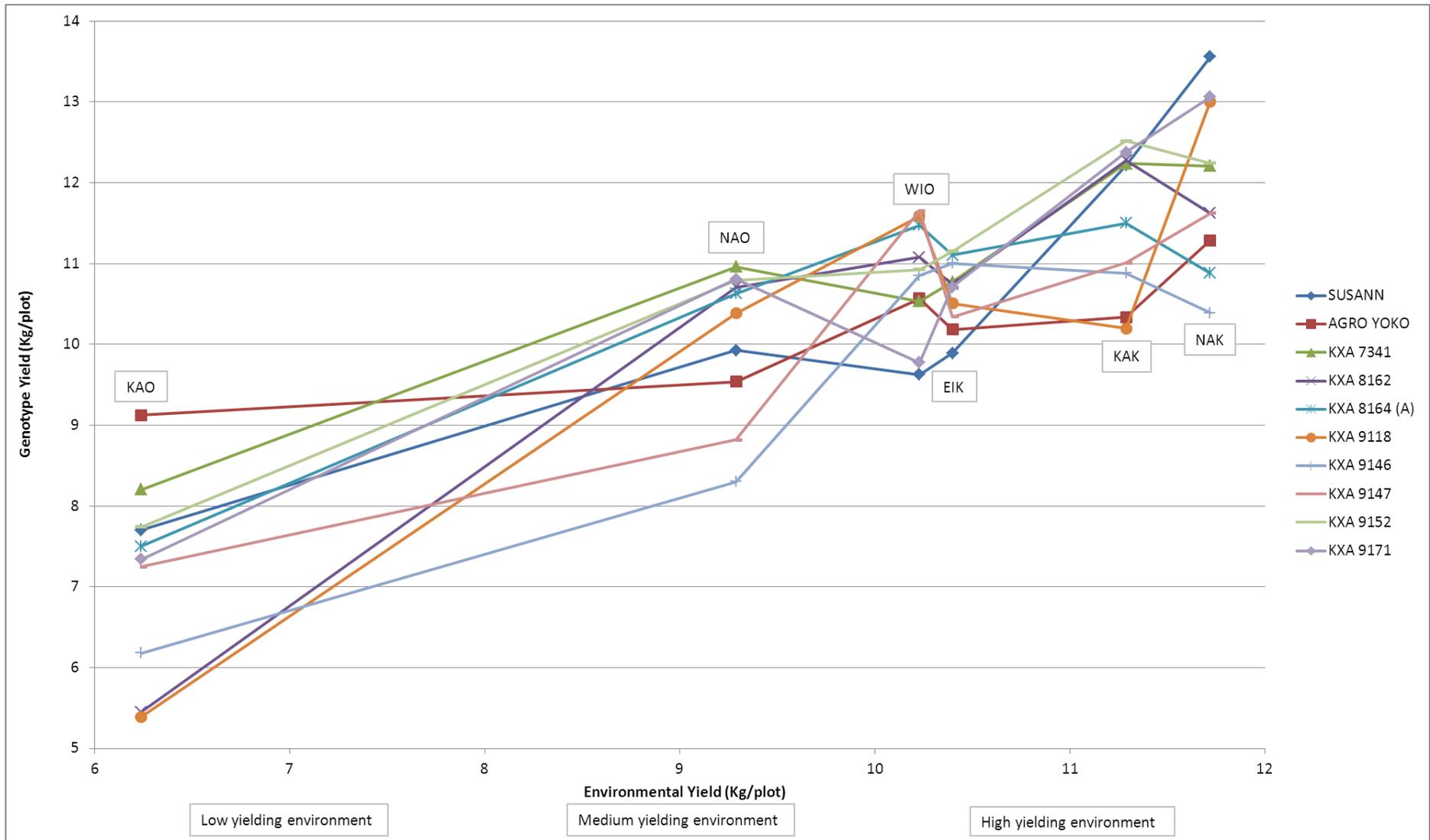
In order to determine and localize the GxE cross-over point, and to answer the research question 5 (the impact of organic testing fields in the efficiency of conventional breeding program), the Genotype vs Environmental Grain Yield Plot was elaborated.

After checking the top 3 genotypes in each environment for grain yield 10 entries were selected to elaborate the scatterplot of Genotype vs environmental yield showed in Figure 5.2. The mean grain yield per environment differed considerably (Table 5.3), being the lowest in KAO (6.2 Kg/plot) and the highest in NAK (11.7 Kg/plot). The yields for NAO, WIO, EIK and KAK were in between those extreme values; the organic environments had on average a lower yield compared to the conventional environments. When the graphic was separated in "yielding environments" only KAO fit in the place of "low yielding environments", NAO was considered a "medium yielding environment" and WIO, EIK, NAK and KAK were considered "high yielding environments". To check the response of the genotypes to the farming system the graphic was divided in two segments, the organic segment (from KAO to WIO) and the conventional segment (from EIK to NAK):

- Organic segment: in general all the genotypes increased significantly their grain yields from KAO through WIO and different cross-over occurred. However the line slopes were steeper in some genotypes, other genotypes displayed parallel lines, and genotype AGRO YOKO showed initially a straight line that was later steeper.
- Conventional segment: in general eight out of the ten genotypes increased significantly their grain yields from EIK through NAK and different cross-over occurred. Genotype SUSSAN showed the steeper slope and the larger increase in grain yield.

All these observations demonstrated that genotypes can behave differently depending on the environments used for the varietal testing. The cross-over interaction can be observed in certain genotypes, but not in all of them. For that reason, entries showing specific adaptation were considered in order to establish where the cross-over interaction can be expected. The best entry in the lowest yielding environment and the best one in the highest yielding environment were chosen. These genotypes were AGRO YOKO (red line) in KAO and SUSSAN (blue line) in NAK; in environments KAO and WIO the difference was significant in favour of AGRO YOKO, in environments NAO and EIK no significant difference was found between both entries; consequently in the segment from KAO to EIK no significant GxE interaction of cross-over type was observed between those genotypes. In the section from EIK to KAK a significant difference in favour of SUSSAN was observed and maintained until NAK. When the same procedure was done with the second best genotype in KAO (KXA 7341) and the second best genotype in NAK (KXA 9171) the cross-over type interaction was found in the section between environments EIK and NAK (figure not shown). Consequently, the GxE cross-over type of interaction, for this set of 64 genotypes under these 6 environmental conditions, was located in the range between 10.4 and 11.7 Kg/plot.

Figure 5.2. Mean grain yield of the best 10 entries at 6 environments vs mean of the 64 entries at each environment during 2009



Least Significant Difference (LSD) = 0.6 Kg/plot at  $\alpha=0.05$  was taken to compare genotype and environmental grain yield differences

### 5.1.6. Stability Analysis

The previous analysis already located the GxE of crossover type, from here the interest was to identify and classify the genetic material under study. In order to answer the research question 5 (the impact of organic testing fields in the efficiency of conventional breeding program) and distinguish stable genotypes, the mean-CV analysis was used. Considering the definition of stable genotype by Francis & Kannenberg (1978), this analysis was focused on identification of the Group I entries: genotypes which, in presence of GxE interaction, showed performances above the group average (high yield) and variation below the group average (low CV%).

Figures 5.3 to 5.5 show Mean-CV plots for grain yield of varieties taking into account all environments (Figure 5.3) and the organic and conventional farming environments, separately (Figures 5.4 and 5.5), in order to reveal differences in stability between the genotypes. Table 5.9 summarizes the varietal groupings with help of each analysis. Group I (stable genotypes) comprised 24 genotypes. The analyses per farming system resulted in 18 Group I genotypes for conventional farming, and 22 Group I genotypes for organic farming. Only 9 Group I genotypes detected in the latter analyses were identical: AMBROSINI, CASSILAS, SYMBOL, KXA 7341, KXA 8164 (A), KXA 8181, KXA 9145, KXA 9148 and KXA 9152. This implicates that those genotypes were the most stable under both farming conditions. These varieties also classified as Group I types by the overall analysis. Some genotypes showed specific adaptation to organic farming: entry AGRO YOKO which performed quite well under organic conditions with little variation, the performance of thus genotype under conventional conditions fitted in Group III. These results showed that wide and specific adapted varieties can be found in these genotypes.

The standard varieties AMANATIDIS, NK FALCONE and RICARDINIO (A) were not part of the Group I and were not considered as stable genotypes in any of the analysis performed. Indeed, all of them formed part of Group IV in the overall analysis, showing always a mean yield below the group mean and poor stability. From the Mean-CV analysis, an important aspect is the CV of each genotype which was used as a measure of consistency among performances. In the same way, and considering that the same entries were employed in different environments, the grand mean CV (horizontal dotted line in the graphics) of each analysis are used to demonstrate how variable on average the genotypes were in a specific farming system compared to another. The same criterion was used to compare locations and environments. For that, Table 5.10 summarize the grand mean CV of genotypes for the three traits and the corresponding highest and lowest genotype CV. For GFM and grain yield, Table 5.10 shows a very similar distribution of the genotype CV, both traits had around 20% in the overall analysis and about 8% in conventional environments; on the other hand, the analysis for the organic environments gave a CV that was around three times the value obtained for the trials under conventional conditions. The range, expressed as the difference between maximum and minimum genotype CVs, was larger in the organic farming system for both of these traits. In the case of GDCn1, the analysis at any level had similar values in the mean, and the range of the genotype CV. In general there were very low genotype CV values for this trait. These results are in agreement with three facts: the high linear correlation between GFM and grain yield; the high *Rep%* of the trait GDCn1 under both conventional and organic farming conditions and its high  $r_{\text{GCON-ORG}}$ ; and the variation among locations, environments and between farming systems.

Figure 5.3. Mean-CV plot of varieties for Grain Yield (Kg/plot) based on 2009 trials in all environments (2 farming systems and 3 locations with 3 repetitions)

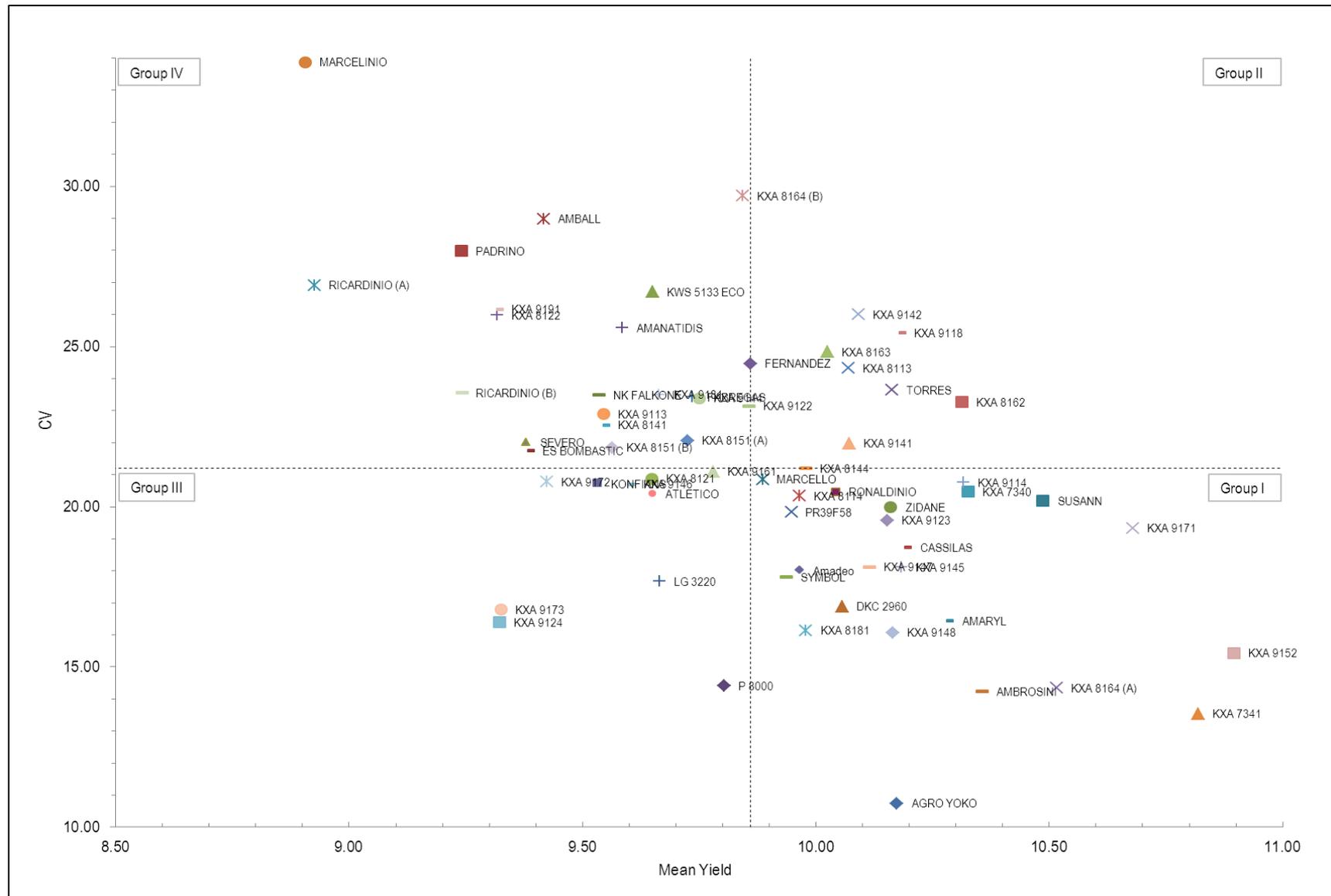
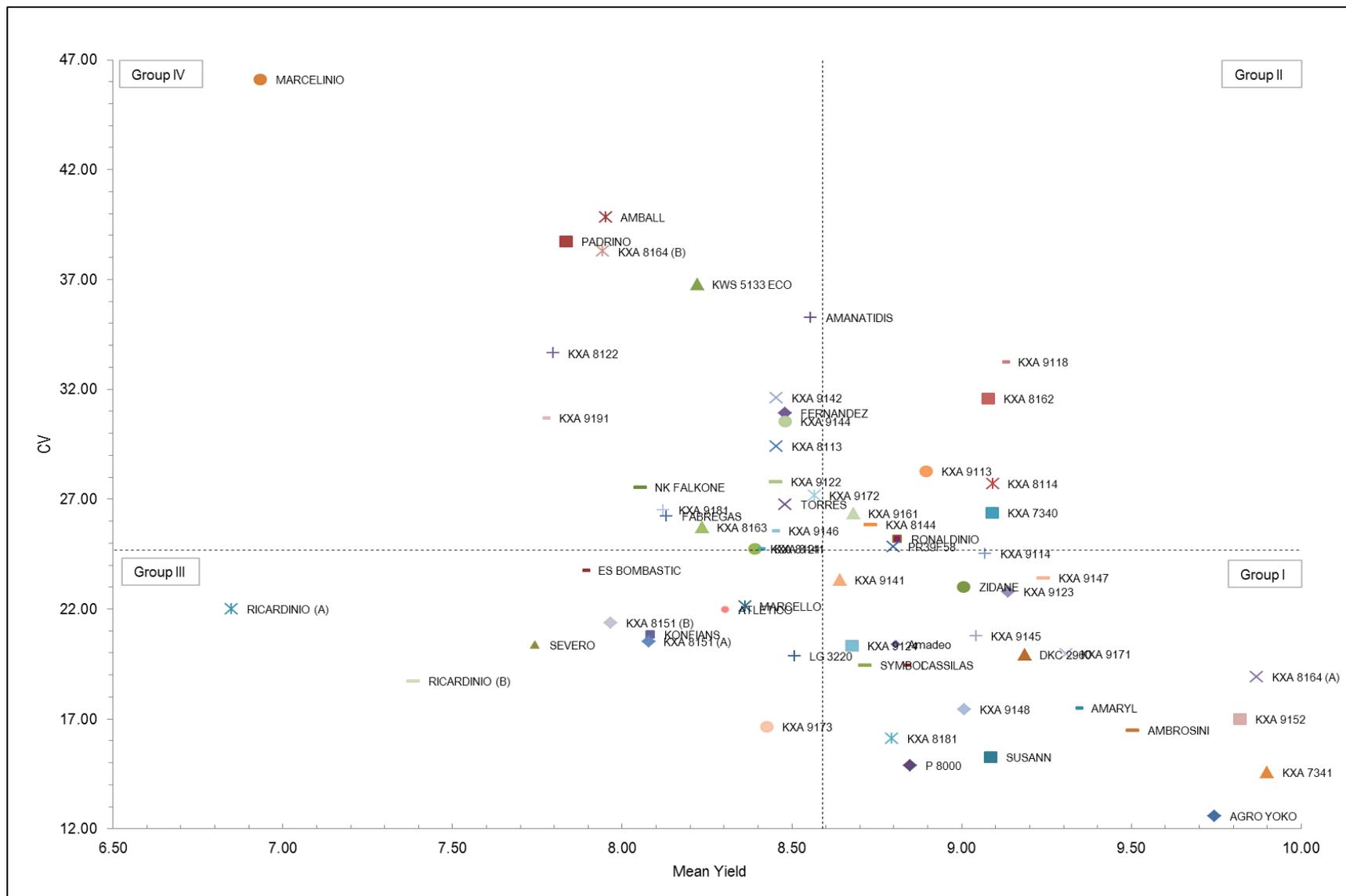




Figure 5.5. Mean-CV plot of varieties for Grain Yield (Kg/plot) based on 2009 trials in organic environments (3 locations with 3 repetitions)



**Table 5.9. Comparison of groups formed for grain yield after Mean-CV analysis of 64 genotypes overall environments, and in conventional and organic farming conditions**

Entry	Denomination	Overall	Farming System	
			CON	ORG
1	Amadeo	I	III	I
2	RONALDINIO	I	I	II
3	SEVERO	IV	III	III
4	KONFIANS	III	III	III
5	MARCELLO	I	I	III
6	ATLETICO	III	III	III
7	LG 3220	III	III	III
8	ES BOMBASTIC	IV	III	III
9	NK FALKONE	IV	III	IV
10	P 8000	III	III	I
11	SUSANN	I	II	I
12	DKC 2960	I	IV	I
13	PR39F58	I	III	II
14	AMBALL	IV	IV	IV
15	ZIDANE	I	II	I
16	AMANATIDIS	IV	IV	IV
17	AMARYL	I	II	I
18	AMBROSINI	I	I	I
19	AGRO YOKO	I	III	I
20	PADRINO	IV	III	IV
21	KWS 5133 ECO	IV	III	IV
22	TORRES	II	I	IV
23	RICARDINIO (A)	IV	III	III
24	MARCELINIO	IV	III	IV
25	FABREGAS	IV	I	IV
26	CASSILAS	I	I	I
27	SYMBOL	I	I	I
28	FERNANDEZ	IV	II	IV
29	KXA 7340	I	I	II
30	KXA 7341	I	I	I
31	KXA 8113	II	I	IV
32	KXA 8114	I	III	II
33	KXA 8121	III	III	IV
34	KXA 8122	IV	III	IV
35	KXA 8141	IV	IV	IV
36	KXA 8144	II	II	II
37	KXA 8151 (A)	IV	II	III
38	KXA 8162	II	I	II
39	KXA 8163	II	II	IV
40	KXA 8164 (A)	I	I	I
41	KXA 8181	I	I	I
42	KXA 9113	IV	IV	II
43	KXA 9114	I	II	I
44	KXA 9118	II	II	II
45	KXA 9122	IV	II	IV
46	KXA 9123	I	II	I
47	KXA 9124	III	IV	I
48	KXA 9141	II	II	I
49	KXA 9142	II	II	IV
50	KXA 8164 (B)	IV	I	IV
51	KXA 9144	IV	III	IV
52	KXA 9145	I	I	I
53	KXA 9146	III	III	IV
54	KXA 9147	I	III	I
55	KXA 9148	I	I	I
56	KXA 9152	I	I	I
57	KXA 9161	III	IV	II
58	KXA 9171	I	II	I
59	KXA 9172	III	IV	IV
60	KXA 9173	III	IV	III
61	KXA 9181	IV	II	IV
62	KXA 9191	IV	IV	IV
63	RICARDINIO (B)	IV	III	III
64	KXA 8151 (B)	IV	I	III
# of entries	GROUP I	24	18	22
	GROUP II	8	15	9
	GROUP III	10	21	11
	GROUP IV	22	10	22

**Table 5.10. Comparison of the grand mean CV of conventional and organic farming systems for 3 traits in grain maize during 2009. N=64 genotypes**

Trait	CV of the Genotypes (%)	Overall	Farming System		Location			Environment					
			CON	ORG	Einbeck	Niederalteich	Kaufering	EIK	WIO	NAK	NAO	KAK	KAO
GFM	Grand mean	<b>20.22</b>	<b>7.74</b>	<b>24.92</b>	<b>6.06</b>	<b>13.75</b>	<b>32.45</b>	<b>4.00</b>	<b>4.18</b>	<b>6.18</b>	<b>10.70</b>	<b>5.27</b>	<b>19.20</b>
	Max	34.28	17.97	47.58	13.94	30.97	63.56	10.55	19.36	26.70	36.06	16.05	46.45
	Min	11.70	3.98	12.57	1.57	3.73	14.12	0.30	0.15	0.67	1.61	0.13	1.01
Grain Yield	Grand mean	<b>21.20</b>	<b>8.26</b>	<b>24.69</b>	<b>6.23</b>	<b>15.38</b>	<b>34.10</b>	<b>4.11</b>	<b>4.33</b>	<b>6.01</b>	<b>11.12</b>	<b>5.34</b>	<b>19.40</b>
	Max	33.87	16.59	46.10	15.67	32.80	64.72	10.35	20.78	26.11	36.60	15.25	48.11
	Min	10.75	3.98	12.57	1.97	5.78	14.40	0.06	0.31	0.65	0.25	0.84	0.91
GDCn1	Grand mean	<b>6.96</b>	<b>6.73</b>	<b>6.98</b>	<b>2.57</b>	<b>2.23</b>	<b>2.38</b>	<b>0.55</b>	<b>0.86</b>	<b>0.82</b>	<b>1.01</b>	<b>0.92</b>	<b>1.62</b>
	Max	9.19	8.66	9.84	4.47	3.88	4.27	1.27	2.05	2.20	2.18	2.06	3.70
	Min	5.24	5.38	4.89	0.89	0.95	1.12	0.12	0.06	0.12	0.26	0.08	0.25

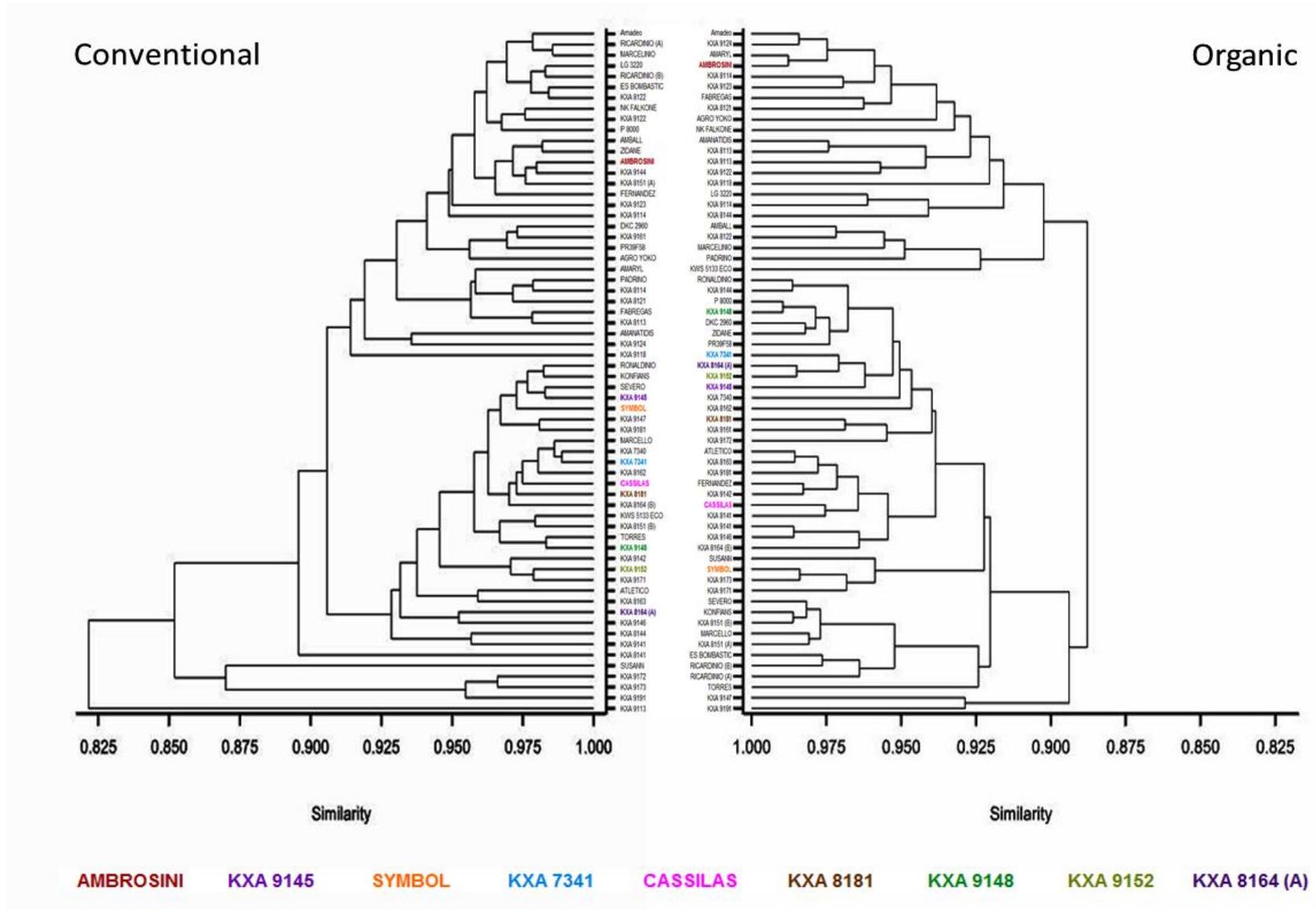
Max = maximum genotype CV, Min = minimum genotype CV

### 5.1.7. Hierarchical Cluster Analysis

Finally, a comparison of conventional and organic dendrograms was elaborated in order to support the results found in  $r_{GCON-ORG}$  and to help answering the research question 1 (the utility of testing conventional varieties under organic conditions for recommendation of organic varieties). For that, cluster analysis were performed under conventional and organic conditions.

With help of a Group Average Agglomerative Cluster analysis based on data for the three grain traits used in this study and collected in trials under conventional and organic farming conditions, respectively resulted in dendrograms for both farming systems (Figure 5.6). The 9 common Group I genotypes in organic and conventional farming for the trait grain yield in the stability analysis (AMBROSINI, CASSILAS, SYMBOL, KXA 7341, KXA 8164 (A), KXA 8181, KXA 9145, KXA 9148 and KXA 9152) were presented in color in the dendrograms to facilitate the comparison between farming systems. Figure 5.6 shows that under conventional conditions the similarity between genotypes was at least 82.5 %, under organic conditions the similarity between genotypes was at least 88.75%. Considering the 9 common Group I genotypes, it was clear that two major clusters were formed under both farming conditions, one containing AMBROSINI (upper cluster) and a second one containing the remaining 8 Group I genotypes (lower cluster). However, such clusters comprised different number of genotypes under each farming condition: the dendrogram for conventional conditions showed 31 entries in the upper cluster and 33 entries in the lower cluster; the dendrogram for organic conditions showed 23 entries in the upper cluster and 41 in the lower cluster. The upper clusters for both farming conditions shared 20 entries and the lower clusters shared 30 entries. These results showed in general that the structure of the clusters within the farming system, and the similarity in the clusters between farming systems considerably varied, which produced asymmetric dendrograms when facing both farming systems.

Figure 5.6. Comparison of dendrograms between conventional and organic farming conditions considering 3 traits (GFM, Grain Yield and GDCn1) for the similarity matrix. N=64 genotypes



## 5.2. Silage Maize

### 5.2.1. Varietal Performance

To answer the research question 1 (the utility of testing conventional varieties under organic conditions for recommendation of organic varieties), individual ANOVAs per environment and combined ANOVAs overall environments, per farming systems and per location were performed to observe the significance of the genotype and GxE interaction effects, analyse the mean differences among farming systems and environments, and calculate genetic parameters under organic and conventional farming systems per each trait in study.

Individual ANOVAs per environment (Table 5.11) show that there were highly significant ( $p < 0.001$ ) differences between the entries in each environment for the three traits under study. For a detailed list of mean performances of genotypes in each environment consult Annex 4.

**Table 5.11. Summary of statistical analysis per environment for 3 traits in silage maize during 2010. N=49 genotypes**

Environment	Trait	TFM					Silage Yield					TDCn2				
	Source of variation	d.f.	m.v.	s.s.	m.s.	F pr.	d.f.	m.v.	s.s.	m.s.	F pr.	d.f.	m.v.	s.s.	m.s.	F pr.
EIS	Genotype	48		4177.0	87.0	<.001	48		131.0	2.7	<.001	48		713.7	14.9	<.001
	Residual	96		571.1	5.9		96		58.7	0.6		96		106.0	1.1	
	Total	146		4840.3			146		194.1			146		825.5		
WSO	Genotype	43	-5	5956.3	138.5	<.001	43	-5	233.4	5.4	<.001	48		563.0	11.7	<.001
	Residual	82	-14	1298.2	15.8		82	-14	117.5	1.4		96		61.6	0.6	
	Total	127	-19	7089.3			127	-19	338.3			146		626.2		
KAS	Genotype	48		9433.6	196.5	<.001	48		195.5	4.1	<.001	48		1082.8	22.6	<.001
	Residual	96		558.3	5.8		96		42.9	0.4		96		58.9	0.6	
	Total	146		10020.8			146		240.5			146		1142.7		
KSO	Genotype	44	-4	4330.8	98.4	<.001	44	-4	200.3	4.6	<.001	48		367.6	7.7	<.001
	Residual	82	-14	2244.0	27.4		82	-14	159.3	1.9		96		68.7	0.7	
	Total	128	-18	10270.8			128	-18	580.0			146		479.9		

d.f. = degrees of freedom, m.v. = missing values, s.s. = sum of squares, m.s. = mean square, F pr = probability value for the F test

The combined ANOVAs over all environments (Table 5.12) indicate that the main factor Genotype and GxE interaction were highly significant ( $p < 0.001$ ) for the three traits under study.

**Table 5.12. Summary of statistical analysis across 4 environments for 3 traits in silage maize during 2010. N=49 genotypes**

Trait	TFM					Silage Yield					TDCn2				
Source of variation	d.f.	m.v.	s.s.	m.s.	F pr.	d.f.	m.v.	s.s.	m.s.	F pr.	d.f.	m.v.	s.s.	m.s.	F pr.
Genotype	48		19055.2	397.0	<.001	48		473.1	9.9	<.001	48		2185.0	45.5	<.001
GxE	135	-9	5180.1	38.4	<.001	135	-9	298.3	2.2	<.001	144		542.2	3.8	<.001
Residual	356	-28	4671.6	13.1		356	-28	378.4	1.1		384		295.3	0.8	
Total	550	-37	49153.4			550	-37	4562.4			587		4980.7		

d.f. = degrees of freedom, m.v. = missing values, s.s. = sum of squares, m.s. = mean square, F pr = probability value for the F test

Tables 5.13 and 5.14 summarize the overall means as well as the mean performance of entries for the three silage maize traits per environment and farming system and the corresponding statistical analyses. The first table also shows the highest and lowest entry means. The performance for silage maize traits were on average high, with a substantial differences between environments and farming systems. Table 5.13 shows that the trait means under conventional conditions were significantly ( $p < 0.05$ ) higher than the corresponding trait means under organic conditions. For TFM and silage yield 5.4 Kg/plot and 2.5 Kg/plot superior respectively. For TDCn2 the difference was of 1.8%. The mean range was larger in the conventional farming system in all of the three traits.

**Table 5.13. Comparison of the trait means of conventional and organic farming systems for 3 traits in silage maize during 2010. N=49 genotypes**

Trait		Overall	Farming System		Environment			
			CON	ORG	EIS	WSO	KAS	KSO
TFM (Kg/plot)	<b>Mean</b>	<b>51.2</b>	<b>53.8</b>	<b>48.4</b> *	<b>53.6</b>	<b>55.5</b>	<b>54.0</b>	<b>41.4</b> *
	Max	62.8	68.6	57.8	67.8	69.0	71.7	52.1
	Min	41.0	41.7	38.1	43.0	43.8	37.7	29.2
Silage Yield (Kg/plot)	<b>Mean</b>	<b>15.7</b>	<b>16.9</b>	<b>14.4</b> *	<b>17.7</b>	<b>17.4</b>	<b>16.1</b>	<b>11.6</b> *
	Max	17.3	19.1	16.2	19.7	20.4	19.1	13.5
	Min	13.6	14.6	12.3	15.7	14.7	13.5	8.9
TDCn2 (%)	<b>Mean</b>	<b>30.8</b>	<b>31.7</b>	<b>29.9</b> *	<b>33.3</b>	<b>31.5</b> *	<b>30.1</b>	<b>28.4</b> *
	Max	35.0	36.6	33.4	37.7	36.9	36.6	31.2
	Min	26.7	26.8	26.6	28.3	27.3	25.2	24.9

Max = maximum performance, Min = minimum performance, \* Significant difference at p=0.05

Since GxE interactions were significant for the three traits in the combined ANOVA, in order to count with a more detailed analysis and determinate genetic parameters, ANOVAs per farming system and locations were run. Table 5.14 indicates that both genotype and GxE interaction main effect were highly ( $p < 0.001$ ) significant in almost both farming systems and locations. For the trait silage yield, under organic conditions the main effect of the genotype was significant at  $p = 0.001$  and the main effect of GxE interaction was significant at  $p = 0.007$ . In Einbeck for the same trait, the main effect of the genotype was significant at  $p = 0.007$  and in Kaufering the main effect of GxE interaction was significant at  $p = 0.001$ .

**Table 5.14. Summary of statistical analysis of conventional and organic farming systems and per location for 3 traits in silage maize during 2010. N=49 genotypes**

Farming System or Location	Trait Source of variation	TFM					Silage Yield					TDCn2				
		d.f.	m.v.	s.s.	m.s.	F pr.	d.f.	m.v.	s.s.	m.s.	F pr.	d.f.	m.v.	s.s.	m.s.	F pr.
CON	Genotype	48		12145.5	253.0	<.001	48		270.3	5.6	<.001	48		1544.8	32.2	<.001
	GxE	48		1465.1	30.5	<.001	48		56.2	1.2	<.001	48		251.8	5.2	<.001
	Residual	192		1129.4	5.9		192		101.6	0.5		192		164.9	0.9	
	Total	293		14873.0			293		636.9			293		2689.5		
ORG	Genotype	48		12145.5	253.0	<.001	45	-3	350.1	7.8	0.001	48		800.5	16.7	<.001
	GxE	48		1465.1	30.5	<.001	42	-6	124.2	3.0	0.007	48		130.2	2.7	<.001
	Residual	192		1129.4	5.9		164	-28	276.8	1.7		192		130.3	0.7	
	Total	293		14873.0			256	-37	3141.1			293		1832.7		
Einbeck	Genotype	48		8939.1	186.2	<.001	48		260.0	5.4	0.007	48		1190.6	24.8	<.001
	GxE	43	-5	1378.1	32.1	<.001	43	-5	112.7	2.6	<.001	48		86.1	1.8	<.001
	Residual	178	-14	1869.3	10.5		178	-14	176.2	1.0		192		167.7	0.9	
	Total	274	-19	12249.0			274	-19	537.3			293		1679.5		
Kaufering	Genotype	48		11408.1	237.7	<.001	48		303.4	6.3	<.001	48		1224.4	25.5	<.001
	GxE	44	-4	2457.2	55.9	<.001	44	-4	97.4	2.2	0.001	48		226.0	4.7	<.001
	Residual	178	-14	2802.3	15.7		178	-14	202.2	1.1		192		127.6	0.7	
	Total	275	-18	31132.2			275	-18	2206.7			293		1853.2		

d.f. = degrees of freedom, m.v. = missing values, s.s. = sum of squares, m.s. = mean square, F pr = probability value for the F test

## 5.2.2. Genetic Parameters

Based on the combined ANOVAs across farming systems the variance components were calculated for comparison of parameters under conventional and organic conditions. In Table 5.15 for the trait TFM, the estimate for  $\sigma^2_G$  was slightly higher under conventional than the corresponding one for organic conditions. The estimate for  $\sigma^2_{GE}$  was similar for both farming systems and for  $\sigma^2_e$  was very much higher under organic farming than under conventional farming. In both farming conditions  $\sigma^2_{GE}$  was lower than  $\sigma^2_G$ . The  $\sigma^2_e$  was lower than  $\sigma^2_{GE}$  under conventional conditions and higher under organic conditions. *Rep%* was 11% higher under conventional conditions.

**Table 5.15. Estimates of genetic parameters under conventional and organic conditions for 3 traits in silage maize during 2010. N=49 genotypes**

Parameter	TFM		Silage Yield		TDCn2	
	CON	ORG	CON	ORG	CON	ORG
$\sigma^2_G$	37.09	25.55	0.74	0.80	4.49	2.33
$\sigma^2_{GE}$	8.21	8.04	0.21	0.42	1.46	0.68
$\sigma^2_e$	5.88	21.60	0.53	1.69	0.86	0.68
Rep%	87.9	77.0	79.2	62.0	83.7	83.7

$\sigma^2_G$  = genetic variance,  $\sigma^2_{GE}$  = GxE interaction variance,  $\sigma^2_e$  = error variance, Rep% = repeatability

In the case of silage yield, the estimate for  $\sigma^2_G$  was similar under both farming systems. The estimate for  $\sigma^2_{GE}$  was almost the double, and for  $\sigma^2_e$  was much higher under organic than under conventional conditions, respectively. For both farming systems  $\sigma^2_{GE}$  was lower than  $\sigma^2_G$ . The  $\sigma^2_e$  was higher than the  $\sigma^2_{GE}$  under conventional farming and very much higher under organic farming. Rep% was 17% higher in conventional conditions. For TDCn2, the estimate for  $\sigma^2_G$  and for  $\sigma^2_{GE}$  were almost the double and for  $\sigma^2_e$  was slightly higher under conventional than the corresponding ones for organic farming conditions. For both farming conditions  $\sigma^2_{GE}$  was lower than  $\sigma^2_G$ . The  $\sigma^2_e$  was lower than  $\sigma^2_{GE}$  under conventional conditions and similar under organic conditions for this trait. Rep% was exactly the same under both farming systems.

### 5.2.3. Correlation Coefficients

After the means, variances and genetic parameters were described for each trait, the linear relationship (r) between the three traits under study was assessed in order to support the discussion about these results. Table 5.16 shows that the traits TFM and silage yield were highly significant and positively correlated in both farming systems and over all the environments. On the other hand, traits TFM and TDCn2 were highly significant, negatively correlated. The traits silage yield and TDCn2 indicated in general negative and significant correlation.

**Table 5.16. Estimates of linear correlations between 3 traits in silage maize during 2010. N=49 genotypes**

Farming System	Pearson's Correlation	Traits		
		TFM-Silage Yield	TFM-TDCn2	Silage Yield-TDCn2
CON	r	0.89	-0.92	-0.66
	t pr.	<.001	<.001	<.001
ORG	r	0.91	-0.84	-0.57
	t pr.	<.001	<.001	<.001
Overall	r	0.90	-0.93	-0.68
	t pr.	<.001	<.001	<.001

t pr. = t-approximation test, with approximately a t-distribution on n-2 degrees of freedom

Additionally, in order to answer the research questions 1 (the utility of testing conventional varieties under organic conditions for recommendation of organic varieties), genotypic correlation between the means of the genotypes under conventional and organic conditions ( $r_{GCON-ORG}$ ) was evaluated per trait. Table 5.17 shows that traits TFM and TDCn2 had a highly significant, total correlation between farming systems. For silage yield the correlation between farming systems was high and highly significant.

**Table 5.17. Estimates of correlations between genotypic means with conventional and organic farming conditions for 3 traits in silage maize during 2010. N=49 genotypes**

Trait	Pearson's Correlation	Farming System	Einbeck	Kaufering
TFM	$r_{\text{GCON-ORG}}$ t pr.	0.99 <.001	0.78 <.001	0.78 <.001
Silage Yield	$r_{\text{GCON-ORG}}$ t pr.	0.80 <.001	0.50 <.001	0.67 <.001
TDCn2	$r_{\text{GCON-ORG}}$ t pr.	1.07 <.001	0.91 <.001	0.83 <.001

t pr. = t-approximation test, with approximately a t-distribution on n-2 degrees of freedom

#### 5.2.4. Stability Analysis

In order to answer the research question 5 (the impact of organic testing fields in the efficiency of conventional breeding program) and distinguish stable genotypes, the mean-CV analysis was used. Considering the definition of stable genotype by Francis & Kannenberg (1978), this analysis was focused on identification of the Group I entries: genotypes which, in presence of GxE interaction, showed performances above the group average (high yield) and variation below the group average (low CV%).

Figures 5.7 to 5.9 show Mean-CV plots for silage yield of varieties taking into account all environments (Figure 5.7) and the organic and conventional farming environments, separately (Figures 5.8 and 5.9), in order to reveal differences in stability between the genotypes. Table 5.18 summarizes the varietal groupings with help of each analysis. Group I (stable genotypes) comprised 19 genotypes. The analyses per farming system resulted in 16 Group I genotypes for conventional farming, and also 16 Group I genotypes for organic farming. Only 7 Group I genotypes detected in the latter analyses were identical: BARROS, KXA 9144, KXA 9329, KXA 0141, KXA 0144, KXA 0152 and KXA 0154. This implicates that those genotypes were the most stable under both farming conditions. These varieties also classified as Group I types by the overall analysis. Figures 5.8 and 5.9 revealed that most of the stability in silage yield were due to the modification in the CV of the genotypes, more than to their silage yield performance, which produced in general vertical changes (in the Y-axis). For instance, genotype GROSSO (A) was part of Group I under conventional conditions; however, under organic conditions it increased its CV and still kept its silage yield above the mean, fitting in Group II. KXA 9361, FERNANDEZ and AMARETTO showed similar behavior as GROSSO (A). The opposite was observed in LG3258, under conventional conditions this entry fitted in Group II, however under organic condition, besides it increased its CV, it fitted in Group I. Similar behavior was observed in KXA 0122.

The standard varieties FABREGAS and NK FALCONE were not part of the Group I and were not considered as stable genotypes in any of the analysis performed. Indeed, FABREGAS fitted in Group II and NK FALCONE was part of Group IV in the overall analysis, showing always CV above the group mean and poor stability. The grand mean CV (horizontal dotted line in the graphics) of each analysis were used to demonstrate how variable in average the genotypes were in a specific farming system compared to another, and the same criterion was used to compare locations and environments. For that, Table 5.19 summarize the grand mean CV of genotypes for the three traits and the corresponding highest and lowest genotype CV. For silage yield, Table 5.19 shows a CV of genotypes of 18.3% in the overall analysis, and 7% in conventional environments; on the other hand, the analysis for the organic environments gave a CV that is more than three times the value obtained for the trials under conventional conditions. The range, expressed as the difference between maximum and minimum genotype CVs, was larger in the organic farming system for this trait. The trait TFM showed a similar distribution of the genotype CV as silage yield. In the case of TDCn2, the analysis at any level had similar values in the mean, and the range of the genotype CV. In general there were very low genotype CV values for this trait. These results are in agreement with three facts: the high positive linear correlation between TFM and silage yield, the high and similar *Rep%* of the trait TDCn2 under both conventional and organic farming conditions and its high  $r_{\text{GCON-ORG}}$ , and the variation among locations, environments and between farming systems.

Figure 5.7. Mean-CV plot of varieties for Silage Yield (Kg/plot) based on 2010 trials in all environments (2 farming systems and 2 locations with 3 repetitions)

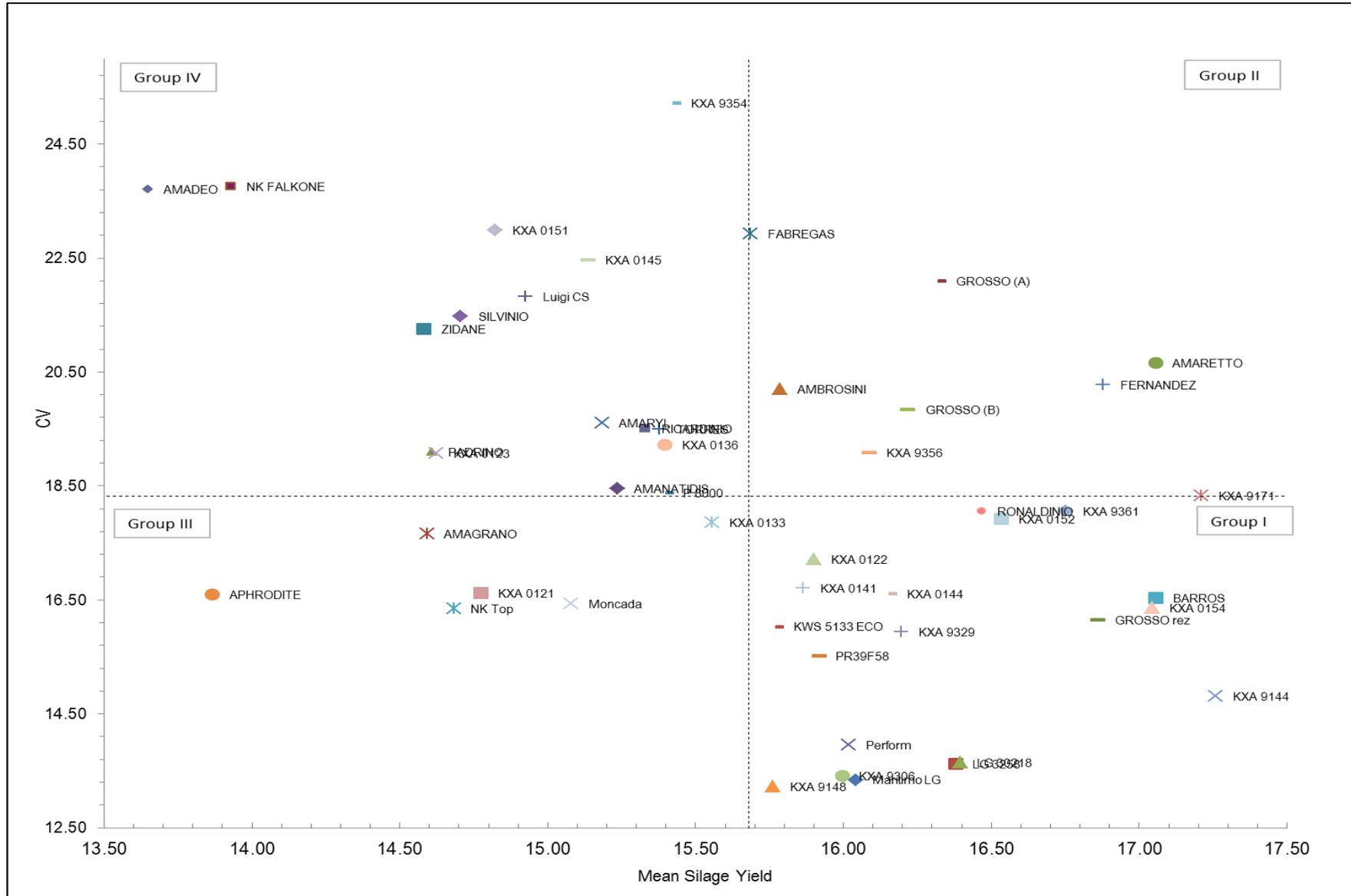


Figure 5.8. Mean-CV plot of varieties for Silage Yield (Kg/plot) based on 2010 trials in conventional environments (2 locations with 3 repetitions)

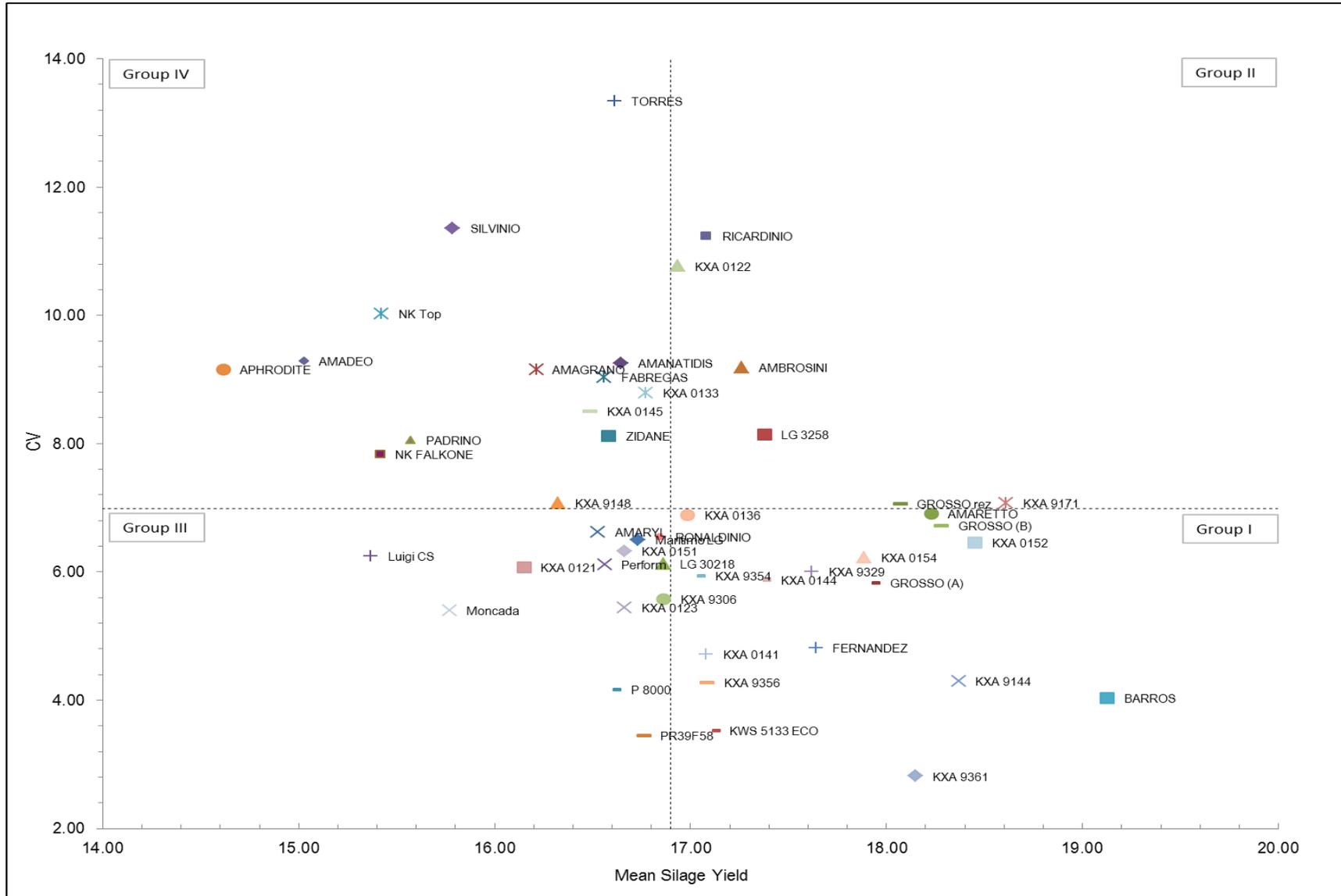
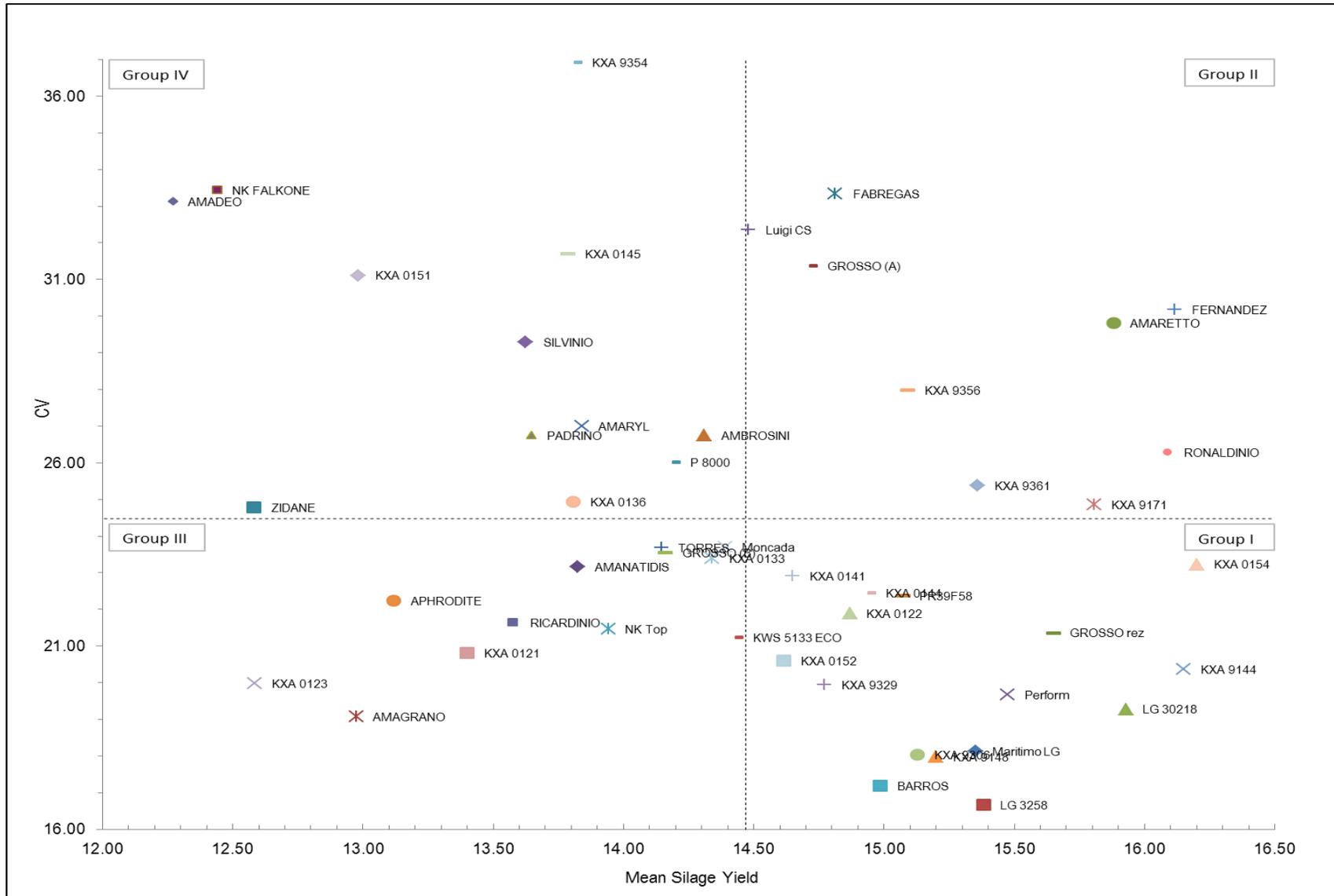


Figure 5.9. Mean-CV plot of varieties for Silage Yield (Kg/plot) based on 2010 trials in organic environments (2 locations with 3 repetitions)



**Table 5.18. Comparison of groups formed for silage yield after Mean-CV analysis of 49 genotypes overall environments, and in conventional and organic farming conditions**

Entry	Denomination	Overall	Farming System	
			CON	ORG
1	AMADEO	IV	IV	IV
2	NK FALKONE	IV	IV	IV
3	PADRINO	IV	IV	IV
4	RICARDINIO	IV	II	III
5	FABREGAS	II	IV	II
6	RONALDINIO	I	III	II
7	TORRES	IV	IV	III
8	GROSSO (A)	II	I	II
9	GROSSO rez	I	II	I
10	AMANATIDIS	IV	IV	III
11	ZIDANE	IV	IV	IV
12	AMBROSINI	II	II	IV
13	AMARYL	IV	III	IV
14	AMAGRANO	III	IV	III
15	AMARETTO	II	I	II
16	Luigi CS	IV	III	II
17	P 8000	IV	III	IV
18	PR39F58	I	III	I
19	Maritimo LG	I	III	I
20	LG 3258	I	II	I
21	LG 30218	I	III	I
22	Perform	I	III	I
23	NK Top	III	IV	III
24	APHRODITE	III	IV	III
25	FERNANDEZ	II	I	II
26	KWS 5133 ECO	I	I	III
27	GROSSO (B)	II	I	III
28	SILVINIO	IV	IV	IV
29	BARROS	I	I	I
30	KXA 9148	I	IV	I
31	KXA 9144	I	I	I
32	KXA 9171	II	II	II
33	KXA 9306	I	III	I
34	KXA 9329	I	I	I
35	KXA 9354	IV	I	IV
36	KXA 9356	II	I	II
37	KXA 9361	I	I	II
38	KXA 0121	III	III	III
39	KXA 0122	I	II	I
40	KXA 0123	IV	III	III
41	KXA 0133	III	IV	III
42	KXA 0136	IV	I	IV
43	KXA 0141	I	I	I
44	KXA 0144	I	I	I
45	KXA 0145	IV	IV	IV
46	KXA 0151	IV	III	IV
47	KXA 0152	I	I	I
48	KXA 0154	I	I	I
49	Moncada	III	III	III
# of entries	GROUP I	19	16	16
	GROUP II	8	6	9
	GROUP III	6	13	12
	GROUP IV	16	14	12

**Table 5.19. Comparison of the grand mean CV of conventional and organic farming systems for 3 traits in silage maize during 2010. N=49 genotypes**

Trait	CV of the Genotypes (%)	Overall	Farming System		Location		Environment			
			CON	ORG	Einbeck	Kaufering	EIS	WSO	KAS	KSO
TFM	Grand mean	<b>15.01</b>	<b>5.97</b>	<b>20.06</b>	<b>6.54</b>	<b>18.86</b>	<b>4.41</b>	<b>5.55</b>	<b>4.16</b>	<b>19.20</b>
	Max	23.46	13.25	33.52	14.79	32.46	11.36	17.92	9.53	38.82
	Min	8.13	2.44	10.63	2.42	10.53	0.51	0.92	1.22	0.40
Silage Yield	Grand mean	<b>18.32</b>	<b>6.99</b>	<b>24.48</b>	<b>5.91</b>	<b>20.78</b>	<b>4.02</b>	<b>5.24</b>	<b>3.69</b>	<b>17.21</b>
	Max	25.22	13.34	36.93	12.60	32.59	11.58	15.57	8.44	33.32
	Min	13.22	2.82	16.66	2.17	12.23	0.71	0.28	0.71	1.40
TDCn2	Grand mean	<b>7.11</b>	<b>6.36</b>	<b>6.50</b>	<b>4.01</b>	<b>4.63</b>	<b>2.84</b>	<b>2.20</b>	<b>2.16</b>	<b>3.11</b>
	Max	10.86	13.80	12.03	7.51	10.05	5.17	5.25	4.50	9.98
	Min	3.97	1.65	2.78	1.70	0.82	0.43	0.46	0.09	0.27

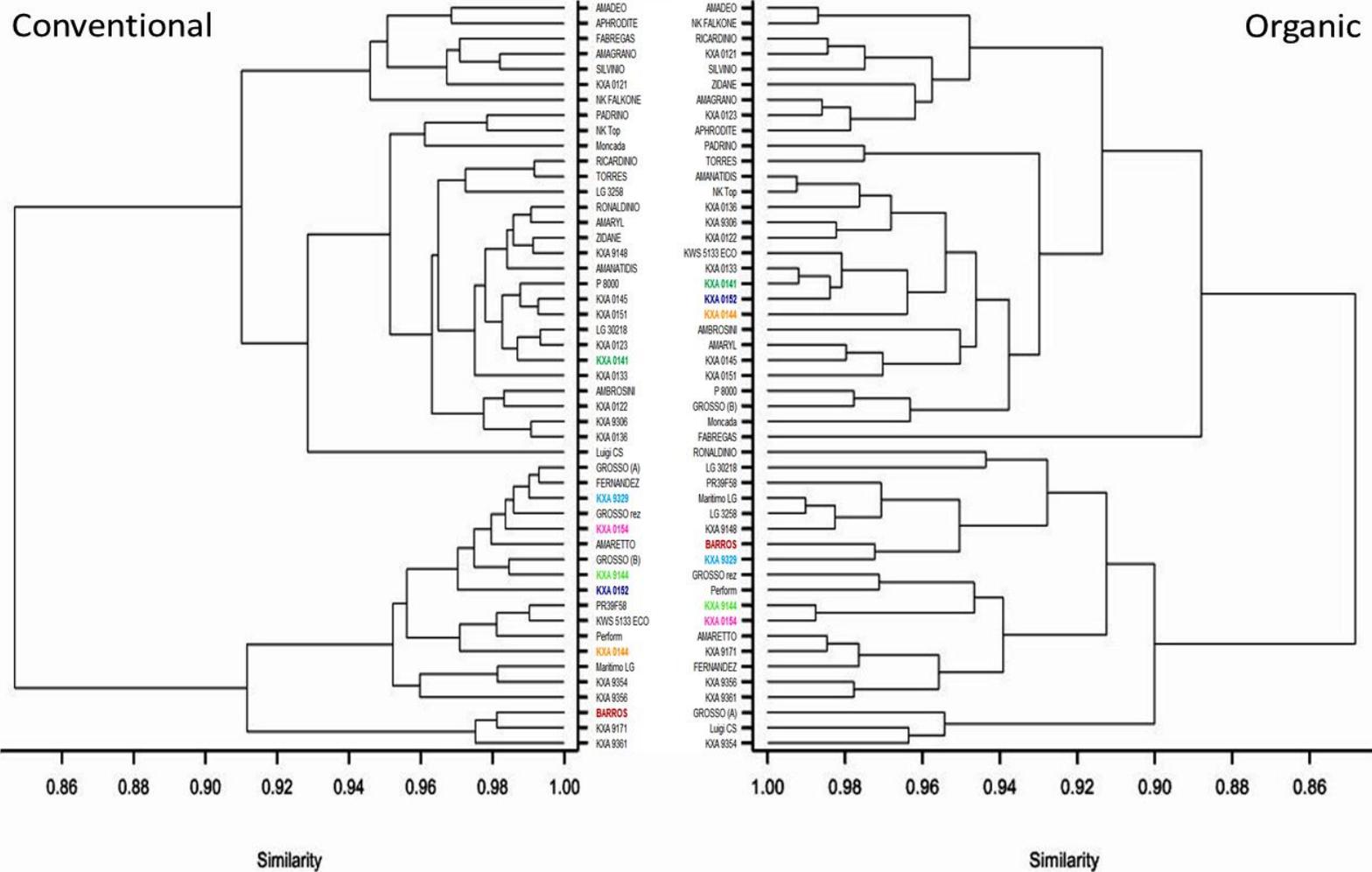
Max = maximum genotype CV, Min = minimum genotype CV

### 5.2.5. Hierarchical Cluster Analysis

Finally, a comparison of conventional and organic dendrograms was elaborated in order to support the results found in  $r_{\text{GCON-ORG}}$  and to help answering the research question 1 (the utility of testing conventional varieties under organic conditions for recommendation of organic varieties). For that, cluster analysis were performed under conventional and organic conditions.

With help of a Group Average Agglomerative Cluster analysis based on data for the three silage traits used in this study and collected in trials under conventional and organic farming conditions, respectively resulted in dendrograms for both farming systems (Figure 5.10). The 7 common Group I genotypes in organic and conventional farming for the trait silage yield in the stability analysis (BARROS, KXA 9144, KXA 9329, KXA 0141, KXA 0144, KXA 0152 and KXA 0154) were presented in color in the dendrograms to facilitate the comparison between farming systems. Figure 5.10 shows that under conventional and organic conditions the similarity between genotypes was at least 84%. Considering the 7 common Group I genotypes, it was clear that two major clusters were formed under both farming conditions. Under conventional conditions one contained KXA 0141 (upper cluster), and the second one contained the remaining 6 Group I genotypes (lower cluster); under organic conditions the upper cluster contained KXA 0141, KXA0152 and KXA 0144, and the lower cluster contained the remaining 4 Group I genotypes. However, such clusters comprised almost the same number of genotypes under both farming condition: the dendrogram for conventional conditions showed 30 entries in the upper cluster and 19 entries in the lower cluster; the dendrogram for organic conditions showed 29 entries in the upper cluster and 20 in the lower cluster. The upper clusters for both farming conditions shared 25 entries and the lower clusters shared 15 entries. These results showed in general that the structure of the clusters within the farming system, and the similarity in the clusters between farming systems did not vary that much, which produced somehow symmetric dendrograms when facing both farming systems.

Figure 5.10. Comparison of dendrograms between conventional and organic farming conditions considering 3 traits (TFM, Silage Yield and TDCn2) for the similarity matrix. N=49 genotypes



KXA 0141 KXA 9329 KXA 0154 KXA 9144 KXA 0152 KXA 0144 BARROS

## VI. DISCUSSION

This study was based on VCU Test data; however, in order to get conclusive results 3 traits for grain maize and 3 traits for silage maize were not enough. Additionally, the data were only for one year testing for each grain maize (2009) and silage maize (2010), and repetition through years was missing. In order to develop the discussion, different studies were consulted and referred, most of them about comparative testing in grain maize, which presented genetic parameters for different traits under conventional and organic conditions (Annex 7); especially to answer the research question 3 (the relevant traits for organic farming systems). Comparative studies for silage maize were not known. Other studies during the discussion include literature about VCU Test, especially to answer the research question 4 (the appropriateness of VCU testing protocol for evaluating organic maize varieties), cereal breeding, maize breeding and breeding for low-input/organic conditions. Therefore, to answer research questions 1 to 5 the results from this study, literature review and consultation with maize experts was employed. Research questions 6 and 7 (adapting maize breeding program to organic agriculture) are the application of the lessons learned from testing conventional varieties under organic conditions to recommend varieties for organic agriculture.

- 6.1. How useful are the results of testing conventional varieties under organic conditions for recommendation of organic varieties with respect to: means, components of variance, repeatability (heritability), and genotypic mean correlation between conventional and organic trials?

**MEANS:** the difference between conventional and organic farming means varies according to the trait (Tables 5.3 and 5.13). In general, grain yield, GFM, GDCn1 (grain maize test); silage yield, TFM and TDCn2 (silage maize test) have all significant reduction under organic conditions. These results for grain yield, agree with Burger *et al.* (2008) where they found 8.4% to 21.4% of reduction, with Messmer *et al.* (2010) where they found 3% to 18% of reduction, and with Brandes (2011) where he found 2.9% in Einbeck to 66% in Kaufering of reduction under organic conditions. Presterl *et al.* (2003) found a reduction from 14.5% to 55.4% in grain yield under low-N conditions compared with high-N conditions. However, Lorenzana & Bernardo (2008) did not find significant differences between the mean grain yields across locations in the organic and conventional systems, justifying this lack of difference with the favourable environment conditions and the heterogeneity of the genetic material employed in the study. For GDCn1, these results are in agreement with Burger *et al.* (2008) and Messmer *et al.* (2010), where they stated that in general GDCn1 content at harvest was slightly lower under organic than under conventional conditions. Additionally, Brandes (2011) found a reduction in the range of 2% to 4% for GDCn1, and Lorenzana & Bernardo (2008) found significant increase in grain moisture (opposite trait to GDCn1) under organic conditions.

**GENETIC VARIANCE ( $\sigma^2_G$ ):** all estimates of  $\sigma^2_G$  are significant for the 6 traits in study (Tables 5.2 and 5.12). In grain maize: trait GDCn1 presents similar estimates under both farming systems, in GFM and grain yield organic farming presents slightly higher to higher estimates than conventional ones (Table 5.5). In silage maize: for silage yield, the estimates of  $\sigma^2_G$  are similar under both farming systems, however TFM and TDCn2 show slightly higher and higher estimates under conventional than under organic conditions, respectively (Table 5.15). For grain yield, these results agree with Goldstein *et al.* (2012) who indicated that the variance between the entries in organic trials tend to be higher; and with Presterl *et al.* (2003), who observed that on average, components of  $\sigma^2_G$  were 2.3 times higher at low-N than at high-N conditions. However, Burger *et al.* (2008) indicated that  $\sigma^2_G$  were of similar size under both farming systems. For GDCn1, Presterl *et al.* (2003) found that  $\sigma^2_G$  were on average, 43% higher at low-N than at high-N conditions.

**GxE INTERACTION VARIANCE ( $\sigma^2_{GE}$ ):** all estimates of  $\sigma^2_{GE}$  are significant for the 6 traits (Table 5.2 and 5.12). In grain maize, under organic farming conditions  $\sigma^2_{GE}$  is around twice larger than under conventional conditions for the traits grain yield, GFM, and GDCn1 (Table 5.5). This supports the idea that GxE interactions are most important in extreme conditions (Wolfe *et al.*, 2008) and is in agreement with Presterl *et al.* (2003) who found higher  $\sigma^2_{GE}$  at the low-N conditions compared to high-N conditions for grain yield and GDCn1. For grain yield, under both farming conditions,  $\sigma^2_{GE}$  is larger than  $\sigma^2_G$ , which agrees with Burger *et al.* (2008) who found that the  $\sigma^2_{GE}$  was 1.5 to 3 times larger than the  $\sigma^2_G$  in most experiments, however, they also reported that there was good agreement between conventional and organic for  $\sigma^2_{GE}$ , which is not the case in this study. For GFM  $\sigma^2_{GE}$  is similar to the  $\sigma^2_G$  under conventional farming systems, but slightly higher under organic conditions. In the case of

GDCn1  $\sigma^2_G$  is always larger than the  $\sigma^2_{GE}$  under both conventional and organic conditions. In silage maize (Table 5.15): the  $\sigma^2_{GE}$  is around twice larger than under conventional conditions for silage yield. TFM shows similar  $\sigma^2_{GE}$  for both systems, and TDCn2 presents almost the double  $\sigma^2_{GE}$  under conventional than under organic conditions. Under conventional and organic conditions, the three traits present lower  $\sigma^2_{GE}$  than  $\sigma^2_G$ .

According to the variance components (Tables 5.5 and 5.15), for traits GFM and grain yield the variability is explained mostly for the GxE interaction. On the other hand, traits GDCn1, TFM and TDCn2 present variability mostly due to the genotypes than to the GxE interaction. The trait silage yield, in a medium point, is partially influenced by both genetic and GxE interaction factors.

For GFM and grain yield, these results suggest that the reduction in the performances under organic farming system depends mostly on the difference between organic and conventional conditions, which produce larger variance components where the farming system controls less the environment: under organic conditions, and usually larger  $\sigma^2_{GE}$  than  $\sigma^2_G$  in both farming systems. These results also confirm the high correlation between both traits (Table 5.6) and indicate that the same set of genes control such characteristics. Reversely, trait GDCn1 shows a poor response to the environment, which produce larger  $\sigma^2_G$  than  $\sigma^2_{GE}$  and similar  $\sigma^2_G$  in both farming conditions. Now, considering that the same genotypes were employed in both farming systems and consequently no genotypic variation is expected; then the small, but significant, organic reduction in GDCn1 is explained, at least partially, by the agricultural management and the portion of GxE variation. As management differences (Annex 1) should be noticed that the organic trials were sowed later (WIO), earlier (NAO), or the same date (KAO) and the harvest date was later (WIO and NAO) or the same date (KAO) than the conventional plots.

Traits TFM and TDCn2 also show in both farming systems larger  $\sigma^2_G$  than  $\sigma^2_{GE}$ , however, the fact that the entries are conventionally bred seems to predispose a better expression of these traits under conventional conditions; consequently, under organic conditions these traits cede some of the variation to the GxE interaction or the experimental error, which produce lower  $\sigma^2_G$ . Finally, trait silage yield is also influenced mostly by the genotypes, however the GxE interaction of cross-over type also influence the expression of this trait, which result in larger variance components under organic conditions and larger  $\sigma^2_G$  than  $\sigma^2_{GE}$  in both farming conditions. Here again, considering that the same genotypes were employed in both farming systems and consequently no genotypic variation is expected, most of the variation observed in the three silage maize traits is explained by the agricultural management and the portion of GxE variation. As management differences (Annex 2) should be noticed that the organic plot were sowed later (WSO) or the same date (KSO) and harvested later (WSO) or earlier (KSO) than the conventional plots. When sowing the same date and harvesting earlier than the conventional environment (like happened in Kaufering) shorter plants are expected in the organic plots at the harvest moment, considering also the deeper sowing of seed.

**ERROR VARIANCES ( $\sigma^2_e$ ):** the error variances (Tables 5.5 and 5.15) for all the traits, except for TDCn2, are larger under organic conditions than under conventional conditions. These results agree with Goldstein *et al.* (2012) who stated that maize grown under organic farming conditions usually reckons with a significantly higher experimental error, and with Presterl *et al.* (2003) who indicated that  $\sigma^2_e$  was increased at the low-N conditions for grain yield. However, Burger *et al.* (2008) found similar magnitude of error variances under organic and conventional conditions in grain yield. Trait TDCn2, in general, presents similar  $\sigma^2_e$  under both conventional and organic farming conditions. The  $\sigma^2_e$  depends on the genetic material and the environments selected for the VCU Test. Additionally, the farming systems obviously have great influence in the  $\sigma^2_e$ ; under organic conditions the experimental error will be larger since plants have to comply with specific stress conditions that do not occur under conventional farming conditions, like the lack of seed treatments, pesticide and mineral N fertilizer applications, or the weed pressure. Moreover, such experimental error will be even larger if the varieties in the VCU Test do not have the special characteristics required to get over those stress.

**REPEATABILITY:** *Rep%* values differ per trait (Tables 5.5 and 5.15): for grain yield and GFM the estimates are low in both farming conditions, for silage yield the estimates are high under conventional conditions and much lower under organic conditions, for GDCn1, TFM and TDCn2 the estimates are high in both farming conditions. These results, for grain yield, agree with Burger *et al.* (2008) when indicated that heritability ( $h^2$ ) coefficients were moderate to low. Brandes (2011) found about 7% lower  $h^2$  under organic environments than under conventional ones, Presterl *et al.* (2003) found that average  $h^2$  were similar at low-N and high-N environments, and Burger *et al.* (2008) obtained on average the

same level of  $h^2$  under organic and conventional conditions. Lorenzana & Bernardo (2008) found  $h^2$  numerically higher in the conventional system than in the organic system. For GDCn1, Brandes (2011) found consistently high to very high  $h^2$  in both conventional and organic test environments in 3 material groups, with partly lower heritability under organic conditions. To calculate  $Rep\%$ , one needs to have the  $\sigma^2_G$  in the numerator and the  $\sigma^2_G$ , the  $\sigma^2_{GE}$  and the  $\sigma^2_e$  in the denominator. These  $Rep\%$  varied greatly per trait according to the variance components. Consequently, those different results indicates that a higher  $\sigma^2_e$ , as it happens under organic conditions in 5 out of 6 traits, does not necessarily mean a lower  $Rep\%$  estimate, especially since the  $\sigma^2_G$  in organic trials also tend to be higher (Goldstein *et al.*, 2012). This is in agreement with Brandes (2011), who found for grain yield absolutely larger experimental error under organic conditions in 3 of his 4 material groups during 2009, but either the  $\sigma^2_G$  or  $\sigma^2_{GE}$  also increased, so that the  $h^2$  were similar in magnitude than under conventional conditions. Burger *et al.* (2008) also found  $h^2$  in the same order under both farming conditions for grain yield; however, in their work the  $\sigma^2_e$  were of similar magnitude in both systems. Additionally, Presterl *et al.* (2003) indicated that the higher  $\sigma^2_G$  at low-N conditions did not result in a higher average estimate of  $h^2$  due to  $\sigma^2_{GE}$  and  $\sigma^2_e$  were also increased in low-N conditions for grain yield and GDCn1.

Trait GFM and silage yield produce lower  $Rep\%$  in organic conditions due principally to the higher  $\sigma^2_{GE}$  and  $\sigma^2_e$  under organic conditions. This implicates the existence of GxE interaction of cross-over type when testing under organic environments. TFM produce lower  $Rep\%$  in organic conditions due to a lower  $\sigma^2_G$  and a higher  $\sigma^2_e$  under organic conditions. This indicates that the entries employed for the VCU test, selected in conventional environments, present a poor average adaptation to low yielding conditions for this trait. Additionally, the trait grain yield shows slightly (7%) higher  $Rep\%$  under organic conditions because the  $\sigma^2_G$  is greater in the organic environment, which counterbalance the increased  $\sigma^2_e$ . These results agree with Ceccarelli (1994), who indicated the  $\sigma^2_G$  may be greater under stress than non-stress conditions and lead to higher heritability under low-input conditions.

GENOTYPIC CORRELATION BETWEEN MEANS ( $r_{GCON-ORG}$ ): all  $r_{GCON-ORG}$  are positive and significant (Tables 5.7 and 5.17). GDCn1, TFM and TDCn2 present  $r_{GCON-ORG}$  of 1, showing close to a linear relationship between the conventional and organic environments for those traits. This implies that the trait is influenced by the same set of genes under both farming conditions. Estimates for the other traits are: 0.80 for silage yield, 0.70 for GFM and 0.51 for grain yield, showing low linear relationship between farming systems. This suggests, at least partially, that a different set of genes influencing grain yield in each farming system. These results agree with Messer *et al.* (2010) who found on average  $r_{GCON-ORG}$  of 0.54 for grain yield and 0.95 for GDCn1 (Goldstein *et al.*, 2012); Brandes (2011) found on average a  $r_{GCON-ORG}$  of 0.6 for grain yield and 1 for GDCn1; and Presterl *et al.* (2003) found  $r_G$  between high-N and low-N conditions on average high for GDCn1 and low for grain yield, indicating also that  $r_G$  for grain yield decreased significantly with increasing levels of N-deficiency stress. Burger *et al.* (2008), found that  $r_{GCON-ORG}$  for grain yield varied greatly between experiments, ranging from 0.07 to 1.66; and estimates for GDCn1 were high to very high, ranging from 0.91 to 1.06. On the other hand, Lorenzana & Bernardo (2008) found  $r_{GCON-ORG}$  of 0.84 for grain yield concluding that the differential response of the genotypes to the farming systems was absent for grain yield. As a remark, it is possible that these results explain the dendrograms comparison obtained after the hierarchical cluster analysis (Figures 5.6 and 5.10) where the grain maize traits show very different dendrograms under conventional and organic conditions, possibly due to the fact that only GDCn1 have a high correlation between both farming systems and the other two traits are poorly correlated. Oppositely, the dendrogram comparison of the silage maize traits is much similar since the three traits are highly correlated in both farming systems.

After all these results being analysed, in order to determinate how useful is recommending organic varieties from conventional varieties tested under organic conditions in the VCU Test, it must be taken into account the traits, the use of conventionally breed varieties, the genetic material and the environments during the evaluation.

For traits like GDCn1 in grain maize, TDCn2 and TFM in silage maize, the environment where the VCU Test is carried out does not have so much relevance for the results, since such traits are not largely influenced by the environment, present high  $Rep\%$  (with lower values under organic conditions) and  $r_{GCON-ORG}$  close to 1; consequently the results from both farming systems for the VCU Test are reliable and predictable. In the case of silage yield, the trait is mostly influenced by the genotype; though GxE interaction of cross-over type and agricultural management are also relevant when determining its value. Under organic farming, usually the planting of silage maize is later and deeper

and the harvesting is later than under conventional conditions (to avoid bird damage and weeds competition). As a consequence, at harvest moment shorter plants are found in the organic plots; however, the feeding value (defined as the proportion of cob in relation to the whole plant) is higher (around 45%) under organic conditions. This characteristic is a very important advantage for organic farmers (Van Eekeren, 2011 personal communication). Then, since the  $r_{\text{GCON-ORG}}$  point of view the environment for testing this characteristic is not so relevant, however, since the nutritional value point of view the farming system matters. Finally, GFM and grain yield are highly influenced by GxE interaction, present low *Rep%* in both farming conditions and medium to low  $r_{\text{GCON-ORG}}$ , respectively; thus results obtained under conventional environments are not repeatable under organic conditions when GxE interaction of cross-over type is observed. For that reason the organic environments are indispensable during the VCU Test.

The fact that varieties bred for conventional farming can be recommended for organic farming is a limitation. Some of these materials present a poor average adaptation to low yielding conditions and can affect the expression of determinate traits under organic conditions. Supposing that some genotypes from a conventional breeding program proves to be good under organic environments every year during the VCU Test, thus those could be indicated as adequate for organic agriculture. Nonetheless, in this situation, the organic market have to depend on the low probability that such genotypes exist, since species and varieties adapted to favourable growing conditions are, in general, not well adapted to stress conditions (Ceccarelli, 1996), due to the lack of a broad range of adaptability to maintain performances in organic environments (Wolfe *et al.*, 2008). Even though, due to economic reasons, for large breeding companies the conventional germplasm is an ideal source to develop organic varieties, which is perhaps acceptable if the organic market is still unprofitable for them.

In general during the VCU test, differences that can be accounted come from genotypes, locations and environments. Entries in the varietal maize test are heterogeneous, including commercial and experimental varieties, which have special use for grain, silage or both purposes, different maturity groups and probably diverse genetic backgrounds, favouring some genotypes during the grain or silage test. From the three locations employed in this experiment, Einbeck is located in the north of Germany (Lower Saxony State) and the other two, Niederalteich and Kaufering, in the south of Germany (Bavaria), consequently weather and edaphic conditions of the regions come into account. Finally, the environments varied from each other mainly for the farming systems and their respective intensity, but also the agricultural field management (fertilizations, crop protection, sowing and harvesting dates, crop rotations, and proximity of the conventional and organic trails) enter into account.

Therefore, the parallel test under conventional and organic environments is useful for traits with low *Rep%* and  $r_{\text{GCON-ORG}}$ ; however, the use of conventionally bred genotypes is a limiting factor for the recommendation of organic varieties in the VCU Test. Additionally, the agricultural management of the environments should be homogenize in order to reduce the differences between locations and account the differences only due to farming systems, especially for traits with high  $\sigma^2_G$ ; and both conventional and organic environments should be characterized and fully defined for the evaluation, especially for traits with high  $\sigma^2_{\text{GE}}$ .

6.2. Is the ranking for performance of varieties the same under both conventional and organic conditions for grain yield and dry matter content?

As showed in Table 5.8, performance testing under conventional and organic conditions does not produce the same ranking for grain yield, whereas the farming system is irrelevant for GDCn1 ranks. The trait grain yield shows a general low rank correlation between farming systems, however in some locations the rank correlation is not even evidenced. This is in agreement with Vlachostergios & Roupakias (2008), who working with lentil varieties indicated that significant differences for GxE interaction in grain yield depends on the variety response in each environment. The ranking plot (Figure 5.1) for grain yield illustrates that the majority of the best genotypes in conventional systems are ranked lower in organic systems and vice versa, nevertheless, genotypes which rank high in both systems can also be observed. These results agree with Murphy *et al.* (2007) in his winter wheat yield study, where he indicates a broad non-system specific adaptation and the potential of selecting varieties able to perform good in both farming systems; and with Vlachostergios & Roupakias (2008) in the lentils study, where they identified genotypes exhibiting broad adaptability and stability over years and culture environments. It has to be noticed that in this maize study, such broadly adapted

genotypes are few and most of the genotypes shows a medium or extreme rank change. Then, for varietal testing the ranking of high yielding genotypes only in conventional systems are untruthful predictors for high yielding genotypes under organic conditions when GxE interaction of cross-over type occurs, therefore, in this situation the yield evaluation have to be done in both systems in order to determinate which varieties are better under conventional, organic or both conditions. In the case of GDCn1, the ranking plot show that most of the genotypes which rank high under conventional conditions, also rank high under organic conditions, however some genotypes can show a moderate change in ranking. Then, in general, most of the best varieties in organic systems also are the best under conventional system for this trait and varietal testing evaluations do not need to be done in both farming systems when the GxE interaction of cross-over type is not observed.

The better explanation for such opposite patterns in both traits is the *Rep%* and the  $r_{\text{GCON-ORG}}$  of the traits. Table 5.5 shows that grain yield has low *Rep%* estimates under both farming conditions, and Table 5.7 shows low  $r_{\text{GCON-ORG}}$  between farming systems overall environments and per location. On the other hand, GDCn1 shows high *Rep%* estimates under both farming conditions, and high  $r_{\text{GCON-ORG}}$  between farming systems overall environments and per location. This is logical because, as discussed in the previous question, the GxE interaction affects in a great sense the grain yield and is not so relevant for the trait GDCn1. It should be observed that the ANOVAs (Tables 5.4) indicate always a highly significant GxE interaction in all the traits, even per farming system or per location. Nevertheless, the fact that GxE interaction is significant is not enough to prove that the ranks change (GxE interaction of cross-over type), since interaction can occur with insignificant genotype changes in ranks (Murphy *et al.*, 2007). Furthermore, the difference in ranking does not imply optimal suitability to organic farming, especially since the entries under evaluation are bred for conventional farming and lack of special features to overcome organic conditions. Here, it is more convenient to test conventionally bred varieties against organically bred varieties under both farming conditions in order to observe varietal performances, ranks and suitability to conventional and/or organic agriculture.

6.3. Which traits are important for organic farming systems? Which of those can be evaluated under conventional conditions and which ones under organic or low-input conditions?

Obviously, yield is the most relevant agricultural character even in conventional or organic agriculture and for grain or silage maize, as it is observed in Table 1.1. Nevertheless, there are also relevant traits for adapting to organic conditions. As a remark, during the stability analysis, it was observed that relevant and common traits in the most stable varieties under both farming systems were low to medium chilling sensitivity, very low to medium lodging, and very low to low susceptibility to stem rot. Nonetheless, these traits were not the only reason to perform adequately under organic or low input conditions. The main required traits for maize varieties to be suitable for organic agriculture can be summarized in five points (Schmidt & Burger, 2008 and 2010):

- Seed: high genetic vigour, germination, and “thousand-grain weight” (TGW).
- Seedling: high weed suppression and competitive ability against weeds and birds (cold tolerance and fast youth development compared with weeds and bird damage).
- High nutrient uptake efficiency (widespread root system, nutrient obtaining by excretion of root exudates or through symbiosis with mycorrhizal fungi).
- Temporary nutrient deficiency stress tolerance (especially to drought stress).
- Resistance to ear and foliar diseases, plague and abiotic stress.

Additionally, the most import traits according to different objectives in organic agriculture for grain and silage maize are (Goldstein, 2011; Van Eekeren, 2011):

- High grain nutritional value: protein content and quality, minerals, vitamins
- Better grain palatability
- Standability
- ‘Resistance’ to transgenic pollen
- Feeding value (digestibility and starch content)

From these characteristics, it has to be recognized that at least half of them have already been improved in conventional breeding programs, meaning that they are of mutual interest for both conventional and organic agriculture. These are in general: kernel weight (TGW), low temperature tolerance (better plant and root growth, tolerance to chilling injury, and early vigour during spring), drought tolerance (leaf rolling, anthesis-silking interval shortened, sterility resistance and decreased

canopy temperature under drought stress), resistance to diseases and plague (especially to first- and second-generation of European corn borer), higher harvest index under biotic stress that induce sterility, resistance to root and stalk lodging, tolerance to soil N deficiency, better recovery from stress, high grain protein content and higher feeding value (plant cell wall digestibility and intake) for silage maize. For a detailed review of the trait improvements for grain and silage maize in USA and EU the reader should consult Duvick (2005) and Barrière *et al.* (2005; 2006). However, some of the methods and techniques that produce fruitful results under conventional conditions cannot be applied in organic fields, since they attempt against the concept of naturalness (Lammerts van Bueren *et al.*, 2002, 2004 and 2006). A clear example in maize is the sharply increased resistance to both generation of European corn borers and two species of rootworm, through transgenic hybrids. The remaining traits required to suit in organic agriculture are obviously of less relevance to conventional farmers, since synthetic inputs can be applied and compensate for the deficiency. These traits are: high genetic vigour, high germination, weed suppression and competition against weeds and birds (fast youth development), widespread root system, excretion of root exudates and mycorrhizal fungi symbiosis for higher nutrient uptake efficiency. Other traits, relevant for specific organic programs, and also ignored by conventional breeding are: high grain mineral and vitamin content and quality, better grain palatability and resistance to transgenic pollen.

Unfortunately,  $h^2$ , genotypic ( $r_G$ ) and phenotypic ( $r_P$ ) correlation coefficients studies for most of the required traits to be suitable for organic agriculture are scarce in maize. For the literature review in parameters for other traits (Annex 7) and the results from this study, it is concluded, according to the  $r_G$  and  $r_P$  estimates, that under conventional conditions, with correlated results in organic conditions, the next trait can be evaluated for maize: grain dry matter content, ear height (related to grain yield), plant height (related to standability and competitions against weeds), total fresh matter, total dry matter content (related to silage yield), and in a less degree N-response (related to nutrient efficiency). Additionally, Goldstein *et al.* (2012) mentioned that correlation between response under both farming systems are also high for the next characteristics: field emergence, cold tolerance, tendency to tillering, early maturity, productivity, resistance to lodging, and diseases resistance. In contrast, traits with low correlation coefficients between either conventional and organic farming systems or high- and low-N-conditions in grain maize are: root lodging, stalk lodging (related to standing ability), stay green (related to kernel weight), grain fresh matter and grain yield; reason for why those traits should be evaluated directly under organic or low input conditions.

#### 6.4. How suitable is the current VCU testing protocol for evaluating organic maize varieties?

DUS evaluation and VCU Test are criteria needed for a variety to be included in the National List of an EU member state. DUS evaluations are very well organized through the EU member states and follow international guidelines. For the contrary, the VCU Test largely differs among member states (Schnock, 2006; Van Waes, 2009). For a detailed description of the German VCU Test the reader should refer Schnock (2006). The traits of cultivation, yield and quality evaluated under the German VCU protocol (Schnock, 2006) are evaluated in the field plots and in the laboratory. During the field plots the next traits are evaluated: chilling sensitivity, tillering ability, silking date, plant height, European corn borer (*Ostrinia nubilalis*), maize smut (*Ustilago maydis*), stalk rot (*Fusarium*), northern leaf blight (*Helminthosporium turcicum*), lodging, number of plants before harvest, and ripeness of leaves (only silage maize). During the laboratory stage the next traits are calculated: for grain maize: grain yield (based on GFM and GDCn1), TGW, share of broken grains; for silage maize: silage yield (based on TFM and TDCn2), starch content, digestibility. For both grain and silage maize: maturity group (early, mid-early, mid-early to late, based on GDCn1 for grain maize and/or TDCn2 for silage maize).

From this list, it can be noticed that cold tolerance, standability, stay green and resistance to the main maize biotic stress are the central characteristics evaluated for the VCU Test in the field stage; during laboratory stage grain and silage yield are assessed, together with quality features for those yields. Such traits are of course relevant under organic conditions as exposed in the previous question. Nonetheless, it is clear that in the case of maize in Germany, even when organic fields are included in the VCU Test, much of the requirements for maize varieties to fit in organic environments (genetic vigour and germination, weed suppression and competitively against weeds and birds, nutrient uptake efficiency and temporary nutrient deficiency stress tolerance) are missing and are not assessed in the varietal testing protocol. Osman *et al.* (2008a) and Micheloni & Plakolm (2007) found the same issue in VCU test for organic cereals and several examples can be found in different EU member states. In such varietal test no attention is paid to variety traits that may result very useful information for variety

choice in organic farming (Micheloni & Plakolm, 2007). Consequently, the absence of varietal tests that makes possible to register varieties better suited to organic conditions, may frustrate breeders who want to select for the organic sector (Osman *et al.*, 2008a).

Another consideration is the choice of the standard varieties for the VCU Test. Normally the standard varieties are chosen between the best new and old varieties in the catalogue or many cultivated stable varieties are used (Van Waes, 2009). Ceccarelli *et al.* (1992), indicated that the reference cultivar is a key factor in interpreting the yields in low-yielding (organic) conditions of cultivars with high yields in high-yielding (conventional) conditions. They proposed that comparison should be done with the top-yielding lines in low-yielding conditions (the best organic lines), since this comparison provide information on how much additional yield gain is possible by selecting directly in low-yielding conditions. For grain maize, the standard varieties used for the Federal Office of Plant Varieties in 2009 were: NK FALCONE (Syngenta's grain/silage variety admitted in 2007), AMANATIDIS (KWS' grain variety admitted in 2008) and RICARDINIO (KWS' grain/silage variety admitted in 2008). For silage maize the standard varieties in 2010 were: NK FALCONE and FABREGAS (KWS' silage variety admitted in 2009). Besides most of these standard varieties are recently admitted modern varieties, at least two of them, RICARDINIO (in grain maize test) and FABREGAS (in silage maize test), are recommended for organic or low-input farming conditions (Schmidt & Burger, 2010). However, any of them were considered as stable genotypes in the stability analyses (Table 5.9 and 5.18) for grain and silage yield.

Therefore, answering the question, the conventional VCU Test protocol is partially suitable for evaluating organic maize varieties. For registration and recommendation of organic varieties, besides the conventional and organic test in parallel, and the inclusion of some standard varieties that perform adequately in low-input conditions; there is a need to introduce key characteristics required for organic farming (listed in the previous question), to be evaluated under organic conditions, in order to determinate if the material is valuable for usage in organic agriculture.

6.5. What is the impact of the incorporation of an organic testing field in conventional breeding program on its efficiency?

As genotypes, also the environments are variable in conventional and organic farming systems. Across the environments employed in grain and silage maize for these VCU Test during 2009 and 2010, large differences were found in terms of performances, correlations and variability. In the stability analysis, from the comparison of grand mean CV of the genotypes (Tables 5.10 and 5.19) for the traits grain and silage yields it is clear that the organic farming system and the organic environments presented more variability than the conventional farming system and the conventional environments respectively, showing that under organic condition the experimental error was higher. Additionally, location Kaufering and environments KAO and KSO presented the highest variability, suggesting that KAO (during 2009) and KSO (during 2010) were the less controlled environment, leading to great mean differences between farming systems in location Kaufering (Table 5.3 and 5.13). On the other hand, location Einbeck presented similar grand mean CV of the genotypes for both farming systems, leading to not significant mean differences between farming systems in Einbeck for this traits. Moreover, the performance correlation (Table 5.7 and 5.17) between conventional and organic farming systems varies per location. This heterogeneity mainly between organic environments indicates that some organic conditions might be more similar to conventional than to other organic environments, as is the case for WIO and EIK in location Einbeck. The stability analysis indicates that for grain yield only 9 out of 64 entries are consistently stable in both farming systems, which implicate that 55 entries responded different depending of the test environments, corroborating the great effect of the GxE interaction in this trait. For silage yield only 7 out of 49 entries are consistently stable in both farming systems. However, Figures 5.8 and 5.9 reveals the pattern of mostly vertical changes (in the CV axis) in this trait, suggesting that the GxE interaction does not influence largely the variation in the entries, which confirms that the genotype effect determinate most of the variation for this trait, and partially the GxE interaction. Thus, for the trait grain yield, with high GxE effect, the genotype vs environmental plot (Figure 5.2) evidenced the GxE cross-over type of interaction according to the best genetic material and the environments employed for the VCU Test, and located the GxE cross-over point, for this set of 64 entries under those 6 environmental conditions, in the range between 10.4 and 11.7 Kg/plot. These results locate environments EIK, WIO, NAO and KAO below the cross-over point and KAK and NAK above this point. Nonetheless, it should be noticed that some entries have a cross-over before this point, others have even more than one cross-over point (genotypes from Group II and

IV in the stability analysis) and other ones probably never have a cross-over interaction (genotypes from Group III in the stability analysis).

Therefore, the inclusion of organic testing fields in conventional breeding programs enables (1) to observe and compare the behaviour of each entry in conventional and organic environments, (2) the location of the GxE cross-over point, and (3) to distinguish stable genotypes under conventional, organic or both conditions; which increase the efficiency of the program when it tries to meet the organic sector. For that reason, breeding programs are recommended to include and characterize organic environments in the breeding procedure for evaluation of traits highly influenced by the GxE interaction, like grain yield. Additionally, the standard varieties should also include organic varieties, bred and selected in organic environments, in order to compare the new varieties below the cross-over point, and designate varieties suitable for organic farming.

#### 6.6. Adaptation of maize breeding program to low input farming conditions.

In order to adapt maize breeding program to organic farming the definition of appropriate and meaningful environments should be considered. According to Ceccarelli (1994), most of the debate between wide and specific adaptation and between the breeding strategies is due to the definition of stress environments, which are often very different. Wolfe *et al.* (2008) indicates the importance to recognize that within organic and conventional agricultures exist several sub-systems based, for instance, on market levels for organic agriculture, and on inputs levels for conventional agriculture. For both farming systems it is relevant to recognise the structure of the systems and the impact of the different inputs employed or not employed and consequently, the VCU Test and the Breeding Programs need to be done in both conventional and organic conditions. In order to identify the range of targeted environments it is necessary to perform thorough GxE studies over years in many different organic environments assessing  $h^2$ ,  $r_{\text{GCON-ORG}}$  and different variance components, plus the inclusion of farming practices per environment. Burger *et al.* (2008), stated that if the same selection intensity is reached under conventional and organic conditions, and the  $h^2$  are the same, the efficiency of indirect selection only depends on the  $r_{\text{GCON-ORG}}$ . A final factor is the economic consideration, since precise information of the parameter estimates depends on the actual cost of organic experiments in a particular environment (Przystalski *et al.*, 2008). Consequently, considering genetic parameters, GxE interaction of cross-over type, and the economic aspects, the maize breeding program to be adapted faces 3 different scenarios:

- A. Whether  $r_{\text{GCON-ORG}}$  is close to 1 and the organic systems are above the cross-over point: Presterl *et al.* (2003) stated that if both conventional and organic farming are equally important for large breeding companies, and assuming that traits have similar  $h^2$  and  $\sigma^2_{\text{G}}$  under both farming systems, combined improvement would be the most effective design as long as  $r_{\text{GCON-ORG}}$  ranges between 0.65 and 1. However, the likelihood of obtaining significant correlation under conventional and organic conditions will depend partly on the nature of the systems under considerations and partly on the interaction of those systems with the environment conditions during the period of observation. Additionally, Burger *et al.* (2008) recognized that if the goal is to select hybrids with adaptation to both farming systems, then organic environments should be included in some part of the testing system. Besides, it may seem this is the perfect situation for large breeding companies, some minor adaptation have to be done to a conventional breeding program in order to breed for the organic sector:
  - a. Parental lines: modern varieties
  - b. Adaptation: wide adapted genotypes. Here, it should be noticed that the term “wide adapted” needs to be specified in relation to the target range environments (Ceccarelli, 1989), in our case adapted to environments above the cross-over GxE point.
  - c. Maize population structure: hybrids
  - d. Selection environment: mainly conventional (indirect selection) with inclusion of organic environments in a later phase of the program.
  - e. Centralised Plant Breeding.
  - f. Evaluation of traits under conventional conditions plus the inclusion of traits required to cope organic farming conditions, if necessary.
- B. Whether  $r_{\text{GCON-ORG}}$  is low, the organic systems are below the cross-over point, and belong to a regional or local market farming; therefore it is not profitable for large breeding companies: Presterl *et al.* (2003), indicated in their study that  $r_{\text{GCON-ORG}}$  for grain yield decrease when GxE

increase in grain yield, and that for  $r_{\text{GCON-ORG}}$  below 0.65 a subdivision of the breeding program for specific adapted varieties will be the most suitable strategy. Additionally, Ceccarelli (1994) defined the cost of wide adaptation in wheat as the yield disadvantage of 10-30% of wide adapted lines when compared with lines selected for specific adaptation. Burger *et al.* (2008), indicated that if the goal is to develop varieties with specific adaptation to organic conditions, then direct selection completely under organic farming conditions is the adequate strategy; and Wolfe *et al.* (2008), indicated that success in local selection depends on a broad parental collection of genetic sources. In this situation, by force, the specific goals should be reached with a different strategy, which implies the organization of the target group and development of an own organic breeding program, considering furthermore that selection for organic farming should be focus in the needs of the whole system, as well as the target goal (Wolfe *et al.*, 2008). The adaptation of the breeding program would be like this:

- a. Parental lines: old varieties, landraces, under-utilized species adapted to low-input conditions, modern varieties.
- b. Adaptation: specific adapted genotypes. In our case, adapted to environments below the cross-over GxE point.
- c. Maize population structure: open pollinated (OP) varieties, modern synthetic OP populations, and alternative hybrids (sister line crosses, triple hybrids, double hybrids, top-crosses between OP populations and inbreds).
- d. Selection environment: exclusively organic (direct selection).
- e. Decentralised Participatory Plant Breeding
- f. Evaluation of traits of relevance only for organic farming and for the specific project in the organic breeding program.

C. Whether  $r_{\text{GCON-ORG}}$  is low, the organic systems are below the cross-over point, and belong to a global commodity farming; therefore it is of economic interest for large breeding companies: it have to be noticed that varieties from conventional breeding programs can fulfil partly the requirements of organic farming. However, better adapted varieties are needed to optimise organic farming systems and to act in accordance with required product quality for organic conditions (Wolfe *et al.*, 2008). In this situation maize varieties should be selected according to the priorities in the farming system, and such decision mainly depends on the magnitude of economic interest in the organic market. A main consideration is that the base of breeding for organic agriculture demands the use of genetic diversity to improve or support the broad range of environments and farmer managements (Wolfe *et al.*, 2008). The adaptation of the breeding program would be like this:

- a. Parental lines: old varieties, landraces, under-utilized species adapted to low-input conditions, modern varieties.
- b. Adaptation: specific adapted genotypes to conventional and organic conditions, or wide adapted genotypes if found. Here “wide adapted” means adapted to environments below the cross-over GxE point.
- c. Maize population structure: hybrids
- d. Selection environment: conventional for conventional adapted hybrids, and organic for organic adapted hybrids (direct selection), or both for wide adapted genotypes, if found.
- e. Centralised Plant Breeding for conventional farming and Centralised Participatory Plant Breeding for organic farming, if possible.
- f. Evaluation of traits with high  $r_{\text{GCON-ORG}}$  under conventional conditions, plus the inclusion and evaluation of traits required to cope organic environments and traits with low  $r_{\text{GCON-ORG}}$  under organic conditions.

6.7. In which phase or the program is better to test under organic conditions?

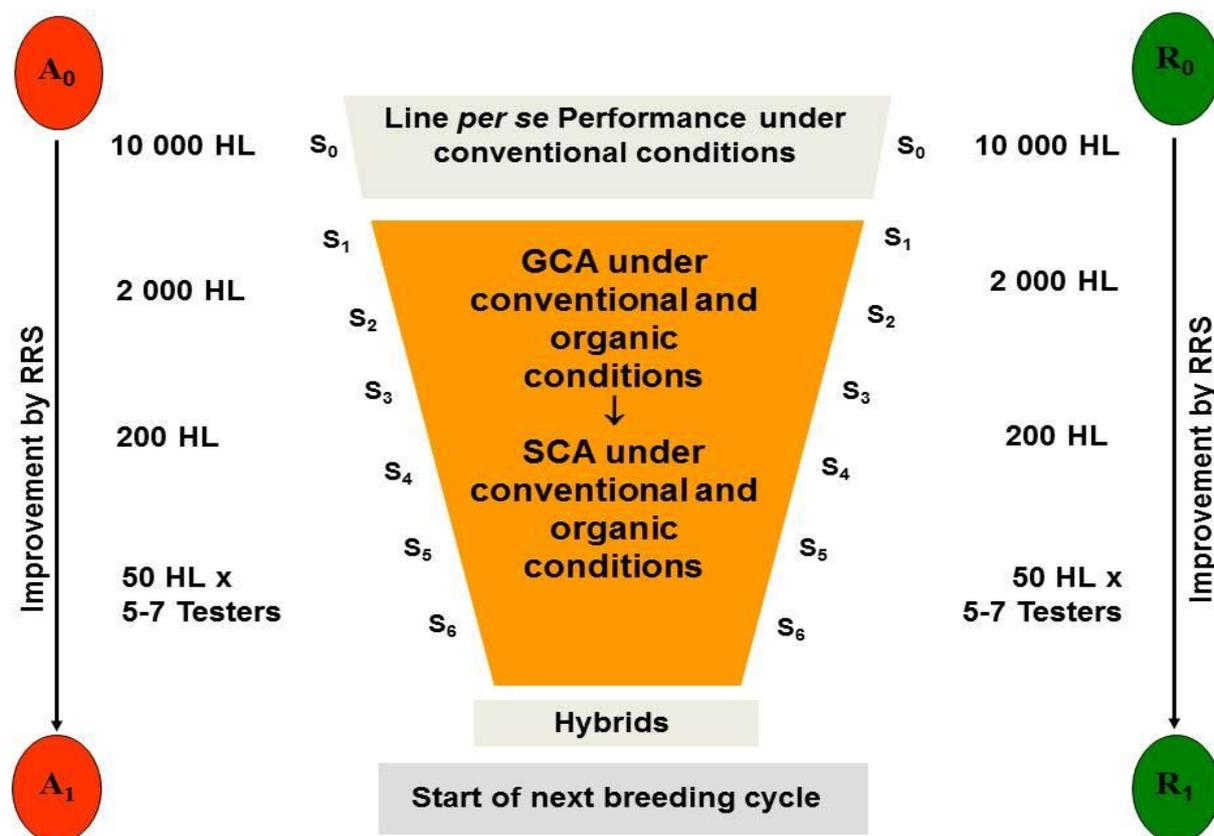
Assuming the most common situation for North-West Europe, when the organic environments are below the cross-over point of the GxE interaction and a conventional breeding program wants to breed organic maize varieties (scenario C in the previous question), then the testing under organic conditions is established according to the  $r_{\text{GCON-ORG}}$  of the characters in question. A classification of the traits according to their response correlation between organic and conventional farming systems is showed in question 3. In order to indicate the organic testing in the maize Breeding Program, the description is based in the production of two-way hybrids, originated from two to four inbred parental lines, which are genetically distant and derived from different heterotic pools. These heterotic pools are maintained and

improved by simultaneous reciprocal recurrent selection (RRS). In general, the Breeding Program consists of 4 steps (Maliepaard, 2011):

1. Development of 10 000 homozygous inbreds lines (HL) per heterotic pool, and visual selection of 2 000 HL among them on Line *per se* Performance.
2. Evaluation of testcrosses with a single tester of the opposite heterotic pool, and selection of 200 HL with superior General Combinatory Ability (GCA) per heterotic pool.
3. Evaluation of factorial crosses with several testers of the opposite heterotic pool among HL with high GCA, selection of 50 superior HL with high Specific Combinatory Ability (SCA) per heterotic pool.
4. These 50 HL are employed to produce superior hybrids with high SCA and GCA. At the same time, these 50 HL are recombined within pools to start a new cycle of RRS.

Figure 6.1 shows an outline of the adapted maize breeding program. The base population  $A_0$  (landraces, under-utilized species adapted to low-input conditions, modern varieties with high degree of heterosis for the traits of interest) becomes  $A_1$  after evaluation with testers from population R and selection of lines with high GCA and SCA. Reciprocally, the starting population  $R_0$  becomes  $R_1$  after evaluation with testers from population A and selection of lines with high GCA and SCA.

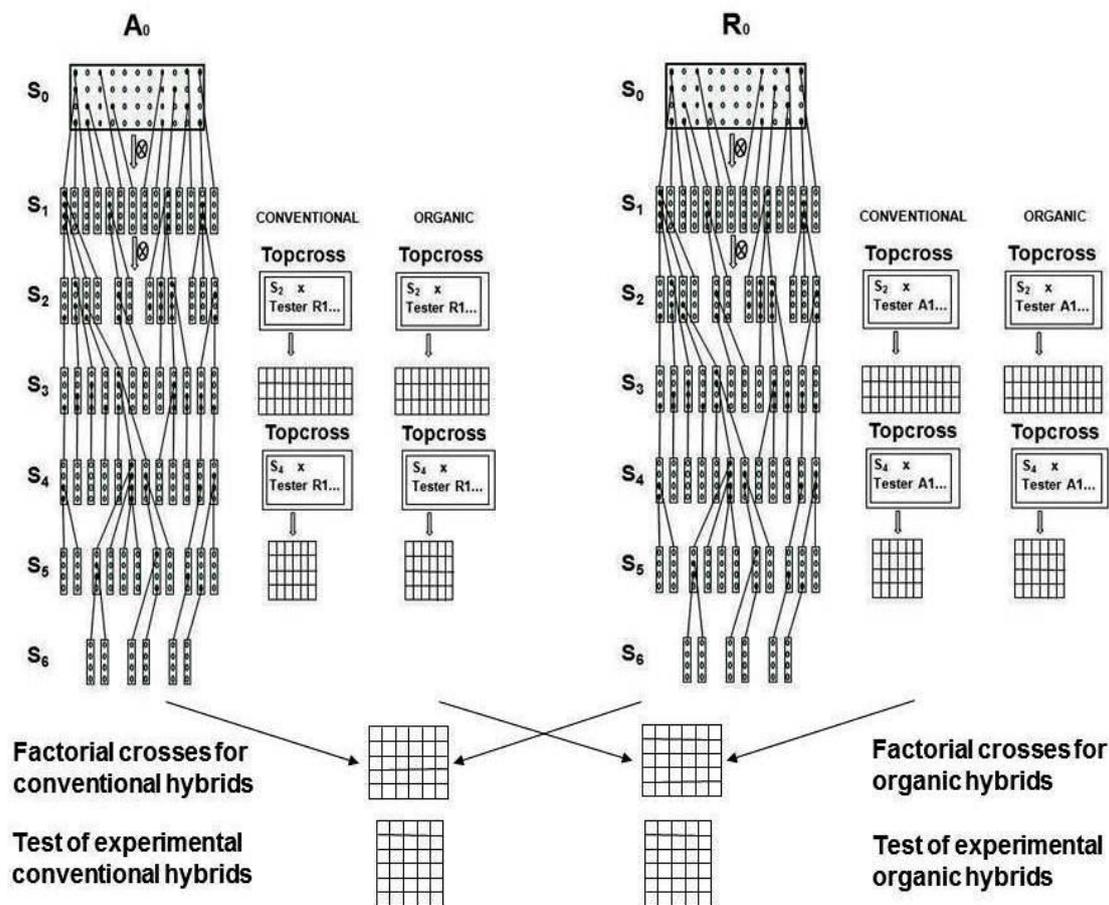
Figure 6.1. Overview of the adapted Maize Breeding Program to low input farming conditions



Adapted from: Schmidt (2008) & Maliepaard (2011)

Figure 6.2 shows that the homozygous inbred lines are developed by continued selfing up to  $S_5$  or  $S_6$  or by double haploids (DH). During the evaluation of lines, testing is done exclusively under conventional conditions during the Line *per se* Performance (Step 1), since the high correlated traits between both conventional and organic farming systems are evaluated in this step. Testing under organic and conventional conditions in parallel begins during the testcross performance: GCA and SCA (Step 2 and 3), since low to medium correlated traits are evaluated in these steps. In this point, the traits required to perform adequately under organic farming should be also assessed and added to the organic evaluation. The testcrosses are made with  $S_1$  or  $S_2$  HL (early testing: GCA) and with  $S_3$  or  $S_4$  HL (SCA + GCA). The experimental hybrids are tested in multi-location trials.

Figure 6.2. Adaptation of Maize Breeding Program to low input farming conditions. The inclusion of organic environments in the breeding scheme



Adapted from: Maliepaard (2011)

Finally, in order to produce hybrid seed, isolated fields are employed, female lines are detasselled (mechanically emasculated) or male sterility is used if possible, and male lines are planted in proportion 1:4 (male:female) with several sowing dates of male rows to ensure pollination of all the female lines.

## VII. CONCLUSIONS

VCU Testing of conventional varieties, including organic environments, taught that:

- 7.1. For traits with high correlation between genotypic means under conventional and organic conditions, testing and/or breeding under conventional or organic conditions produce reliable and predictable performances between both farming systems, which variation is mostly due to genetic factors. In the case of the trait silage yield, also influenced mostly by genetic factors, organic agricultural management is relevant when determining its value.
- 7.2. For traits with low correlation between genotypic means under conventional and organic conditions, like grain yield, the parallel testing under conventional and organic environments and the inclusion of standard varieties that perform adequately in low-input conditions, are convenient when the organic environments are located below the cross-over point of the GxE interaction. However, the evaluation of only conventionally bred genotypes, and the lack of characteristics required to cope organic environments in the evaluation, are limiting factors for the accurate recommendation of organic varieties.
- 7.3. If a conventional breeding program tries to meet the organic sector, it is always necessary to include and characterize organic environments in the breeding scheme during the stage of testcross performance; and evaluate the traits required to deal with organic environments, in order to identify accurately specific adapted organic genotypes.

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## **IX. ANNEX**

**ANNEX 1. Field management, cultural practices and climatic data for grain maize during 2009**

Year	2009					
Location	EIK	WIO	NAK	NAO	KAK	KAO
Elevation (m)	130	165	-	319	600	600
Soil	Clay	Clay	-	Sandy clay	Silty clay	Loess clay
Soil type	Brown earth	-	-	Loess loam	Loess loam	Parabrown earth
Acker number	80	75	-	63	70	70
Soil test	Nov 2008	Sep 2008	-	Mar 2007	Jul 2010	Jan 2008
Humus (%)	-	-	-	-	2.1	-
pH	7.3	6.6	-	7.3	6.0	6.8
P <sub>2</sub> O <sub>5</sub> (mg/100)	18.0	7.9	-	23.0	6.0	10.0
K <sub>2</sub> O (mg/100)	14.0	12.0	-	12.0	10.0	16.0
Mg (mg/100)	4.0	4.5	-	19.0	15.0	11.0
Mineral N content	-	Mar 2009	-	-	-	Mar 2009
0-30 soil layer (Kg/ha)	-	10.0	-	-	-	20.0
30-60 soil layer (Kg/ha)	-	20.0	-	-	-	21.0
60-90 soil layer (Kg/ha)	-	15.0	-	-	-	-
Sum	-	45.0	-	-	-	41.0
Sowing date	25 Apr 2009	04 May 2009	26 Apr 2009	22 Apr 2009	25 Apr 2009	25 Apr 2009
Flowering date	-	-	-	10 Jun 2009	-	-
Harvest date	24 Oct 2009	27 Oct 2009	23 Oct 2009	25 Oct 2009	27 Oct 2009	27 Oct 2009
Notes	-	-	-	Dry sowing	-	Heavy weed pressure
				Average rainfall: 820 mm per annum		It hailed during the 2/3-leaves stage, weakening the plants
				Average annual temperature: 8.1°C		It strongly rained after sowing, provoking siltation.

Crop Rotation											
Year	2004		2005		2006		2007		2008		Location
Main crop	Maize		Maize		Maize		Maize		Maize		EIK
Intercrop											
Main crop					Carrot		Oat		Clover		WIO
Intercrop											
Main crop	Winter wheat		Grain maize		Winter wheat		Grain maize		Winter wheat		NAK
Intercrop		Oil radish				Oil radish				Oil radish	
Main crop	Grain maize		Peas		Grain maize		Sunflower		Green fallow		NAO
Intercrop			(Intercrop failed)						Clover mixture		
Main crop	Winter wheat		Grain maize		Winter wheat		Grain maize		Winter wheat		KAK
Intercrop		Oil radish				Oil radish				Oil radish	
Main crop	Winter wheat		Grain maize		Winter wheat		Beans		Winter wheat		KAO
Intercrop											

Crop Fertilization								
Date	Fertilizer (Commercial name)	Amount	Nutrient (Kg/ha)					Location
			N	P <sub>2</sub> O <sub>5</sub>	K <sub>2</sub> O	CaO	MgO	
01 Nov 2008	40 er Kali	600 Kg			240			EIK
01 Nov 2008	Triple-Phosphat	300 Kg		160				
		Sum		160	240			
								WIO
								NAK
								NAO
04 Apr 2009	Kartoffelfruchtwasserko nzentrat	3000 Kg	69	33	240	3.3	20.4	KAK
22 Apr 2009	Diammonium phosphate (DAP)	200 Kg	36	92				
22 Apr 2009	Urea	200 Kg	92					
		Sum	197	125	240	3.3	20.4	
08 Apr 2009	Kartoffelfruchtwasser	3000 Kg	22	6	95			KAO
25 Apr 2009	Cattle manure	20 m <sup>3</sup>	3.7	1.5	5			
		Sum	25.7	7.5	100			

Soil Preparation		
Date	Device	Location
Nov 2008	Plough	EIK
Apr 2009	Combination	
		WIO
		NAK
05 Nov 2008	Plough furrow at 25 cm.	NAO
09 Apr 2009	Frontal drag + harrow/crumblers combination for seedbed preparation	
21 Apr 2009	Frontal drag + harrow/crumblers combination for seedbed preparation	
29 Apr 2009	<i>Einböck</i> blind tines	
20 May 2009	Becker 6-row hoe with goosefoot shares	
23 May 2009	<i>Einböck</i> currycomb	
04 Jun 2009	<i>Becker</i> 6-row hoe for slight ridging	
Autumn 2008	Plough furrow with packer	KAK
25 Apr 2009	Harrow with crumblers	
-	Plough furrow about 20 cm.	KAO
-	Large drag harrow	
-	Large drag harrow for seedbed preparation	

Crop Protection								
Date	Application	Active ingredient	Stage	Application rate (L/ha)		Jet	Pressure (bar)	Location
				Product	Water			
	Calaris	Terbuthylazin + mesotrione		1.25	250			EIK
	Dual Gold	S-metolachlor		1				
								NAK
25 May 2009	Callisto	Mesotrione	3-leaves	0.75	220	IDN03	3	KAK
25 May 2009	Gardo Gold	Terbuthylazin + S-metolachlor	3-leaves	2.7	220	IDN03	3	
25 May 2009	Boric Acid		3-leaves	3.0 Kg	220	IDN03	3	

Monthly values for Station 10438 Kassel (103 Km S from Einbeck and 94 Km S from Wiebrechtshausen)

STAT	JJJJMM	QN	TXM	TMM	TNM	FMM	NMM	SOS	RSS	TXX	TNN	FXX	RSX
2532	200901	3	0.2	-2.7	-6.2	2.00	-999	67.0	23.3	7.2	-19.0	12.3	10.2
2532	200902	3	4.0	1.1	-1.7	1.90	-999	46.8	62.8	9.6	-9.6	18.2	12.1
2532	200903	3	8.5	4.5	0.8	1.90	5.92	74.7	80.1	14.6	-4.1	15.9	13.6
<b>2532</b>	<b>200904</b>	<b>3</b>	<b>18.7</b>	<b>12.9</b>	<b>7.4</b>	<b>1.90</b>	<b>3.04</b>	<b>225.7</b>	<b>29.8</b>	<b>25.1</b>	<b>2.7</b>	<b>14.1</b>	<b>7.7</b>
<b>2532</b>	<b>200905</b>	<b>3</b>	<b>19.4</b>	<b>14.0</b>	<b>8.5</b>	<b>1.90</b>	<b>4.96</b>	<b>204.8</b>	<b>83.8</b>	<b>28.4</b>	<b>3.7</b>	<b>17.2</b>	<b>15.9</b>
<b>2532</b>	<b>200906</b>	<b>3</b>	<b>20.0</b>	<b>14.9</b>	<b>10.0</b>	<b>1.90</b>	<b>5.20</b>	<b>165.6</b>	<b>52.5</b>	<b>28.2</b>	<b>5.4</b>	<b>14.1</b>	<b>13.4</b>
<b>2532</b>	<b>200907</b>	<b>3</b>	<b>23.6</b>	<b>18.2</b>	<b>13.1</b>	<b>1.80</b>	<b>5.20</b>	<b>205.1</b>	<b>76.8</b>	<b>30.1</b>	<b>9.3</b>	<b>15.5</b>	<b>14.5</b>
<b>2532</b>	<b>200908</b>	<b>3</b>	<b>25.0</b>	<b>18.6</b>	<b>12.5</b>	<b>1.60</b>	<b>3.60</b>	<b>235.9</b>	<b>41.4</b>	<b>34.7</b>	<b>6.8</b>	<b>15.4</b>	<b>10.0</b>
<b>2532</b>	<b>200909</b>	<b>3</b>	<b>19.8</b>	<b>14.5</b>	<b>10.2</b>	<b>1.70</b>	<b>4.80</b>	<b>134.2</b>	<b>54.8</b>	<b>30.0</b>	<b>6.0</b>	<b>14.2</b>	<b>15.2</b>
<b>2532</b>	<b>200910</b>	<b>3</b>	<b>12.0</b>	<b>8.2</b>	<b>4.6</b>	<b>1.80</b>	<b>5.76</b>	<b>81.6</b>	<b>58.9</b>	<b>21.0</b>	<b>-1.5</b>	<b>16.3</b>	<b>14.5</b>
2532	200911	3	10.0	7.7	5.6	2.30	6.72	32.3	89.7	12.5	2.8	16.4	19.5
2532	200912	3	2.7	0.3	-2.4	2.20	6.40	34.4	80.2	9.1	-17.1	14.4	23.1

Monthly values for Station 10788 Straubing (35 Km W from Niederalteich)

STAT	JJJJMM	QN	TNN	TNM	TMM	TXM	TXX	SOS	NMM	RSS	RSX	FMM	FXX
10788	200912	3	-15.8	-3.2	-0.2	2.2	8.5	28.7	5.4	74.8	14.1	2.4	16.2
10788	200911	3	-2.0	1.9	5.8	9.3	14.1	69.8	4.2	58.7	22.5	2.3	16.1
<b>10788</b>	<b>200910</b>	<b>3</b>	<b>-3.3</b>	<b>4.2</b>	<b>8.1</b>	<b>11.9</b>	<b>26.0</b>	<b>80.7</b>	<b>4.9</b>	<b>53.1</b>	<b>12.6</b>	<b>2.0</b>	<b>17.0</b>
<b>10788</b>	<b>200909</b>	<b>3</b>	<b>4.4</b>	<b>9.3</b>	<b>15.3</b>	<b>21.3</b>	<b>28.1</b>	<b>197.0</b>	<b>3.3</b>	<b>37.9</b>	<b>13.3</b>	<b>1.6</b>	<b>15.0</b>
<b>10788</b>	<b>200908</b>	<b>3</b>	<b>5.4</b>	<b>12.7</b>	<b>19.2</b>	<b>25.5</b>	<b>30.0</b>	<b>250.6</b>	<b>3.5</b>	<b>43.5</b>	<b>11.9</b>	<b>1.8</b>	<b>18.7</b>
<b>10788</b>	<b>200907</b>	<b>3</b>	<b>8.4</b>	<b>12.7</b>	<b>18.7</b>	<b>24.5</b>	<b>34.1</b>	<b>221.0</b>		<b>82.6</b>	<b>32.5</b>	<b>1.9</b>	<b>20.2</b>
<b>10788</b>	<b>200906</b>	<b>7</b>	<b>4.6</b>	<b>10.6</b>	<b>16.1</b>	<b>21.4</b>	<b>28.5</b>	<b>230.2</b>		<b>95.5</b>	<b>27.2</b>	<b>2.0</b>	<b>17.7</b>
<b>10788</b>	<b>200905</b>	<b>10</b>	<b>2.0</b>	<b>8.8</b>	<b>14.7</b>	<b>20.3</b>	<b>29.6</b>	<b>235.0</b>	<b>5.3</b>	<b>106.1</b>	<b>30.4</b>	<b>2.0</b>	<b>32.0</b>
<b>10788</b>	<b>200904</b>	<b>10</b>	<b>0.3</b>	<b>5.2</b>	<b>12.6</b>	<b>19.4</b>	<b>23.1</b>	<b>281.6</b>	<b>3.3</b>	<b>54.6</b>	<b>26.7</b>	<b>2.1</b>	<b>14.3</b>
10788	200903	10	-2.7	1.5	4.4	7.8	14.8	78.6	6.8	57.1	7.8	2.4	20.0
10788	200902	10	-15.7	-3.3	-0.4	2.1	9.7	50.3	6.9	44.6	7.4	2.3	21.8
10788	200901	10	-17.6	-8.1	-4.4	-1.2	5.3	75.3	5.4	19.5	8.7	1.5	15.0

Monthly values for Station 10962 Hohenpeißenberg (35 Km S from Kaufering)

STAT	JJJJMM	QN	TNN	TNM	TMM	TXM	TXX	SOS	NMM	RSS	RSX	FMM	FXX
10962	200912	3	-18.4	-3.4	-0.9	2.4	12.1	56.8	6.5	75.1	15.1	2.8	22.2
10962	200911	3	-1.0	3.8	6.8	11.0	19.9	101.6	5.6	71.1	19.6	2.7	24.7
<b>10962</b>	<b>200910</b>	<b>3</b>	<b>-3.7</b>	<b>4.7</b>	<b>7.3</b>	<b>10.9</b>	<b>24.3</b>	<b>111.8</b>		<b>79.6</b>	<b>15.8</b>	<b>2.3</b>	<b>20.3</b>
<b>10962</b>	<b>200909</b>	<b>3</b>	<b>4.9</b>	<b>10.4</b>	<b>13.3</b>	<b>16.9</b>	<b>27.3</b>	<b>168.9</b>	<b>5.0</b>	<b>79.3</b>	<b>13.4</b>	<b>2.1</b>	<b>16.8</b>
<b>10962</b>	<b>200908</b>	<b>3</b>	<b>8.5</b>	<b>13.5</b>	<b>17.7</b>	<b>22.0</b>	<b>28.5</b>	<b>266.1</b>	<b>4.1</b>	<b>73.7</b>	<b>16.2</b>	<b>2.2</b>	<b>15.0</b>
<b>10962</b>	<b>200907</b>	<b>3</b>	<b>5.5</b>	<b>12.1</b>	<b>16.3</b>	<b>20.8</b>	<b>31.0</b>	<b>212.2</b>	<b>4.8</b>	<b>102.1</b>	<b>17.9</b>	<b>3.0</b>	<b>23.9</b>
<b>10962</b>	<b>200906</b>	<b>7</b>	<b>4.6</b>	<b>9.7</b>	<b>12.9</b>	<b>17.0</b>	<b>26.2</b>	<b>163.1</b>	<b>5.9</b>	<b>209.2</b>	<b>37.2</b>	<b>3.3</b>	<b>29.4</b>
<b>10962</b>	<b>200905</b>	<b>10</b>	<b>1.8</b>	<b>8.6</b>	<b>12.7</b>	<b>17.2</b>	<b>26.6</b>	<b>220.2</b>	<b>5.1</b>	<b>142.1</b>	<b>20.9</b>	<b>3.3</b>	<b>31.1</b>
<b>10962</b>	<b>200904</b>	<b>10</b>	<b>-0.3</b>	<b>6.1</b>	<b>10.1</b>	<b>15.1</b>	<b>22.3</b>	<b>237.3</b>	<b>4.4</b>	<b>27.0</b>	<b>9.6</b>	<b>3.3</b>	<b>21.7</b>
10962	200903	10	-6.2	-1.6	0.9	4.0	11.0	80.2	6.9	74.6	12.2	4.1	30.8
10962	200902	10	-11.8	-4.2	-1.7	1.3	12.3	88.0	6.2	70.0	11.2	3.9	36.6
10962	200901	10	-11.1	-5.1	-2.7	-0.0	7.6	103.3	5.2	24.3	8.4	2.9	34.9

Abbreviation	Meaning	Unit
QN	Data quality	---
TNN	Minimum temperature at 2m above ground	Degree Celsius (°C)
TNM	Mean daily minimum temperature at 2m above ground	Degree Celsius (°C)
TMM	Mean temperature at 2m above ground	Degree Celsius (°C)
TXM	Mean daily maximum temperature at 2m above ground	Degree Celsius (°C)
TXX	Maximum temperature at 2m above ground	Degree Celsius (°C)
SOS	Sum of sunshine duration	Hours
NMM	Mean cloud coverage	Oktas
RSS	Total amount of precipitation	mm
RSX	Maximum daily rainfall amount	mm
FMM	Mean wind power	Beaufort Scale (Bft)
FXX	Maximum wind speed at 10m height (peak height)	m/sec

**For Data quality (QN):** **3** Data are for individual (especially automatically measured) elements systematically examined grossly and corrected if necessary; **7** Data that have been tested systematically. All values are provided with quality bytes. Rejected values are marked, but still not corrected in the rule; **10** Data that are systematically checked and corrected or confirmed. The quality of the test corresponds to each time valid procedures

## ANNEX 2. Field management, cultural practices and climatic data for silage maize during 2010

Year	2010			
Location	EIS	WSO	KAS	KSO
Elevation (m)	130	170	600	600
Soil	Clay	Clay	Silty clay	Loess clay
Soil type	Brown earth	-	Loess loam	Parabrown earth
Acker number	80	70	75	70
<b>Soil test</b>	Nov 2008	Sep 2008	2004	Jan 2008
Humus (%)	-	-	2.3	-
pH	7.3	6.7	7.0	6.8
P <sub>2</sub> O <sub>5</sub> (mg/100)	18.0	6.9	16.0	10.0
K <sub>2</sub> O (mg/100)	14.0	17.0	14.0	16.0
Mg (mg/100)	4.0	5.0	16.0	11.0
<b>Mineral N content</b>	-	09 Mar 2010	16 Mar 2010	15 March 2010
0-30 soil layer (Kg/ha)	-	21.0	20.0	26.0
30-60 soil layer (Kg/ha)	-	27.0	26.0	16.0
60-90 soil layer (Kg/ha)	-	34.0	16.0	-
Sum	-	82.0	62.0	42.0
<b>Sowing date</b>	24 Apr 2010	03 May	26 Apr 2010	26 Apr 2010
<b>Flowering date</b>	-	-	-	-
<b>Harvest date</b>	7,8 Oct 2010	12-14 Oct 2010	13 Oct 2010	11 Oct 2010
<b>Notes</b>				Deeper sown against crows and pre-emergent weeding
				Less weed pressure compared with previous year
				Output of the second manure transfer in the series: positive, better utilization

Crop Rotation											
Year	2005		2006		2007		2008		2009		Location
Main crop	Maize		Maize		Maize		Maize		Maize		EIS
Intercrop											
Main crop	Wheat		Winter barley		Clover		(Kart) Potato???		Winter barley		WSO
Intercrop									Legume mixture		
Main crop	Winter wheat		Winter wheat		Potato		Winter wheat		Winter wheat		KAS
Intercrop				Oil radish		Oil radish					
Main crop	Winter barley		Beans		Winter wheat		Spring barley		Rye		KSO
Intercrop											

Crop Fertilization								
Date	Fertilizer (Commercial name)	Amount	Nutrient (Kg/ha)					Location
			N	P <sub>2</sub> O <sub>5</sub>	K <sub>2</sub> O	CaO	MgO	
01 Nov 2009	40 er Kali	600 Kg			240			EIS
01 Nov 2009	Triple-Phosphat	300 Kg		160				
		Sum		160	240			
	No organic fertilization							WSO
12 Mar 2010	Kartoffelfruchtwasserkonzentrat	4000 Kg	92	44	320	4.4	27.2	KAS
27 Mar 2010	Diammonium phosphate (DAP)	180 Kg	32.4	82.8				
27 Mar 2010	Urea	150 Kg	69					
		Sum	193.4	126.8	320	4.4	27.2	
20 Apr 2010	Cattle manure between rows	20 m <sup>3</sup>	3.7	1.5	5.1			KSO
25 May 2010	Cattle manure between rows	20 m <sup>3</sup>	3.7	1.5	5.1			
		Sum	7.4	3.0	10.2			

Soil Preparation		
Date	Device	Location
Nov 2009	Plough	EIS
Apr 2010	Combination	
Apr 2010	Plough	WSO
Apr 2010	Flat harrow further processed by INS BB	
13 Nov 2009	Plough furrow with packer	KAS
24 Apr 2010	Harrow with crumblers	
20 Oct 2009	Plough furrow about 20 cm.	KSO
20 Apr 2010	Large drag harrow for manure incorporation and seedbed preparation	

Crop Protection								
Date	Application	Active ingredient	Stage	Application rate (L/ha)		Jet	Pressure (bar)	Location
				Product	Water			
	Laudis	Terbuthylazin + Mesotrione		1.60	250			EIS
	Sucessor T	S-metolachlor		2.4				
05 Jun 2010	Callisto	Mesotrione	4-leaves	0.7	220	IDN03	3	KAS
05 Jun 2010	Gardo Gold	Terbuthylazin + S-metolachlor	4-leaves	1.2	220	IDN03	3	
05 Jun 2010	Dual Gold	S-metolachlor	4-leaves	1.2	220	IDN03	3	
05 Jun 2010	Boric Acid		4-leaves	5.0 Kg	220	IDN03	3	

Monthly values for Station 10438 Kassel (103 Km S from Einbeck and 94 Km S from Wiebrechtshausen)

STAT	JJJJMM	QN	TXM	TMM	TNM	FMM	NMM	SOS	RSS	TXX	TNN	FXX	RSX
2532	201001	3	-1.7	-3.5	-5.8	2.00	-999	22.6	42.2	3.7	-14.8	13.0	4.9
2532	201002	3	2.8	-0.3	-3.5	2.10	6.88	32.2	46.0	13.0	-11.5	22.9	12.1
2532	201003	3	9.3	4.6	0.6	2.20	5.36	134.8	67.4	20.7	-9.5	21.4	18.3
<b>2532</b>	<b>201004</b>	<b>3</b>	<b>15.2</b>	<b>9.4</b>	<b>3.7</b>	<b>2.10</b>	<b>4.08</b>	<b>206.2</b>	<b>12.8</b>	<b>24.2</b>	<b>-1.6</b>	<b>15.3</b>	<b>4.3</b>
<b>2532</b>	<b>201005</b>	<b>3</b>	<b>14.7</b>	<b>10.5</b>	<b>6.6</b>	<b>2.10</b>	<b>6.24</b>	<b>125.5</b>	<b>69.1</b>	<b>25.2</b>	<b>1.6</b>	<b>15.2</b>	<b>11.4</b>
<b>2532</b>	<b>201006</b>	<b>3</b>	<b>22.9</b>	<b>16.8</b>	<b>10.8</b>	<b>1.90</b>	<b>3.44</b>	<b>287.3</b>	<b>60.9</b>	<b>29.1</b>	<b>4.6</b>	<b>22.0</b>	<b>21.6</b>
<b>2532</b>	<b>201007</b>	<b>3</b>	<b>26.7</b>	<b>20.6</b>	<b>14.5</b>	<b>1.70</b>	<b>3.84</b>	<b>275.9</b>	<b>37.0</b>	<b>34.3</b>	<b>8.4</b>	<b>15.3</b>	<b>9.7</b>
<b>2532</b>	<b>201008</b>	<b>3</b>	<b>21.6</b>	<b>16.5</b>	<b>12.3</b>	<b>1.80</b>	<b>5.76</b>	<b>130.8</b>	<b>116.8</b>	<b>29.0</b>	<b>6.3</b>	<b>14.5</b>	<b>22.8</b>
<b>2532</b>	<b>201009</b>	<b>3</b>	<b>17.1</b>	<b>12.4</b>	<b>8.4</b>	<b>1.80</b>	<b>-999</b>	<b>117.9</b>	<b>54.3</b>	<b>22.7</b>	<b>4.3</b>	<b>14.8</b>	<b>23.1</b>
<b>2532</b>	<b>201010</b>	<b>3</b>	<b>12.7</b>	<b>8.4</b>	<b>4.9</b>	<b>1.90</b>	<b>5.12</b>	<b>104.4</b>	<b>24.9</b>	<b>21.9</b>	<b>-0.6</b>	<b>13.1</b>	<b>6.8</b>
2532	201011	3	7.2	5.2	2.9	2.10	7.36	19.9	67.2	16.1	-7.3	19.5	18.5
2532	201012	3	-1.5	-3.7	-6.6	2.30	-999	14.5	62.5	6.1	-12.7	16.6	10.0

Monthly values for Station 10962 Hohenpeißenberg (35 Km S from Kaufering)

STAT	JJJJMM	QN	TNN	TNM	TMM	TXM	TXX	SOS	NMM	RSS	RSX	FMM	FXX
10962	201012	3	-10.7	-4.9	-2.5	0.5	14.1	54.5	6.9	69.6	12.7	2.7	24.4
10962	201011	3	-7.5	0.7	3.1	6.1	18.2	89.2	6.3	45.4	7.2	3.1	32.0
<b>10962</b>	<b>201010</b>	<b>3</b>	<b>-1.5</b>	<b>4.4</b>	<b>6.8</b>	<b>9.9</b>	<b>22.9</b>	<b>110.6</b>	<b>5.7</b>	<b>62.4</b>	<b>13.0</b>	<b>2.4</b>	<b>15.1</b>
<b>10962</b>	<b>201009</b>	<b>3</b>	<b>3.1</b>	<b>7.6</b>	<b>10.7</b>	<b>14.4</b>	<b>21.0</b>	<b>148.5</b>	<b>5.5</b>	<b>67.3</b>	<b>19.7</b>	<b>2.2</b>	<b>16.6</b>
<b>10962</b>	<b>201008</b>	<b>3</b>	<b>4.7</b>	<b>11.4</b>	<b>14.6</b>	<b>18.9</b>	<b>27.2</b>	<b>147.9</b>	<b>6.0</b>	<b>242.4</b>	<b>57.3</b>	<b>2.5</b>	<b>23.3</b>
<b>10962</b>	<b>201007</b>	<b>3</b>	<b>8.7</b>	<b>13.6</b>	<b>17.7</b>	<b>22.2</b>	<b>29.4</b>	<b>251.5</b>	<b>4.6</b>	<b>188.0</b>	<b>46.2</b>	<b>2.2</b>	<b>15.9</b>
<b>10962</b>	<b>201006</b>	<b>3</b>	<b>3.6</b>	<b>11.1</b>	<b>14.3</b>	<b>17.6</b>	<b>30.5</b>	<b>194.0</b>	<b>5.2</b>	<b>184.6</b>	<b>63.7</b>	<b>2.3</b>	<b>15.0</b>
<b>10962</b>	<b>201005</b>	<b>3</b>	<b>1.1</b>	<b>5.6</b>	<b>8.5</b>	<b>12.3</b>	<b>23.9</b>	<b>97.2</b>	<b>6.9</b>	<b>128.9</b>	<b>19.8</b>	<b>2.5</b>	<b>18.1</b>
<b>10962</b>	<b>201004</b>	<b>3</b>	<b>-2.8</b>	<b>3.1</b>	<b>7.1</b>	<b>12.1</b>	<b>24.1</b>	<b>209.8</b>	<b>4.7</b>	<b>38.9</b>	<b>12.8</b>	<b>2.5</b>	<b>15.4</b>
10962	201003	3	-12.8	-1.2	2.0	6.7	23.1	150.4	5.9	31.0	7.8	3.1	23.7
10962	201002	3	-12.3	-4.0	-1.2	2.0	11.4	86.7	6.6	42.5	9.9	2.6	23.0
10962	201001	3	-11.6	-6.9	-4.6	-2.1	6.5	66.4	6.4	42.5	8.4	2.4	24.4

Abbreviation	Meaning	Unit
QN	Data quality	---
TNN	Minimum temperature at 2m above ground	Degree Celsius (°C)
TNM	Mean daily minimum temperature at 2m above ground	Degree Celsius (°C)
TMM	Mean temperature at 2m above ground	Degree Celsius (°C)
TXM	Mean daily maximum temperature at 2m above ground	Degree Celsius (°C)
TXX	Maximum temperature at 2m above ground	Degree Celsius (°C)
SOS	Sum of sunshine duration	Hours
NMM	Mean cloud coverage	Oktas
RSS	Total amount of precipitation	mm
RSX	Maximum daily rainfall amount	mm
FMM	Mean wind power	Beaufort Scale (Bft)
FXX	Maximum wind speed at 10m height (peak height)	m/sec

**For Data quality (QN): 3** Data are for individual (especially automatically measured) elements systematically examined grossly and corrected if necessary.

### ANNEX 3. Denomination and year of admission or application for admission of hybrids to the German national list, their use and statistics for 3 traits in the 6 trials of grain maize performance test in 2009

Entry	Denomination	Admission year	Application year	Use	GFM (kg/plot)								GDCN1 (%)						Grain Yield (kg/plot)						
					EIK	WIO	NAK	NAO	KAK	KAO	Genotype mean	EIK	WIO	NAK	NAO	KAK	KAO	Genotype mean	EIK	WIO	NAK	NAO	KAK	KAO	Genotype mean
1	Amadeo	2004		grain and silage maize	15.5	16.1	15.1	12.0	16.7	10.8	14.4	67.0	63.7	77.3	74.8	67.9	66.5	69.5	10.4	10.3	11.6	9.0	11.3	7.2	10.0
2	RONALDINIO	2006		silage maize	15.9	17.2	16.0	12.5	17.3	10.1	14.8	65.7	64.6	74.2	72.1	66.5	62.9	67.7	10.4	11.1	11.9	9.0	11.5	6.3	10.0
3	SEVERO	2006		grain maize	16.1	13.7	15.3	12.7	17.0	9.3	14.0	65.8	61.4	74.7	71.9	65.1	62.3	66.9	10.6	8.4	11.4	9.1	11.0	5.8	9.4
4	KONFIANS	2005		grain maize	15.1	14.4	16.0	13.0	17.2	9.5	14.2	65.6	62.8	73.6	71.6	65.3	64.6	67.3	9.9	9.0	11.8	9.3	11.3	5.9	9.5
5	MARCELLO	2006		grain and silage maize	16.6	15.0	15.6	13.7	18.5	9.6	14.8	65.3	61.5	74.4	71.8	64.1	63.0	66.7	10.8	9.2	11.6	9.8	11.8	6.0	9.9
6	ATLETICO	2006		silage maize	15.3	15.7	15.7	12.7	17.6	9.7	14.4	65.2	63.5	73.9	70.5	64.9	62.4	66.8	10.0	9.9	11.6	8.9	11.5	6.0	9.7
7	LG 3220	2007		grain and silage maize	15.8	15.5	14.1	10.9	16.7	11.6	14.1	66.1	63.3	76.9	74.9	67.2	65.4	69.0	10.4	9.8	10.8	8.2	11.2	7.6	9.7
8	ES BOMBASTIC	2007		grain and silage maize	15.7	14.7	15.0	11.7	16.2	9.9	13.7	66.4	63.5	76.5	73.4	66.5	63.7	68.3	10.4	9.3	11.5	8.6	10.8	5.7	9.4
9	NK FALKONE	2007		grain and silage maize	15.4	15.3	15.3	11.3	16.3	9.4	13.8	67.2	64.6	76.2	72.4	67.8	64.4	68.8	10.4	9.9	11.7	8.2	11.0	6.1	9.5
10	P 8000	2009		grain and silage maize	15.2	15.6	14.4	12.3	16.2	11.4	14.2	67.1	63.7	77.7	74.3	67.0	65.3	69.2	10.2	10.0	11.2	9.1	10.9	7.4	9.8
11	SUSANN	2009		grain and silage maize	15.7	16.0	18.9	14.5	19.1	12.4	16.1	62.8	60.4	71.9	68.6	64.0	61.9	64.9	9.9	9.6	13.6	9.9	12.2	7.7	10.5
12	DKC 2960	2006		grain and silage maize	15.5	16.3	16.1	14.2	15.7	10.7	14.7	66.0	63.2	75.7	73.4	66.3	63.6	68.0	10.2	10.3	12.2	10.4	10.4	6.8	10.1
13	PR39F58	2003		grain and silage maize	16.2	16.9	15.5	13.0	16.9	9.9	14.8	65.3	63.1	75.0	73.2	65.6	62.4	67.4	10.6	10.7	11.6	9.6	11.1	6.2	9.9
14	AMBALL	2006		grain maize	14.7	16.2	15.4	13.3	16.6	6.1	13.7	66.7	63.4	76.4	73.0	66.7	64.3	68.4	9.8	10.3	11.8	9.7	11.1	3.9	9.4
15	ZIDANE	2007		grain maize	15.1	17.5	16.6	12.8	16.9	10.5	14.9	65.8	63.5	76.3	72.5	66.9	64.1	68.2	10.0	11.1	12.7	9.3	11.3	6.6	10.2
16	AMANATIDIS	2008		grain maize	15.1	17.3	13.8	12.7	16.1	7.5	13.7	67.6	65.5	76.9	74.6	69.0	64.8	69.7	10.2	11.3	10.6	9.5	11.1	4.9	9.6
17	AMARYL	2008		grain and silage maize	14.2	16.6	15.7	13.3	17.6	11.4	14.8	68.3	65.2	76.3	73.6	68.3	65.2	69.5	9.7	10.8	12.0	9.8	12.0	7.4	10.3
18	AMBROSINI	2009		silage maize	15.3	17.2	15.5	13.3	17.1	11.4	15.0	67.2	64.3	75.3	74.2	67.9	66.9	69.3	10.3	11.0	11.7	8.9	11.6	7.6	10.4
19	AGRO YOKO	2009		silage maize	15.4	16.7	14.9	12.9	15.5	13.8	14.9	66.3	63.3	75.8	73.7	66.6	66.3	68.7	10.2	10.6	11.3	9.5	10.3	9.1	10.2
20	PADRINO	2007		grain and silage maize	14.4	15.7	11.7	12.6	16.2	5.7	12.7	68.0	65.3	76.7	74.4	70.0	68.4	70.5	9.8	10.3	9.0	9.4	11.4	3.9	9.0
21	KWS 5133 ECO	2007		grain maize	16.0	15.6	14.9	14.3	17.4	6.9	14.2	66.5	64.3	73.9	71.1	66.4	65.1	67.9	10.7	10.0	11.0	10.2	11.6	4.5	9.7
22	TORRES	2007		grain and silage maize	16.2	14.3	16.9	14.3	17.8	9.5	14.8	66.9	64.1	75.8	72.6	66.5	62.1	68.0	10.9	9.2	12.8	10.4	11.9	5.9	10.2
23	RICARDINIO (A)	2008		grain and silage maize	16.0	13.6	14.4	9.4	17.0	8.4	13.1	66.4	61.5	76.2	72.6	67.1	63.7	67.9	10.6	8.4	11.0	6.8	11.4	5.3	8.9
24	MARCELINIO	2008		grain and silage maize	15.4	15.6	14.2	10.5	17.1	4.6	12.9	67.1	64.6	76.0	73.8	67.1	65.0	68.9	10.4	10.1	10.8	7.7	11.5	3.0	8.9
25	FABREGAS	2009		silage maize	15.3	15.6	14.8	11.1	17.2	8.8	13.8	68.8	64.7	77.4	75.9	70.2	66.4	70.6	10.5	10.1	11.5	8.4	12.1	5.9	9.7
26	CASSILAS	2009		silage maize	16.7	16.9	16.2	12.5	18.3	11.3	15.3	64.8	62.3	74.4	70.8	64.5	62.9	66.6	10.8	10.5	12.1	8.8	11.8	7.1	10.2
27	SYMBOL	2009		grain maize	16.4	15.6	16.2	14.4	17.1	10.5	15.0	64.6	61.6	73.2	69.4	64.5	62.7	66.0	10.6	9.6	11.8	10.0	11.0	6.6	9.9
28	FERNANDEZ	2009		silage maize	15.0	16.3	16.5	13.4	17.4	8.6	14.5	67.5	63.8	74.7	71.9	65.0	62.3	67.5	10.1	10.4	12.3	9.6	11.3	5.4	9.9
29	KXA 7340	2007			16.7	17.0	15.8	14.7	18.5	9.9	15.4	65.4	62.5	74.2	71.5	65.2	62.3	66.9	10.9	10.6	11.7	10.5	12.1	6.2	10.3
30	KXA 7341	2007			16.6	16.9	16.4	15.4	18.9	13.1	16.2	65.0	62.3	74.5	71.0	65.0	62.5	66.7	10.8	10.5	12.2	11.0	12.2	8.2	10.8
31	KXA 8113	2008			15.7	16.4	15.8	12.2	17.2	8.0	14.2	68.5	65.4	77.8	75.1	70.2	67.8	70.8	10.7	10.7	12.3	9.2	12.0	5.5	10.1
32	KXA 8114	2008			14.6	17.0	15.0	12.0	15.5	10.3	14.1	68.3	66.1	77.8	75.7	70.3	66.6	70.8	9.9	11.3	11.7	9.1	10.9	6.9	10.0
33	KXA 8121	2008			15.1	16.4	14.1	10.9	16.8	9.4	13.8	67.5	64.6	77.3	76.0	69.1	66.9	70.2	10.2	10.6	10.9	8.3	11.6	6.3	9.6
34	KXA 8122	2008			15.9	15.4	14.1	12.3	16.4	6.8	13.5	67.1	63.7	76.7	74.9	67.0	63.9	68.9	10.7	9.8	10.8	9.2	11.0	4.4	9.3
35	KXA 8141	2008			15.8	16.0	13.2	12.0	17.9	10.2	14.2	65.9	61.8	75.3	73.1	65.9	63.5	67.6	10.4	9.9	9.9	8.8	11.8	6.5	9.5
36	KXA 8144	2008			15.8	17.1	15.1	11.5	17.4	10.7	14.6	65.9	62.8	78.0	76.0	66.0	63.0	68.6	10.4	10.8	11.7	8.8	11.5	6.7	10.0
37	KXA 8151 (A)	2008			15.6	14.3	16.0	13.0	17.7	9.6	14.4	66.2	61.8	75.2	70.9	66.5	63.6	67.4	10.3	8.9	12.0	9.2	11.8	6.1	9.7
38	KXA 8162	2008			16.3	17.6	15.5	14.8	18.7	8.8	15.3	66.0	63.0	75.0	72.4	65.7	62.0	67.4	10.7	11.1	11.6	10.7	12.3	5.5	10.3
39	KXA 8163	2008			16.2	15.8	17.3	13.1	18.3	9.1	15.0	65.4	62.2	74.8	71.1	65.2	61.2	66.6	10.6	9.8	12.9	9.3	11.9	5.6	10.0
40	KXA 8164 (A)	2008			17.0	18.3	14.4	14.7	17.9	11.8	15.7	65.3	62.9	75.8	72.3	64.4	63.5	67.4	11.1	11.5	10.9	10.6	11.5	7.5	10.5
41	KXA 8181	2008			16.8	15.4	15.4	13.3	17.1	11.6	14.9	65.1	62.5	74.4	70.9	64.8	63.2	66.8	10.9	9.6	11.5	9.4	11.1	7.3	10.0
42	KXA 9113	2009			14.2	17.5	15.7	12.0	13.1	9.4	13.7	66.4	64.5	78.0	76.0	68.1	66.6	69.9	9.4	11.3	12.2	9.1	8.9	6.2	9.5
43	KXA 9114	2009			15.9	17.1	16.4	12.9	17.0	10.6	15.0	66.7	62.6	77.4	75.6	67.4	64.0	68.9	10.6	10.7	12.7	9.7	11.4	6.8	10.3
44	KXA 9118	2009			15.8	18.0	17.3	13.9	15.3	8.1	14.7	66.3	64.3	75.3	74.5	66.9	66.7	69.0	10.5	11.6	13.0	10.4	10.2	5.4	10.2
45	KXA 9122	2009			15.9	16.3	16.1	11.7	16.0	9.5	14.3	67.0	63.4	76.3	75.1	67.6	65.7	69.2	10.7	10.4	12.3	8.8	10.8	6.2	9.9
46	KXA 9123	2009			15.5	17.0	16.3	12.2	15.3	10.2	14.4	66.9	65.1	77.9	77.1	68.2	68.5	70.6	10.4	11.1	12.7	9.4	10.4	6.9	10.2
47	KXA 9124	2009			15.0	15.9	11.8	12.0	15.8	10.2	13.4	67.3	64.8	76.6	74.7	68.3	66.4	69.7	10.1	10.3	9.0	9.0	10.8	6.8	9.3
48	KXA 9141	2009			15.8	17.1	16.2	11.8	18.2	10.5	14.9	65.4	63.8	76.3	72.1	64.8	61.8	67.4	10.4	10.9	12.4	8.5	11.8	6.5	10.1
49	KXA 9142	2009			16.5	17.3	17.4	12.3	17.7	9.2	15.1	65.4	63.5	73.8	70.4	65.3	62.3	66.8	10.8	11.0	12.8	8.7	11.6	5.7	10.1
50	KXA 8164 (B)	2008			16.7	18.0	16.5	11.2	18.2	7.3	14.7	65.2	62.4	75.8	70.7	65.2	63.1	67.1	10.9	11.2	12.5	8.0	11.9	4.6	9.8
51	KXA 9144	2009			15.6	17.2	15.3	12.6	16.7	8.5	14.3	66.0	63.9	75.5	72.0	66.9	63.2	67.9	10.3	11.0	11.6	9.1	11.2	5.3	9.8
52	KXA 9145	2009			16.3	17.0	16.3	13.2	17.5	11.0	15.2	65.1	62.8	74.5	71.3	64.3	64.0	67.0	10.6	10.7	12.2	9.4	11.2	7.0	10.2
53	KXA 9146	2009			16.8	17.3	13.9	11.7	17.1	10.0	14.5	65.4	62.7	74.8	70.9	63.6									

## ANNEX 4. Denomination and year of admission or application for admission of hybrids to the German national list, their use and statistics for 3 traits in the 4 trials of silage maize performance test in 2010

Entry	Denomination	Admission year	Application year	Use	TFM (Kg/plot)					TDCn2 (%)					Silage Yield (Kg/plot)				
					EIS	WSO	KAS	KSO	Genotype mean	EIS	WSO	KAS	KSO	Genotype mean	EIS	WSO	KAS	KSO	Genotype mean
1	AMADEO	2004		grain and silage maize	47.9	47.7	39.5	29.2	41.1	33.7	32.8	35.4	31.2	33.3	16.1	15.7	14.0	8.9	13.6
2	NK FALKONE	2007		grain and silage maize	43.0	46.5	44.7	29.8	41.0	36.5	34.3	33.9	30.4	33.8	15.7	15.9	15.1	9.0	13.9
3	PADRINO	2007		grain and silage maize	49.2	50.7	48.7	36.7	46.3	32.8	32.1	30.8	30.4	31.5	16.1	16.3	15.0	11.0	14.6
4	RICARDINIO	2008		grain and silage maize	54.7	48.4	48.3	38.7	47.5	34.3	32.7	31.8	29.4	32.1	18.8	15.8	15.4	11.3	15.3
5	FABREGAS	2009		silage maize	47.5	51.2	43.1	36.3	44.5	37.7	36.9	35.4	29.8	35.0	17.9	18.9	15.2	10.8	15.7
6	RONALDINIO	2006		silage maize	54.1	60.4	50.4	43.0	52.0	32.7	32.5	31.8	29.3	31.6	17.7	19.6	16.0	12.6	16.5
7	TORRES	2007		grain and silage maize	55.0	51.2	45.9	39.9	48.0	33.9	32.1	31.9	29.8	31.9	18.6	16.5	14.6	11.8	15.4
8	GROSSO (A)	2010		grain and silage maize	59.8	63.8	61.3	39.3	56.0	31.5	29.4	27.8	27.3	29.0	18.8	18.7	17.0	10.7	16.3
9	GROSSO rez	2010		grain and silage maize	59.0	62.1	63.3	49.8	58.5	32.4	29.3	26.9	26.5	28.8	19.1	18.2	17.0	13.1	16.9
10	AMANATIDIS	2008		grain maize	51.5	50.4	51.5	39.2	48.1	34.5	32.6	30.1	28.8	31.5	17.8	16.4	15.5	11.2	15.2
11	ZIDANE	2007		grain maize	52.6	46.7	50.0	34.9	46.0	33.4	32.1	31.3	29.5	31.6	17.6	15.0	15.6	10.2	14.6
12	AMBROSINI	2009		silage maize	53.1	51.4	49.3	37.1	47.7	35.0	34.0	32.3	30.0	32.8	18.6	17.5	15.9	11.1	15.8
13	AMARYL	2008		grain and silage maize	53.3	54.5	52.5	36.3	49.2	32.5	31.6	30.0	28.9	30.7	17.3	17.2	15.8	10.5	15.2
14	AMAGRANO	2010		grain maize	48.0	43.8	40.7	35.7	42.1	36.6	34.3	36.6	30.7	34.6	17.5	15.0	14.9	10.9	14.6
15	AMARETTO	2010		grain and silage maize	59.3	67.4	62.7	44.6	58.5	32.0	29.5	27.9	26.7	29.0	19.0	19.9	17.5	11.9	17.1
16	Luigi CS	2010		grain maize	51.8	63.1	51.2	38.7	51.2	30.7	29.2	29.0	27.7	29.2	15.9	18.4	14.8	10.6	14.9
17	P 8000	2009		grain and silage maize	49.1	55.6	54.3	40.7	50.0	34.8	31.4	29.7	26.8	30.7	17.1	17.5	16.1	10.9	15.4
18	PR39F58	2003		grain and silage maize	53.1	58.1	58.4	46.5	54.0	31.5	30.9	28.8	26.2	29.4	16.7	18.0	16.8	12.2	15.9
19	Maritimo LG	2007		grain and silage maize	51.0	56.1	59.2	47.6	53.5	34.4	31.2	26.9	27.8	30.1	17.5	17.5	15.9	13.2	16.0
20	LG 3258	2009		grain and silage maize	55.7	54.9	54.7	47.6	53.2	33.4	32.0	29.5	27.9	30.7	18.6	17.5	16.2	13.2	16.4
21	LG 30218	2010		silage maize	52.4	54.6	53.2	48.5	52.2	33.6	33.9	30.3	27.6	31.3	17.6	18.5	16.1	13.4	16.4
22	Perform	2007		silage maize	57.6	61.2	53.5	52.1	56.1	30.2	28.6	29.4	26.0	28.5	17.4	17.5	15.7	13.5	16.0
23	NK Top	2009		grain and silage maize	49.7	49.8	45.5	40.2	46.3	33.8	33.2	30.9	28.3	31.6	16.8	16.5	14.0	11.4	14.7
24	APHRODITE	2009		silage maize	45.7	46.2	37.7	35.5	41.3	34.4	33.6	35.9	30.2	33.5	15.7	15.5	13.5	10.7	13.9
25	FERNANDEZ	2009		silage maize	59.1	69.0	61.1	44.2	58.3	31.0	29.6	27.8	26.9	28.8	18.3	20.4	17.0	11.9	16.9
26	KWS 5133 ECO	2007		grain maize	55.8	55.7	56.4	42.0	52.5	31.4	30.6	29.7	28.2	30.0	17.5	17.0	16.7	11.8	15.8
27	GROSSO (B)	2010		grain and silage maize	61.1	57.5	63.3	42.6	56.1	31.4	29.8	27.5	26.4	28.8	19.2	17.1	17.4	11.3	16.2
28	SILVINIO	2010		grain maize	47.6	50.0	42.0	35.2	43.7	36.4	33.6	34.0	29.9	33.5	17.3	16.8	14.2	10.4	14.7
29	BARROS	2010		silage maize	65.6	58.2	71.7	50.6	61.5	29.2	28.5	26.6	26.3	27.7	19.2	16.7	19.1	13.3	17.1
30	KXA 9148		2009		51.6	53.1	50.1	48.4	50.8	33.1	32.3	31.1	27.5	31.0	17.1	17.2	15.6	13.2	15.8
31	KXA 9144		2009		58.9	63.5	66.7	48.5	59.4	31.1	29.9	27.7	27.4	29.0	18.3	19.0	18.4	13.3	17.3
32	KXA 9171		2009		67.8	67.0	67.9	48.7	62.8	29.0	28.4	25.9	25.9	27.3	19.7	19.1	17.6	12.6	17.2
33	KXA 9306		2009		48.4	53.1	50.6	44.4	49.1	36.4	32.8	31.9	29.0	32.5	17.6	17.4	16.1	12.8	16.0
34	KXA 9329		2009		60.0	56.7	60.7	46.7	56.0	30.9	29.9	27.5	27.0	28.8	18.5	17.0	16.7	12.6	16.2
35	KXA 9354		2009		54.4	61.4	59.4	35.5	52.7	32.8	29.5	27.3	26.9	29.1	17.9	18.1	16.2	9.5	15.4
36	KXA 9356		2009		56.4	65.3	65.8	46.6	58.5	30.9	28.4	25.5	24.9	27.4	17.4	18.6	16.8	11.6	16.1
37	KXA 9361		2009		64.9	68.9	71.1	46.4	62.8	28.3	27.3	25.2	25.9	26.7	18.4	18.8	17.9	11.9	16.8
38	KXA 0121		2010		48.2	48.4	45.3	38.3	45.1	34.8	32.1	34.3	29.9	32.8	16.8	15.5	15.5	11.3	14.8
39	KXA 0122		2010		50.9	50.4	46.2	41.2	47.2	36.5	34.9	33.1	29.6	33.5	18.6	17.6	15.3	12.2	15.9
40	KXA 0123		2010		51.2	45.6	54.7	37.6	47.3	34.0	32.9	29.1	30.8	31.7	17.4	14.7	15.9	10.5	14.6
41	KXA 0133		2010		52.0	53.6	51.7	39.5	49.2	33.9	31.7	30.7	30.3	31.7	17.6	17.3	15.9	11.4	15.6
42	KXA 0136		2010		48.9	51.1	50.9	38.5	47.3	36.7	32.8	31.6	28.3	32.4	17.9	16.7	16.1	10.9	15.4
43	KXA 0141		2010		52.2	55.5	55.4	41.4	51.1	33.7	31.3	30.0	29.3	31.1	17.6	17.6	16.6	11.7	15.9
44	KXA 0144		2010		58.3	60.4	59.0	46.3	56.0	30.1	30.3	29.1	30.9	30.1	17.6	17.9	17.2	12.0	16.2
45	KXA 0145		2010		48.9	55.3	51.5	35.0	47.7	35.9	31.9	29.9	28.3	31.5	17.6	17.6	15.4	9.9	15.1
46	KXA 0151		2010		49.9	51.3	53.2	33.4	46.9	35.2	32.0	29.8	28.7	31.4	17.5	16.5	15.8	9.4	14.8
47	KXA 0152		2010		58.5	56.0	61.5	42.9	54.7	32.1	30.7	29.5	28.8	30.3	18.8	17.2	18.1	12.1	16.5
48	KXA 0154		2010		57.4	63.7	59.5	48.0	57.1	32.9	30.3	28.5	27.4	29.8	18.8	19.3	16.9	13.1	17.0
49	Moncada	2004		grain maize	46.7	55.2	52.8	41.2	49.0	35.3	31.7	28.6	27.6	30.8	16.5	17.4	15.1	11.3	15.1
<b>Environmental mean</b>					53.6	55.5	54.0	41.4		33.3	31.5	30.1	28.4		17.7	17.4	16.1	11.6	
Trait s.e.					2.4	4.0	2.4	5.2		1.1	0.8	0.8	0.8		0.8	1.2	0.7	1.4	
Trait C.V.(%)					4.5	7.2	4.5	12.6		3.2	2.5	2.6	3.0		4.4	6.9	4.2	12.1	
<b>CON mean</b>									<b>51.2</b>					<b>30.8</b>					<b>15.7</b>
Trait s.e.									3.6					0.9					1.0
Trait C.V.(%)									7.1					2.8					6.6
<b>ORG mean</b>									<b>53.8</b>					<b>31.7</b>					<b>16.9</b>
Trait s.e.									2.4					0.9					0.7
Trait C.V.(%)									4.5					2.9					4.3
<b>Grand mean (Overall)</b>									<b>48.4</b>					<b>29.9</b>					<b>14.4</b>
Trait s.e.									4.6					0.8					1.3
Trait C.V.(%)									9.6					2.8					9.0

## ANNEX 5. Comparison of performance ranks of 64 genotypes per farming system and per location for three traits in grain maize

Genotype		GFM Rank								Grain Yield Rank								GDCn1 Rank							
		Farming System		Einbeck		Niederaltleich		Kaufering		Farming System		Einbeck		Niederaltleich		Kaufering		Farming System		Einbeck		Niederaltleich		Kaufering	
Entry	Denomination	CON	ORG	EIK	WO	NAK	NAO	KAK	KAO	CON	ORG	EIK	WO	NAK	NAO	KAK	KAO	CON	ORG	EIK	WO	NAK	NAO	KAK	KAO
1	Amadeo	21	33	25	30	20	16	20	48	31	43	31	31	27	21	32	52	54	55	50	42	55	53	54	56
2	RONALDINIO	44	40	40	52	42	27	40	37	44	39	33	57	41	23	40.5	37	26	32	25	56	13	28	36	22
3	SEVERO	32	14	46	4	24	30	27	20	25.5	9	38	3	17	25	20	18	23	11	26	5	23	25	19	15
4	KONFIANS	33	17	12	7	40	38	39	26	21	13	9	7	36	34	29	24	17	34	24	24	8	20	24	45
5	MARCELLO	54	29	56	10	32	49	58	30	50	27	56	8	23	49	52	27	9	18	15	6	17	22	7	24
6	ATLETICO	37.5	23	17	22	35	32	46.5	33	22	18	12	22	22	19	37	28	12	12	12	39.5	11.5	8	16	17
7	LG 3220	12	19	33	15	6	5	22	56	11	20	35	15.5	7	6	28	59	45	51	34	32	52	54	48	51
8	ES BOMBASTIC	16	8	29	9	17.5	13	12	16	18	7	34	10	18	12	10	16	41	39	39	37	48	43	34	34
9	NK FALKONE	17	10	20	11	23	8	16	25	25.5	10	26	19	29	5	18	30	50	43	55	53	42	32	52	43
10	P 8000	8	44	14	20	12	23	13.5	53	10	46	18	23	14	27	15	56.5	53	48	51	43	59	48	45	50
11	SUSANN	64	60	31	27	64	58	63	62	62	55	8	13	64	52	60	61	1	1	1	1	1	1	5	4
12	DKC 2960	19.5	56	22	33	43	54	7	47	20	57	20	34	45	58	6	45	33	38	30	30	34	42	31	31
13	PR39F58	39	43	48	42	30	39	25	35	29	40	42	42	28	41	21.5	33	24	29	14	29	26	41	26	16
14	AMBALL	13	11	5	32	28	46	19	3	17	12	6	32	37	46	23	2	43	42	45	36	47	39	40	42
15	ZIDANE	40	49	10	58	56	34	26	43	45	45	11	56	58	32	31	41	38	37	27	39.5	43	34	41	40
16	AMANATIDIS	4	16	9	55	4	33	10	7	7	24	16	61	5	39	21.5	7	59	53	59	63	53	51	59	46
17	AMARYL	24	54	2	39	34	45	46.5	55	40	59	3	47	42	48	56	56.5	57	50	61	60	44.5	44	57	49
18	AMBROSINI	28	57	18	51	31	44	36	54	38	60	24	53	33	50	47	60	47	56	54	51	32	47	53	60
19	AGRO YOKO	6	61	19	40	15	36	6	64	6	62	17	39	16	40	5	64	37	49	36	33	35	45	39	53
20	PADRINO	1	4	3	23	1	28	13.5	2	2	8	7	30	2	35	34	3	61	63	60	61	50	49	61	63
21	KWS 5133 ECO	31	15	43	17.5	16	55.5	42	5	28	16.5	46	25	13	54	43	5	29	40	43	50	11.5	17	32	48
22	TORRES	55	24.5	49	6	59	55.5	50	27	61	29	57	9	59	56	53	22	40	31	48	48	38	36	37.5	8
23	RICARDINIO (A)	23	1	45	3	11	1	29	10	23	1	45	4	12	1	35	8	42	24	38	7	41	35	46	33
24	MARCELINIO	14	3	21	21	9	3	31	1	15	3	28	26	6	3	38	1	46	47	53	54	39	46	47	47
25	FABREGAS	19.5	6	16	19	13	6	38	14	48	14	36	27	20	10	58	21	62	58	64	57	58	60	63	54
26	CASSILAS	57	51	58	41	47	26	56	52	53	42	55	37	44	18	51	50	8	13	6	17	16	10	13	23
27	SYMBOL	49	48	54	17.5	45	57	33	42	32	37	41	12	39	53	19	40	6	3	5	8	5	2	12	21
28	FERNANDEZ	42	28	8	34.5	55	47	41	12	42	28	14	36	53	42	30	10	31	26	58	45	22	24	18	12
29	KXA 7340	56	55	60	45	37	59	59	34	54	51.5	60	41	31	59	59	31	18	16	20	19	15	19	20	13.5
30	KXA 7341	61	64	57	43	52	64	61	63	58	64	53	38	47	64	61	63	15	10	7	15	20	15	17	18
31	KXA 8113	37.5	12	28	37	38	20	37	8	56	26	51	45	51	29	57	13	64	61	63	62	60	56	62	62
32	KXA 8114	5	37.5	4	47	19	19	5	41	12.5	51.5	10	59	30	26	14	47	63	62	62	64	61	59	64	58
33	KXA 8121	9	13	11	36	7	4	24	21	19	21	19	40	10	8	45	36	60	60	57	55	56	61	60	61
34	KXA 8122	11	5	39	12	8	24	17	4	12.5	6	47	17	8	31	17	4	48	46	52	41	51	55	44	35
35	KXA 8141	15	27	32	28	3	18	52	39	8	22	30	21	3	16	48	38	30	28	12	31	50	40	28	29
36	KXA 8144	30	36	34	50	21	10	43	46	39	38	32	46	35	14	40.5	42	44	44	29	26	63	62	30	25
37	KXA 8151 (A)	47	22	26	5	41	37	48	32	49	16.5	23	6	43	30	50	29	32	17	35	11	29	13	37.5	32
38	KXA 8162	53	52	51	60	29	61	60	15	52	50	52	54	25.5	61	62	12	28	22	31	28	27	33	27	6
39	KXA 8163	60	26	47	24	61	40	57	17	60	19	43	18	61	33	55	14	20	7	19	14	24	16	22	1
40	KXA 8164 (A)	46	63	64	63	10	60	51	60	34.5	63	63	62	9	60	39	58	21	30	16	27	37	31	11	30
41	KXA 8181	48	46	61	13	27	43	35	57	34.5	41	61	14	19	37	24	54	14	19	9	21	18.5	14	15	28
42	KXA 9113	3	32	1	59	36	17	1	23	3	44	1	60	48	28	1	34	56	59	40	52	64	63	55	57
43	KXA 9114	45	50	38	48	53	35	28	45	55	49	40	44	57	47	36	43	51	45	44	22	57	58	49	36
44	KXA 9118	34	45	36	62	60	51	3	9	41	53	37	63	62	57	4	11	36	54	37	49	30	50	43	59
45	KXA 9122	29	20	42	34.5	44	11	9	28	43	25	49	35	52	15	12	35	49	52	49	34	44.5	57	51	52
46	KXA 9123	18	37.5	24	46	49	21	4	38	36	54	29	55	56	36	7	48	58	64	47	59	62	64	56	64
47	KXA 9124	2	24.5	7	26	2	15	8	40	1	35	13	33	1	20	11	44	55	57	56	58	49	52	58	55
48	KXA 9141	52	39	35	49	46	14	54	44	51	34	27	49	54	11	49	39	27	23	21.5	46	46	27	14	3
49	KXA 9142	59	34	55	56	62	25	49	19	57	30	54	51	60	13	44	17	16	14	18	38	10	7	23	13.5
50	KXA 8164 (B)	58	18	59	61	54	7	55	6	59	11	58	58	55	4	54	6	25	15	10	18	36	9	21	26
51	KXA 9144	25	30	27	53	25	29	21	11	27	32	22	52	24	24	26	9	35	33	32	47	33	26	42	27
52	KXA 9145	50	53	50	44	51	42	44	49	47	48	39	43	46	38	27	49	11	27	8	25	21	18	9	38
53	KXA 9146	27	31	62	57	5	12	32	36	9	23	62	48	4	7	13	32	10	9	21.5	23	25	12	3	5
54	KXA 9147	36	58	37	64	33	22	30	58	24	56	25	64	25.5	17	16	53	13	20	13	20	18.5	29	10	9
55	KXA 9148	43	47	53	29	26	41	34	51	46	47	59	29	32	43	33	51	39	41	41	44	40	38	35	37
56	KXA 9152	62	62	63	54	57	62	62	61	63	61	64	50	49	62	64	62	22	36	33	31	6	23	29	44
57	KXA 9161	26	35	23	16	39	53	15	31	16	36	15	20	38	55	9	26	19	25	17	35	14	30	25	10
58	KXA 9171	63	59	52	25	63	63	64	59	64	58	50	15.5	63	63	63	55	7	5	23	13	9	5	8	7
59	KXA 9172	10	42	13	31	22	52	11	24	5	33	4	24	11	51	3	19	2	4	3	4	3	4	2	11
60	KXA 9173	7	41	6	8	50	50	2	50	4	31	2	5	34	44	2	46	3	2	4	2	2	3	6	20
61	KXA 9181	51	21	30	14	48	31	53	29	37	15	21	11	40	22	42	23	5	6	11	9	4	11	4	2
62	KXA 9191	41	9	15	38	58	2	23	18	14	4	5	28	50	2	8	20	4	8	2	10	7	6	1	19
63	RICARDINIO (B)	22	2	41	1	17.5	9	18	13	30	2	44	1	21	9	25	15	52	35	46	16	54	37		

**ANNEX 6. Comparison of performance ranks of 49 genotypes per farming system and per location for three traits in silage maize**

Genotype		TFM Rank						Silage Yield Rank						TDCn2 Rank					
		Farming System		Einbeck		Kaufering		Farming System		Einbeck		Kaufering		Farming System		Einbeck		Kaufering	
Entry	Denomination	CON	ORG	EIS	WSO	KAS	KSO	CON	ORG	EIS	WSO	KAS	KSO	CON	ORG	EIS	WSO	KAS	KSO
1	AMADEO	2	2	6	6	2	1	2	2	4	6	2	1	42	44	28	39	46	49
2	NK FALKONE	3	1	1	4	6	2	4	4	2	8	10	2	46	47	46	46	43	45
3	PADRINO	10	12	13	13	12	12	6	16	5	9	8	17	25	40	20	29.5	32	44
4	RICARDINIO	20.5	10	31	7	11	18	31.5	9	42	7	13	22	38	39	33	36	38	34
5	FABREGAS	6	11	4	15	5	10	14	30	31	43	11	13	48	49	49	49	47	37
6	RONALDINIO	25	36	29	37	16	31	25	46	28	47	26	37	30	34	19	34	37	32
7	TORRES	15	19	32	16	9	22	18	18	39	11	5	28	36	36	30	31	39	38
8	GROSSO (A)	41	33	44	44	40	20	41	28	43	41	41	11	10	11	14	8	12	14
9	GROSSO rez	44	43	41	40	43	47	42	42	46	37	40	41	11	6	17	7	6	8
10	AMANATIDIS	20.5	14	19	11	20.5	19	19	14	29	10	15	19	31	30	36	35	29	28
11	ZIDANE	18	6	25	5	14	4	16	5	24	2	18	6	33	33	25	32	35	35
12	AMBROSINI	17	13	27	18	13	13	35	21	38	29	25	18	39	45	39	45	41	41
13	AMARYL	27	18	28	22	23	11	13	15	12	22	20	8	21	25	18	23	28	30
14	AMAGRANO	4	4	7	1	3	9	10	6	18	3	7	16	49	48	47	47	49	46
15	AMARETTO	42	45	43	47	42	34	44	44	45	48	44	31	14	7	15	10	13	9
16	Luigi CS	19	31	21	41	19	17	3	26	3	38	6	10	13	12	6	6	17	20
17	P 8000	23	22.5	12	28	28	24	17	20	11	28	29	15	29	17	37	22	24	10
18	PR39F58	33	38	26	34	33	37	23	33	7	35	37	36	16	14	13	19	16	5
19	Maritimo LG	30	37	17	31	35	41	22	38	19	27	24	45	18	19	35	20	5	21
20	LG 3258	31	32	33	24	30	40	37	39	40	30	31	43.5	22	23	24	27	22	22
21	LG 30218	26	34	24	23	26	44	27	45	25	39	30	48	27	31	26	44	30	19
22	Perform	32	47	37	38	27	49	15	40	15	26	19	49	12	4	5	5	20	4
23	NK Top	8	16	14	9	8	23	5	17	9	13	3	24	34	32	29	41	33	26
24	APHRODITE	1	7	2	3	1	7	1	7	1	4	1	12	45	43	34	42	48	42
25	FERNANDEZ	39	46	42	49	39	32	39	47	33	49	39	30	7	10	9	11	11	11
26	KWS 5133 ECO	34	27	34	29	32	28	34	25	17	18	35	29	17	18	11	17	23	23
27	GROSSO (B)	45	29	46	33	44	29	45	19	47	19	43	20	8	8	12	12	8	7
28	SILVINIO	5	8	5	10	4	6	8	10	13	16	4	7	47	41	44	43	44	39
29	BARROS	49	40	48	35	49	48	49	37	48	14	49	46	3	5	3	4	4	6
30	KXA 9148	16	30	20	20	15	43	11	36	10	21	17	43.5	28	22	23	33	34	17
31	KXA 9144	46	44	40	42	46	45	46	48	34	44	48	47	6	15	10	13	10	16
32	KXA 9171	47	48	49	46	47	46	48	43	49	45	45	38	2	3	2	3	3	3
33	KXA 9306	11	26	9	19	17	33	26	35	26	24	28	40	40	35	43	38	40	31
34	KXA 9329	40	35	45	32	38	39	38	29	36	17	34	39	5	13	8	14	9	13
35	KXA 9354	35	25	30	39	36	8	30	13	30	36	32	4	15	9	21	9	7	12
36	KXA 9356	43	42	35	45	45	38	33	34	14	40	36	26	4	2	7	2	2	1
37	KXA 9361	48	49	47	48	48	36	43	41	35	42	46	32	1	1	1	1	1	2
38	KXA 0121	7	9	8	8	7	15	9	8	8	5	16	21	43	37	38	29.5	45	40
39	KXA 0122	9	20	16	12	10	25	28	31	37	32	12	35	44	46	45	48	42	36
40	KXA 0123	28	3	18	2	29	14	20	1	16	1	23	9	23	42	32	40	18	47
41	KXA 0133	24	21	22	21	22	21	24	22	27	23	22	25	32	38	31	25	31	43
42	KXA 0136	13	15	10	14	18	16	29	12	32	15	27	14	41	28	48	37	36	25
43	KXA 0141	29	24	23	27	31	27	31.5	27	21	31	33	27	24	26	27	21	27	33
44	KXA 0144	37	39	38	36	34	35	36	32	22	34	42	33	9	29	4	15	19	48
45	KXA 0145	14	17	11	26	20.5	5	12	11	23	33	14	5	37	24	42	26	26	24
46	KXA 0151	22	5	15	17	25	3	21	3	20	12	21	3	35	27	40	28	25	27
47	KXA 0152	38	28	39	30	41	30	47	24	41	20	47	34	20	21	16	18	21	29
48	KXA 0154	36	41	36	43	37	42	40	49	44	46	38	42	19	16	22	16	14	15
49	Moncada	12	22.5	3	25	24	26	7	23	6	25	9	23	26	20	41	24	15	18

**ANNEX 7. Heritabilities and coefficients of phenotypic and genotypic correlation between farming systems for some traits in different maize studies from 1989 to 2011**

Trait	Grain Yield (a)		Grain Yield (b)		Grain Yield (c)		Grain Yield (d)		Grain Yield (e)	
Parameter	High N	Low N	CON	ORG	CON	ORG	CON	ORG	CON	ORG
$h^2$	65.3	65.0			41.0	39.6	70.5	63.0	65.0	52.0
$r_P$	0.53		0.36		0.40		0.41		0.39	
$r_G$	0.74		0.54		0.65		0.60		0.84	
Trait	GDCn1 (a)		GDCn1 (b)		GDCn1 (c)		GDCn1 (d)		Grain Moisture (e)	
Parameter	High N	Low N	CON	ORG	CON	ORG	CON	ORG	CON	ORG
$h^2$					82.1	80.3	89.8	81.0	88.0	83.0
$r_P$	0.88		0.89		0.88		0.88		0.87	
$r_G$	0.96		0.95		0.98		1.02		0.92	
Trait	Root Lodging (e)		Stalk Lodging (e)		Stay Green (e)		Plant Height (e)		Ear Height (e)	
Parameter	CON	ORG	CON	ORG	CON	ORG	CON	ORG	CON	ORG
$h^2$	6.0	63.0		27.0	20.0	58.0	76.0	80.0	43.0	61.0
$r_P$	0.44		-0.04		0.4		0.69		0.52	
$r_G$	0.55				0.57		0.93		0.99	
Trait	Grain yield at Low N (f)		Grain yield at High N (f)		N Response (f)		Average Grain Yield (f)			
Parameter	USA	ARG	USA	ARG	USA	ARG	USA	ARG		
$h^2$	51.0	45.0	59.0	51.0	43.0	6.0	63.0	27.0		
$r_P$										
$r_G$	0.57		0.52		0.83		0.54			

$h^2$  = heritability,  $r_P$  = Coefficient of Phenotypic correlation,  $r_G$  = Coefficient of Genotypic correlation

(a) mean of 21 field experiments results from the European dent and flint gene pool run under High-N and Low-N conditions without N fertilizer applications during 1989 and 1999 at several locations in Germany and France (Presterl *et al.*, 2003).

(b) mean of 4 material groups of preselected lines derived from early European breeding material. Evaluated testcross performance under organic and conventional conditions in 3 regions in Germany in 2008 (Messmer *et al.*, 2010).

(c) mean of 9 material groups of comparative field experiments under organic and conventional farming conditions. Evaluated testcross performance in 3 regions of Germany and selection of superior lines was practiced across two stages in 2004 and 2005. In 2006, single-cross hybrids from the 2 pool were tested in fields at organic and conventional systems in 2 regions in Germany (Burger, 2008; Burger *et al.*, 2008).

(d) mean of 6 material groups of comparative testcross performance under organic and conventional farming conditions. In 2009, 4 material groups were tested in 4 regions of Germany. In 2010 2 sites were employed also in Germany (Brandes, 2011).

(e) from test cross mean basis estimate in 119 intermated B73 x Mo17 RILs x LH295 test crosses evaluated across 2 Minnesota locations in 2006 (Lorenzana & Bernardo, 2008).

(f) mean of 50  $S_2$  (from maize populations with 100, 75, 50, and 25% of Argentine flint germoplasm), crossed to a flint and a dent inbred tester. Testcrosses were evaluated under low-N and high-N level in 2 locations in Illinois (high-N) during 1985 and 3 locations in Argentina (low-N) during 1986 (Brun & Dudley, 1989).