Evaluation and differentiation of garlic cultivars growing in Quebec using phenotypic and genotypic traits

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ABSTRACT

Garlic (*Allium sativum L.*) production in Québec represents only 10% of the local market. Local production is restrained in part by a lack of knowledge on the phenotypic and genotypic stability of different cultivars grown locally. The goals of this project were to study the phenotypic variation in a set of garlic cultivars grown in different environments in Québec, differentiate them using DNA polymorphic patterns, and compare the morphological classification to the genetic one. A two-year field trial was conducted in Lac Saint-Jean and Montréal areas with 36 garlic cultivars of different types. In the first part of this study, cultivars were characterized morphologically by measuring the size, shape and arrangement of leaves, pseudostems, scapes, bulbils, bulbs and cloves. Although differences between cultivar types were identified, it was not possible to obtain a unique set of morphological characteristics specific to each cultivar. Also, yield and yield related characteristics varied widely between environments and within environments. In the second part of this study, 35 microsatellite regions were screened and 21 markers have finally been used to differentiate our cultivars. Overall, garlic types were different genetically, but the cultivars within a type were generally identical resulting in redundancy for 61% of our set of cultivars. Since garlic is propagated by clones, it is likely duplicates, but some cultivars like 'Rose de Lautrec' showed major morphological differences with other cultivars found to be genetically identical. We also found that the cultivars, previously reported to show more than one phenotype had also more than one genotype. A more thorough investigation using advanced genotyping technique is needed to get better insight in the genetic identity of cultivars studied herein.

RÉSUMÉ

La production d'ail (*Allium sativum* L.) au Québec représente seulement 10% du marché local. La production locale est restreinte en partie par un manque de connaissance sur la stabilité phénotypique et génotypique des variétés d'ail cultivées. Cette étude avait donc pour but d'étudier la variation phénotypique de cultivars d'ail ayant poussés dans différents environnements, de les différencier en utilisant des marqueurs génétiques polymorphiques et de comparer les classifications morphologique et génétique ainsi obtenues. Pour ce faire, 36 variétés de types différents ont été plantées pendant deux ans dans les régions de Montréal et du Lac Saint-Jean. Dans la première partie de cette étude, les variétés ont été caractérisées morphologiquement en mesurant la taille, la forme et l'arrangement des feuilles, pseudotiges, bulbilles, bulbes et caïeux. Même si des différences entre les types de variétés ont été identifiées, il n'était pas possible d'obtenir des caractéristiques morphologiques uniques pour chaque variété. Le rendement et les traits associés au rendement variaient grandement entre les différents environnements et à l'intérieur de ceux-ci. Dans la deuxième partie de cette étude, 35 régions microsatellites ont été analysées et 21 ont été utilisées pour différencier les variétés. En général, les types d'ail étaient différents génétiquement, mais en général les cultivars d'un même type n'ont pas été différenciés entre eux résultant en 61% de redondance. Comme l'ail est propagé par clones, il pourrait s'agir de doublons, mais certaines variétés comme Rose de Lautrec ont présenté des différences morphologiques majeures avec les autres cultivars trouvés génétiquement identique. Nous avons aussi trouvé que les cultivars qui présentaient plus d'un phénotype avaient aussi plus qu'un génotype. Une investigation plus poussée utilisant

des techniques génétiques plus avancées serait nécessaire pour avoir plus de connaissances sur l'identité génétique des cultivars étudiés.

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LIST OF ABBREVIATIONS

AFLP: Amplified Fragment Length Polymorphisms

AGE: Agarose Gel Electrophoresis

bp: Nucleic acid base pairs

cDNA: Complementary deoxyribonucleic acid

DNA: Deoxyribonucleic acid

EST: Expressed Sequence Tags

FAO: Food and Agriculture Organization of the United Nations

GSSR: Genomic Simple Sequence Repeats

LSD: Least Significant difference

PAGE: Polyacrylamide Gel Electrophoresis

PCA: Principal Component Analysis

PCR: Polymerase Chain Reaction

PIC: Polymorphic Information Content

RAPD: Randomly Amplified Polymorphic DNA

RFLP: Restriction Fragment Length Polymorphisms

RNA: Ribonucleic acid

SNP: Single Nucleotide Polymorphism

SRAP: Sequence-related Amplified Polymorphism

SSLP: Simple Length Polymorphisms

SSR: Simple Sequence Repeats

STR: Short Tandem Repeats

Ta: Annealing Temperature

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PREFACE

The idea of this project was conceived by Dr. Jean-Benoit Charron, Dr. Philippe Seguin, Mr. Hervé Van Der Heyden (MSc), Marie-Pascale Beaudoin (Agr, MAPAQ) and the candidate. This work is composed of original data taken by the candidate or summer students working with the candidate. All photographs of garlic were taken by the candidate. The candidate wrote chapter I, II, IV and V. The chapter III was co-written by the candidate and Dr. Philippe Seguin. All chapters were edited by Dr. Jean-Benoit Charron and Dr. Philippe Seguin.

1. General Introduction

1.1 Introduction

Garlic is cultivated worldwide to produce fresh, crushed or powdered garlic bulbs but also leaves and flowers. Its importance as a vegetable, seasoning and health keeper is better known than ever (Meredith, 2008). As a result, the worldwide production has nearly doubled in the last decade to 30 million tons in 2019 (FAO, 2019) and Québec production has increased 600% since 2017. Although, the Québec production is still modest with 299 tons of garlic produced on 144 hectares of land in 2019 it represents the quarter of the Canadian production (Statistics Canada, 2019). Chinese garlic has a predominant place on the local market despite the fact that Québec garlic is considerate as a more tasteful product that stores better and longer (Lepage, 2018). As a matter of fact, Quebec garlic represents only 10% of the local market (Statistics Canada, 2021). The diversification of garlic cultivars grown in Québec could allow producers to expend their stock more rapidly, offer garlic for a longer period on the market and meet consumer preferences in terms of look, size and flavor. The diversification of garlic cultivars offered on the market, in addition to the increasing desire for fresh and local products, can favour the establishment of local production in Québec.

The promotion of Québec garlic production remains difficult because the identity and characteristics of many cultivars are unclear. The classification system mostly used in Québec is based on the morphological description of cultivars by Engeland (1991). Hardneck (*i.e.*, with a hard flower stem) are subdivided into Rocambole, Porcelain, Creole, Turban, Purple Stripe, Marbled Purple Stripe and Glazed Purple Stripe. Softneck cultivars (*i.e.*, without a flower stem) are divided into Artichoke, Silverskin and Asiatic. However, garlic has the ability to adapt to its environment leading to a wide range of morphological characteristics for a unique cultivar (Meredith, 2008; Volk and Stern, 2009). This has led to a duplication of names for a genetically singular cultivar and a single name for cultivars with different characteristics. In some countries like France, a classification and certification system is in place to favour the uniformization of garlic cultivars sold and to give access to pure lines of planting stock exempt of disease (Messiaen *et al.*, 1993). With such a system, it would be easier to develop a competitive market for growers in Québec.

The first step to create such a system is to develop tools to identify and describe garlic cultivars. Morphologically, traits like number, size and color of cloves and bulbils could be a better indicator than bulb coloration, size and taste that have varied between environments (Volk and Stern, 2009; Polyzos *et al.*, 2019). To cope with morphological variation and possible duplicate, genetic markers can be used to differentiate garlic cultivars. Recently, garlic cultivars from Brazil (Cunha *et al.*, 2014), Argentina (Barboza *et al.*, 2020), India (Kumar *et al.*, 2019), Korea (Ma *et al.*, 2009; Lee *et al.*, 2011), the United States (Ipek *et al.*, 2003) and other countries (Zhao *et al.*, 2011; Jo *et al.*, 2012) have been differentiated using microsatellite markers (or Simple Sequence Repeats -SSR). With the recent release of the garlic genome sequence (Sun *et al.*, 2020), genetic information will be more accessible to better understand garlic's behaviour and cultivars differences. However, microsatellite markers will continue to be efficient tools to differentiate garlic cultivars within a collection as they are highly polymorphic, have a low cost per sample and are reproducible (Powell *et al.*, 1996). Therefore, this project proposes to evaluate the performance of garlic cultivars growing in two regions of Quebec, identify their main morphological characteristic, differentiate them using genetic markers and compare the morphological classification obtained to the genetical one.

1.2 General hypothesis

Each garlic cultivar will be described using a unique set of morphological characteristics and those phenotypically different cultivars will show unique DNA polymorphic patterns. Cultivars that fall under particular phenotypic groups as described by the North American Classification System proposed by Engeland (1991), will be more genetically alike.

1.3 General objectives

The main purpose of this research is to investigate the relationship between garlic morphological traits and their genetic identity for cultivars growing in Québec.

In order to achieve this, the following objectives will be performed:

1. Study the morphological variation among garlic cultivars and growth locations.
2. Develop a molecular assay using SSR markers to differentiate garlic cultivars growing in Québec.
3. Compare the classification obtained by morphological characteristic to the one obtained from genetic analysis.

2. Literature review

2.1 Garlic as a crop

Garlic (*Allium sativum* L.) is a vegetative crop from the family of *Alliaceae* (former *Liliaceae*). The *Allium* genus includes more than 700 species of which the best known are onions (*Allium cepa)*, leeks (*Allium ampeloprasum* L.) and chives *(Allium schoenoprasum* L.) (Jones and Mann, 1963; Keller, 2002). Garlic is the second most important *Allium* crop after onion and is now cultivated worldwide (Jones and Mann, 1963; Brewster, 2008). This crop has been a part of human life for thousands of years. As a matter of fact, evidence of garlic use has been found 5,000 years ago in Egyptian civilization, but the origin of garlic is estimated to be far from this point, in Central Asia, where a wild fertile relative, *Allium longicuspis* Regel, grows. Through the process of domestication, garlic lost its fertility likely due to the selection for larger garlic bulbs (Etoh and Simon, 2002). However, in the last years, male fertility has been regenerated in some cultivars (Etoh, 1986; Hong and Etoh, 1996; Etoh and Simon, 2002; Shemesh-Mayer *et al.*, 2015). The production of true seed is still long and difficult, and garlic propagation continues to be achieved asexually with bulbs or with bulbils. Even though garlic is propagated only by clones, it has adapted to its environment resulting in hundreds of cultivars growing in different conditions. For example, the Canadian Seed Catalogue Index for garlic contains 353 cultivars (Seeds of Diversity, 2021). The exchanges of clones for seedlings between farmers of different locations and the tendency to rename new planting stock may have created duplicates of some cultivars with different names. Ipek *et al.* (2003), Volk *et al.* (2004), Cunha *et al.* (2014) and Barboza *et al.* (2020) have found 41 to 72% of their collection that shared their genotype with at least one other cultivar.

2.1.1 Economical importance

Garlic was grown in one hundred countries for a total area cultivated of 1.6 million hectares of land in 2019 (FAO, 2020). The worldwide production increased from 20 million tons metric of garlic in 2007 to over 30 million tons of garlic in 2019. The largest producer is China with 77% of the worldwide production in 2019 (FAO, 2020) while Canada produced 1248 tons of garlic on nearly 536 hectares for a farm gate value of $14,245,000. Québec is responsible for close to the quarter of Canadian production with 299 tons on 144 hectares for a farm gate value of $3,917,000. This production is 6 times more important than in 2007 but only represent 10% of the local market (Statistics Canada, 2021).

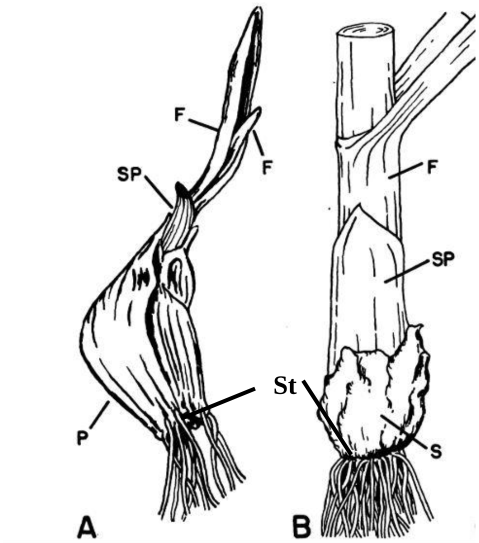
The increase in production in the last decade mainly comes from advances in knowledge about the health properties of garlic. As a matter of fact, garlic bulbs are an important source of antioxidants (Bozin *et al.*, 2008; Chen, Shen, *et al.*, 2013) and have anticancer (Jonkers *et al.*, 1999), antibacterial (Hughes and Lawson, 1991), immune enhancement and cardiovascular (Meredith, 2008) properties. Garlic is now available on the market in various forms: fresh, crushed, dry, in powder or in oil. Bulbs are still the most consumed part of garlic as a condiment but flower and leaves are also eaten (Brewster, 2008).

2.1.2 Garlic growth

Compared to the majority of crops, garlic growth does not come from a seed but from a vegetative structure, normally a clove. Therefore, the main purpose of the plant is not necessary to produce flowers and seeds but to have enough energy to produce a bulb composed of one or more cloves (Jones and Mann, 1953). Moreover, the aboveground stem of the plant looks like a normal one (*i.e*. full stem with buds that supports leaves) but it is a cylindrical pseudostem composed of the sheath of each leaf passing in the middle of the others. The roots and leaves are produced from a stem plate present at the clove base (Mann, 1952; Meredith, 2008). The sprout leaf is the first leaf to grow from the stem plate of the clove. This bladeless leaf leads the way into the clove towards the soil for the subsequent foliage leaves (Fig.2.1). Each new leaf will produce a blade at 180° of the past leaf, and in that way contribute to plant growth. Garlic produces a small, set number of leaves limiting the plant length and reducing its photosynthetic competitiveness (Mann, 1952; Meredith, 2008).

When the plant is mature, environmental conditions will trigger the redistribution of the plant’s energy to the bulb and, in some cultivars, to an inflorescence. At this period, garlic cultivars grown in North America will need a long day photoperiod coupled with high temperatures to achieve multiclove bulbs. Prior to this, garlic needs a cold treatment (*i.e*., vernalization) to gain the ability to develop an inflorescence and a bulb at plant maturity. Without vernalization, garlic will only produce leaves and sometimes a single clove bulb called a round bulb. This treatment is naturally given when garlic is planted in fall. The fall planting will allow garlic to begin its growth. Roots will be developed, and the sprout leaf will begin to growth inside the clove before the frost. This late fall growth will allow garlic to emerge early in the spring. (Mann, 1952; Meredith, 2008).

Unlike most storage structures, a clove is a differentiated leaf produced from lateral buds' primordia on the youngest foliage leaf. Those lateral buds will form two bladeless leaves that will differentiate into a protection leaf and a storage leaf when environmental conditions are met. The storage leaf will swell to form the new clove flesh surrounded by a protection leaf which separates each clove from another and is known as the clove skin. The resulting bulb is a distinct structure composed of multiple cloves enclosed by the remaining foliage leaves. In some cultivars, bud initiation is triggered by up to six new leaves resulting in a multi-layer bulb each separated by their own wrapper (Jones and Mann, 1963; Meredith, 2008).



F : Foliage leaf

P : Protection leaf

S : Depleted storage leaf (clove)

SP : Sprout leaf

St : Stem plate

Fig 2.1 Schematic visualization of a sprouting garlic clove (A) and a young plant (B).
Adapted from Mann (1952).

At the same time, a scape will be produced passing through the pseudostem. The scape can be straight, or coiled in its first stage, then the coiled stalk will uncoil. The umbel capsule at the extremity of the scape will then open to release flowers and bulbils. The shape and colour of the umbel capsule, the flower, and the bulbils will change depending on the cultivar (Jones and Mann, 1963; Meredith, 2008). The nutrient competition between bulbs, bulbils and flowers, the growing condition, and some genetic mutations will lead flowers to become senescent before the production of seed, resulting in garlic sterility. In some cultivars, the artificial removal of bulbils in their early development allows the flower to produce seeds. The first generation seed has a low germination rate (10-35%), but the second and third generation produce more seeds with higher germination rates (65-100%) (Pooler and Simon, 1994; Hong and Etoh, 1996; Simon and Jenderek, 2003). In incomplete bolting cultivars, the scape and inflorescence will form depending on the conditions, but bulbils can also be produced from the pseudostem without a scape (Meredith, 2008).

2.1.3 Cultural practices

Cultivation of garlic is a long process as it takes nearly nine months to produce a marketable bulb. In North America, as previously described, garlic is planted in the fall. The planting time is one of the first parameters that needs to be controlled to obtain a good production. If cloves are planted too soon, leaves will develop outside the clove and will be affected by the freezing temperature of winter resulting in a growth delay or plant death. If planted late, garlic roots may not establish well and yield will be reduced. Garlic can also be planted in the early spring, but the yield is reduced compared with fall planting (Waterer and Schmitz, 1994; Põldma *et al.*, 2005). However, some cultivars must be planted in the spring to grow as they are not adapted to Quebec winters (Waterer and Schmitz, 1994). Spring planted garlic must be artificially vernalized to grow. Pre-chilling requirements can be obtained by passing between four and eight weeks at 4°C. Less than four weeks will not allow the production of a multiclove bulb. After more than eight weeks, clove quality is more likely to deteriorate and early differentiation of buds, before plant maturity, is promoted resulting in unsaleable bulbs: rough bulbs, small bulbs or round bulbs (Jones and Mann, 1963; Bandara *et al.*, 2000; Michael *et al.*, 2018). Longer vernalization can give good results if the temperature is above 4°C but below 10°C (Mann and Minges, 1958). The planting stock is also a variable to control as bigger cloves tend to produce bigger bulbs. Moreover, diseased planting stock will lead to small plants, contamination of soil and in some cases, death of the plant (Kamenetsky, 2007; Volk and Stern, 2009).

As a matter of fact, diseases and pests of planting stock are the main problems affecting garlic crops. Nematodes (*Ditylenchus dipsaci*), viruses (Onion yellow dwarf virus), *Penicillium* and fusarium contaminate tissues and survive in cloves, thus affecting the following season's planting stock (The American Phytopathological Society, 2008). The main methods to limit these problems are to choose disease-free planting stock, rapidly eliminate diseased plants and having a crop rotation of four years with plants from families other than *Alliaceae*. Meristem tissue culture and bulbil plantations are used to produce disease-free plants but they are time-consuming methods (Meredith, 2008).

The plant will also be affected by a range of abiotic factors. Originally, garlic has grown in rocky soils, but such soils do not keep enough water to produce more than a single clove bulb. For the same reason, sandy soil will produce weak plants and small yield. On the other hand, heavy clay soils are too compact, which may restrict root growth and impact bulb development. As garlic roots are not efficient in nutrient uptake from soil, fertilization is needed for a good yield. Too much nutrient like nitrogen can stimulate foliage development and decreased bulb size. If garlic is fertilized for too long, it can reduce bulb size and storage life. Weed presence also has a huge impact on garlic plants as they have a limited production of leaves. Weed pressure at the early stages can reduce growth and, at plant maturity, can inhibit bulb growth. Consequently, weeds need to be removed for plants to grow efficiently and produce sellable bulbs (Meredith, 2008).

One common practice in garlic culture is the scape removal for bolting cultivars. The scape drains energy from the plant that could otherwise be used for bulb growth. Keeping the scape can reduce bulb yield by 5 to 30% (Rosen and Tong, 2001; Pelter *et al.*, 2005; Ulianych *et al.*, 2019). However, a plant with a scape will produce a bulb with a longer storage life. But farmers who sell scapes must remove them at an early stage when they are still coiled, or else they will harden. The timing in scape removal can be optimized to have the minimum yield loss and the maximum conservation time. This optimal time is variable between cultivar and range from the moment when the scape is entirely coiled to the middle of the uncoiling process (Rosen and Tong, 2001; Pelter *et al.*, 2005; Meredith, 2008).

Garlic has to be harvested before its outdoor foliage leaf has been degraded by soil microorganisms, but after those leaves have dried. If harvested too late, the cloves will separate from another in the soil or in storage. If it is harvested too soon, it will store poorly and never achieve a good taste. The only way to be sure if garlic is ready to be harvested is by looking directly at the bulb in the soil. This technique is long and can result in lost bulbs. Therefore, maturity is usually approximated by the number of dry leaves on the plant. The recommendation varies when between one third and three quarters of the plant leaves that are becoming brown (Jones and Mann, 1963; Meredith, 2008).

When bulbs have been harvested and dried, they must be stored properly to allow them to be planted in a few months or to sell them during the winter. Each garlic cultivar does not have the same ability to be stored: some will rapidly lose their properties while others can stay a year without significant changes. The storage condition depends on the utilization of garlic and will affect the life quality of the garlic bulbs. On the one hand, low temperature storage (below 10°C) will break dormancy and leaves will sprout. It is expected to induce vernalization but not for long-term storage. If garlic is kept to be planted in the spring, it must be conserved between 0°C and -3°C to reduce disease spread and shrinking. This range of temperature conserves garlic cloves without degradation and promotes rapid normal growth when the cold treatment is broken. On the other hand, high temperature storage (above 20°C) will promote garlic dehydration and thus reducing storage life. As a result, garlic stored to be sold during winter should be kept near 15°C. To avoid dehydration and disease propagation, garlic bulbs should be kept undivided at 45-50% humidity with good air flow. Furthermore, the storage lifetime depends on harvest conditions: high temperature, high fertilizer, wet conditions and late harvest decrease storage life (Meredith, 2008).

2.2 Genotyping analysis

2.2.1 Microsatellites

Microsatellites also known as Simple Sequence Repeats (SSR), Short Tandem Repeats (STR) and Simple Length Polymorphisms (SSLP) are DNA sequence composed of a 1-6 nucleotide repeat motif (Kole and Abbott, 2008; Mason, 2015). In plants, they have been studied in a wide range of species including pine (*Pinus spp*), oak (*Quercus spp.*), rice (*Oryza spp*.), maize (*Zea mays*), and soybean (*Glycine max*) (Vieira *et al.*, 2016). The distribution of SSRs is not random among the genome. They are more present in the non-coding region or when found in the coding region are related to protein translation (Varshney *et al.*, 2005; Mason, 2015; Vieira *et al.*, 2016).

Microsatellites repeats can be categorized as perfect, imperfect or compound. Perfect SSRs are a repeat sequence of the exact same motif while imperfect SSRs have an insert in the sequence. Compound SSRs have multiple motifs with or without an insertion (Mason, 2015; Vieira *et al.*, 2016). The SSRs obtained by genomic sequencing are called genomic SSRs or GSSRs while SSRs obtained from Expressed Sequence Tags (ESTs), cDNAs and genes are called genic SSRs or EST-SSRs. Those SSRs are present in the coding region and are more conserved (less polymorphic) than genomic SSRs. They are efficient tools for studies about SSR transferability among relative species and functional studies. The existence of public EST databases make them easier to be developed and cheaper (Varshney *et al.*, 2005) but GSSRs are more useful in fingerprinting and varietal identification studies due to their higher mutation rate (Kalia *et al.*, 2011).

2.2.2 Genetic diversity analysis

Microsatellites have been used more often in the last years for genetic diversity analysis as genomic information has become more available and cheaper. Previously, other markers were used such as Restriction Fragment Length Polymorphisms (RFLPs), Amplified Fragment Length Polymorphisms (AFLPs), Randomly Amplified Polymorphic DNAs (RAPDs) and isozymes (Maaß and Klaas, 1995; Ipek *et al.*, 2003; Volk and Stern, 2009; Vieira *et al.*, 2016). AFLPs and RAPDs are dominant markers while RFLPs and SSRs have codominant inheritance. Isoenzymes and RAPDs are less polymorphic than AFLPs and microsatellites (Ipek *et al.*, 2003; Vieira *et al.*, 2016)., and RFLPs have lost their popularity due to the extended use of toxic compounds and the high labour needed to achieve good results (Mason, 2015) and RAPDs are not sufficiently reproducible to be useful in varietal identification. The markers AFLPs and SSRs are the most sufficient tools for polymorphic studies but AFLPs are harder to automate and produce multiple small fragments for a single region (Kalia *et al.*, 2011). Therefore, SSRs are now mostly used for genetic diversity studies because they can reach a high level of precision, are reproducible, use a small amount of starting DNA, cover a large portion of the genome and, once primers are developed, are not too expensive (Powell *et al.*, 1996; Vieira *et al.*, 2016).

The analysis of SSR loci is obtained by PCR amplification. Polymorphic SSR loci will show a difference in length from an addition or a deletion of one or many motifs in the sequence. Those modifications can come from polymerase slippage errors or recombination events (Vieira *et al.*, 2016). The longer the SSR motif is, the more likely it will be polymorphic (Vieira *et al.*, 2016). The fragment visualization can be made by three methods: agarose gel electrophoresis (AGE), polyacrylamide gel electrophoresis (PAGE) or capillary electrophoresis. The first, AGE has a lower resolution but is easy to do and cheaper. The other methods have the same higher resolution (1 bp) but PAGE is more time-consuming and uses toxic compounds than capillary electrophoresis(Sánchez-Pérez *et al.*, 2006; Mason, 2015; Vieira *et al.*, 2016). Those disadvantages have led to extensive development of capillary electrophoresis and thus the diminution of cost. The common method labels PCR products with a fluorophore by the addition of a 5' tailed tag on the forward primer sequence. The most common tag is the M13 sequence (Schuelke, 2000; Hayden *et al.*, 2008). The reaction can be multiplexed using two methods to reduce cost. First, many SSR loci can be amplified in the same PCR reaction by using different tags and fluorophores for each primer used. Second, once amplification has completed, PCR products can be mixed. Different fragments of non-overlapping lengths can be mixed and analyzed together or similar fragment lengths can be coupled with different fluorescent markers (Hayden *et al.*, 2008; Vieira *et al.*, 2016). The utilization of SSR markers analyzed with capillary electrophoresis is thus an effective way to rapidly and precisely access genetic diversity.

2.2.3 Genomic advance on garlic genome

In garlic, little is known about its genome mainly due to its large size. Garlic is a diploid species (2n=2x=16) with a large genome, highly repetitive and heterozygote genome. A complete assembly of the garlic genome (16.24 Gbp) and a large-scale transcriptome have just been released (Sun *et al.*, 2020). Before that, only partial information on the genome was available creating difficulty to characterize the diversity and investigate selection strategies. Sun *et al.* (2012) have sequenced the bud transcriptome resulting in 127,000 unigenes. Shemesh-Mayer *et al.* (2015) have studied the difference between male fertile garlic and male infertile garlic by analyzing transcriptomic information in flowers while Kamenetsky *et al.* (2015) have developed a catalogue of sequenced transcriptomes including green leaf, basal plate, root, inflorescence, flower and bulb clove transcriptomes. Finally, the complete chloroplast genome (153,172 bp) is also available (Filyushin *et al.*, 2016). As a result, 127 biological sequences are available in the Sequence Read Archive (SRA) of NCBI (https://www.ncbi.nlm.nih.gov/) and most of them are RNA sequences. In 2021, the nucleotide database of NCBI contains 102,753 entries for *Allium sativum* (plants) including 22,774 EST sequences and 729 genomic sequences.

Some of those sequences have been used to create SSR primers resulting in 1,714 SSR primers developed from garlic genomes that have been published (Ma *et al.*, 2009; Cunha *et al.*, 2012; Ipek *et al.*, 2015; Liu *et al.*, 2015; Barboza *et al.*, 2018). Among them, 409 have been tested on a minimum of two accessions and 317 produce a PCR amplicon. The majority of SSRs were developed from EST sequences (293/317) as they are more available. 27 SSR regions have been successfully used to study genetic diversity among garlic cultivars from a few countries (Ma *et al.*, 2009; Zhao *et al.*, 2011; Cunha *et al.*, 2012; Jo *et al.*, 2012; Cunha *et al.*, 2014; Chen *et al.*, 2014; Chand *et al.*, 2015; Ipek *et al.*, 2015; Liu *et al.*, 2015; Wang *et al.*, 2016a; Barboza *et al.*, 2018; Kumar *et al.*, 2019). These studies found that garlic expressed a high genomic diversity that can be used to cluster garlic cultivars. However, the relation between accessions that cluster together using SSRs are not consistent. Some authors show a relationship between genetic clustering and geographical origin (Ma *et al.*, 2009; Wang *et al.*, 2016a) while others discovered a more important relationship with flowering ability and ecophysiological traits such as vernalization requirement and dormancy abilities (Ipek *et al.*, 2015; Barboza *et al.*, 2018). Thus, even with the advance in garlic genomic knowledge it is still difficult to classify garlic cultivars.

2.3 Phenotypic diversity of garlic

For garlic to be grown around the world without sexual propagation, it had to present a high phenotypic plasticity. Spontaneous mutations happened through the thousands of years of domestication have been selected leading to the creation of hundreds of lines of garlic with specific traits (Etoh and Simon, 2002).

2.3.1 Morphological characteristics

The extreme variability in morphological traits among garlic cultivars, because of their interaction with the environment and growth conditions, have made the classification of garlic difficult. On the one hand, the number of bulbils and their size, the clove arrangement, number, weight, colour, and skin adherence are cultivar-specific characteristics. On the other hand, the yield, bulb size, colour, and flavour are among traits with more variation. Specifically, the yield depends on the initial planting stock, the incidence of pathogens and viruses, spacing and planting depth, soil, growth, and harvest conditions. The taste and bulb colour depend mostly on the soil characteristics (organic and nutrient content) and the amount of water (Kamenetsky, 2007; Volk and Stern, 2009).

One of the horticultural characteristics mainly used for classifying garlic cultivars is the ability or not to produce a scape. Bolting or hardneck cultivars of garlic have this ability. Cultivars that have completely lost their abilities to produce scapes and flowers are called non-bolting or softneck (Etoh and Simon, 2002). However, some cultivars have been found to produce scapes depending on storage conditions, planting date, photoperiod, and temperature conditions during growth. Others will produce bulbils through the pseudostem without producing a complete scape. They have been classified as incomplete-bolting or weakly bolting cultivars (Mann and Minges, 1958; Etoh and Simon, 2002; Meredith, 2008). Different studies have identified genetic differences between bolting and non-bolting types via isoenzymes, RAPDs, AFLPs and SSRs (Pooler and Simon, 1993; Maaß and Klaas, 1995; Al-Zahim *et al.*, 1997; Volk *et al.*, 2004; Barboza *et al.*, 2020).

2.3.2 Classification systems

Classification systems have evolved over the years as genetic information has been added to the morphological data. Also, garlic can express different traits depending on where in the world they are cultivated. As a result, some specific systems have emerged to classify garlic growth in specific countries. Subspecies *sativum*, *ophioscorodon*, *pekinense* and *longicuspis* are used to classify garlic clones but their utilization is controversial. These groups are not considered as botanical subspecies and their distinguishing characteristics sometimes overlap (Jones and Mann, 1963; Etoh and Simon, 2002; Meredith, 2008). The main characteristics used in this classification system are the origin, bolting capacity and inflorescence characteristics.

These groups present a lot of variation, thus Engeland (1991) analyzed U.S. garlic grown in Washington state and divided *sativum* and *ophioscorondon* groups into different types. These types are used in North America to characterize garlic cultivars. This classification described garlic with traits that vary within the environment as bulbs size and colour and more stable traits as the number of bulbils and cloves. The bolting, single layer cloves cultivars of the *ophioscorodon* subspecies are divided into Rocambole, Porcelain, Purple Stripe, Marbled Purple Stripe, Glazed Purple Stripe, Creole and Turban. The multilayer cloves, softneck or weakly bolting cultivars of the *sativum* subspecies are divided into Artichoke, Silverskin, and Asiatic (Engeland, 1991; Meredith, 2008). Studies using AFLP and RAPD markers have compared genetic identity to the classification described by Engeland and most of the varieties show distinct genetic traits. However, results obtained from Turban, Creole and Asiatic cultivars are not as robust as the others (Al-Zahim *et al.*, 1997; Volk *et al.*, 2004).

At the same time, in France, Messiaen *et al.* (1993) subdivided garlic into five groups with the first three being the main European groups. The groups are mainly divided by origin, dormancy, planting time, and clove arrangement. French garlic plants must be passed through a certification process prior to being sold as planting stock. The goal of this process is to give access to unique cultivar planting stocks of good quality with distinct traits and exempt from pests and diseases. To do so, garlic cloves are first planted for six generations in a field free of any *Allium* species for 5 years and at least 300 m away from any *Allium.* Garlic plants lacking trait-uniformity as well as diseased plants are removed at each generation (Messiaen *et al.*, 1993, Prosemail and GNIS, 2018). Garlic cultivars are then categorized by their morphological characteristics and separated into four clusters: Autumn-planted purple garlic (group III), Autumn-planted white garlic (group III), Hardneck Garlic for Autumn and Spring planting (group I) and Softneck Spring-planted pink garlic (group II). Each unique cultivar is described by morphological traits used by the Community Plant Variety Office (CPVO, 2001), then labelled for commercialization with its origin, bulb and clove skin colour, planting dates, sprouting time, and dormancy ability (Prosemail and GNIS, 2018).

Other countries use phenotypic descriptors to evaluate their stocks (CPVO, 2001; IPGRI *et al.*, 2001; USDA, 2020), but none are as advanced as France for identification certification.

Connecting text to Chapter 3

The previous chapter has reviewed the recent literature on garlic and garlic production in Quebec. It highlighted some of the challenges that the industry is currently facing. One of the main challenges for Québec garlic producers is the access to quality seeds, which is caused in part by a lack of knowledge on garlic cultivars, their traits and their adaptation to Québec climate. One of the key objectives of the following chapter is to provide a better understanding of the stability of phenotypic traits of garlic cultivars grown in contrasted environments of Québec and determine how they fit in the classification system described by Engeland (1991).

3. Phenotypic characterisation and evaluation of garlic (*Allium sativum* L.) cultivars growing in different environments in Québec, Canada

Abstract

Multiple garlic cultivars are currently grown in Canada. However, most farmers in Québec grow the cultivar Music. To increase the presence of local garlic on the local market, the diversification of the cultivars offered is needed but a lack of knowledge on the behaviour of garlic in the province limits this process. In this study, 36 garlic cultivars were grown in four contrasting environments in Québec to assess variation in performance and evaluate them phenotypically based on 28 characteristics related to the bulb, clove, bulbils and plant morphology. Yield and yield-related traits were variable between and within environments while bulbils color, shape and number were the most stable with clove color and number. Two cultivars showed two sets of well-defined traits even when grown in the same environments. Those different phenotypes have clustered with different types of garlic using Gower dissimilarity matrix. Due to the close phenotype of many cultivars and the high variation obtained between environments, genetic analyses are needed to ascertain the identity of the cultivars. This study could serve as a cornerstone to better understand the phenotypic variation of garlic cultivars grown in Québec.

3.1 Introduction

Garlic (*Allium sativum* L.) is a crop adapted to a wide range of environments which is grown in hundreds of countries. However, of the 28 million tons of garlic grown worldwide annually more than 77% is produced in China (FAO, 2021). In recent years, the demand for fresh and local produce has increased; garlic is one example of a crop that can easily be grown locally in North America including the Province of Quebec. Its production in Quebec has increased by 660% in the last decade, however, local production currently still represents only 10% of the garlic market (Statistics Canada, 2019). Reasons for this small market share of locally produced garlic include high production costs and a short period of availability (*i.e.*, from August to December at best). Even if Quebec garlic represents a small portion of the market, the demand remains high due to its perceived more intense flavor and better conservation compared to imported overseas garlic (Lepage, 2018). One possible approach to increase the production of garlic and its market share in Quebec could in part reside in the diversification of garlic cultivars produced. Diversification could lengthen the availability of locally produced garlic as harvest and conservation range would most likely increase thus making local garlic more appealing for local retailers. Also, more cultivars available on the market would distinguish Quebec garlic from the quite uniform imported garlic in terms of looks, size and flavor thus potentially meeting the preferences of more consumers.

Even if garlic is propagated asexually, it has accumulated multiple mutations over time resulting in a high phenotypic variability (Singh *et al.*, 2018). As a result, multiple attempts have been made to classify garlic into morphological groups (Etoh and Simon, 2002). In North America, the description of Engeland (1991) is most commonly used. This classification is based on morphological traits that are relatively stable across environments such as the number of bulbils and cloves, but also traits that tend to vary across environments such as bulbs size and colour. This classification divides cultivars into ten distinct groups. The bolting, single layer cloves cultivars are divided into Rocambole, Porcelain, Purple Stripe, Marbled Purple Stripe, Glazed Purple Stripe, Creole and Turban. The multilayer cloves, softneck or weakly bolting cultivars are divided into Artichoke, Silverskin, and Asiatic (Engeland, 1991; Meredith, 2008). Genetics differences using markers like Randomly Amplified Polymorphic DNA (RAPD) (Al-Zahim *et al.*, 1997) and Amplified Fragment Length Polymorphisms (AFLP) (Volk *et al.*, 2004) have confirmed the classification of cultivars into those groups except the ones from Asiatic, Creole and Turban. All groups of Engeland's classification can be grown in Canada, however, they do not perform equally. In addition, of the 200 garlic cultivars named in the seed of diversity catalogue in Canada (Seeds of Diversity, 2019), few have been studied. Despite this apparent diversity, in Quebec the cultivar 'Music' is used by the majority of local producers because of its good adaptation and high yields (Landry and Khanizadehl, 1994). The knowledge of the adaptation and growth habits of other cultivars that can be grown in Quebec remains limited.

One of the main challenges in using new cultivars is the unstable characteristics of garlic cause by its high phenotypic plasticity. As a result, a specific cultivar can look quite different when grown in different environments, even in the same province or region. This could be an issue for producers trying to diversify their stock of garlic since it could be harder to have sufficient uniformity to meet retailers or consumers quality criteria. The yield, bulb size, colour, morphology and flavour could change depending on growth condition like precipitation and temperature or soil characteristics (Kamenetsky, 2007; Volk and Stern, 2009; Atif *et al.*, 2020). For example, in 2018, large differences in precipitations between regions of Quebec, resulted in bulbs of the most commonly used cultivar locally (*i.e.*, Music) to be purple in the Montérégie area, but bulbs remained the typical white color in other regions (Marie-Pascale Beaudoin, Agr, Personal Communication).

The lack of information regarding the adaptation of garlic cultivars that can be grown in Quebec and garlic plasticity makes it challenging for local producer to diversify their stock of garlic and meet consumer preference for a consistent uniform product. Therefore, the aim of this chapter is to characterize garlic cultivars grown in different environments of Quebec. Specifically, the objective of this chapter is to i) identify among 36 cultivars the ones with the greatest production potential and the most adapted in Quebec, ii) examine the phenotypic variation of these garlic cultivars when grown locally and iii) identify traits that could be used to establish a Quebec garlic classification system.

3.2 Material and Methods

3.2.1 Plant materials

A total of 36 garlic cultivars were grown in 2019 and 2020 in two contrasting regions of Québec, Canada, one in the south in the Montréal area [Sainte-Anne-de-Bellevue (45.410056°, -73.938378° and 45.407886°, -73.935997°) later referred to as S19 and S20] and the other in the North in the Lac Saint-Jean area [L'Ascension-de-Notre-Seigneur (48.677066°, -71.619716°) in 2019 and Alma (48.639461°, -71.741653°) in 2020, later referred as N19 and N20, respectively] (Table 3.1). In the southern environments, the normal growing degree-day (5°C base; GDD5) between April and October range between 1868 and 2018 while in the northern environment it is between 1263 and 1413. The soil in fields used in both regions and years were predominantly composed of clay. The general characteristics of fields used at each site and years are presented in Table 3.1, while the prevailing temperatures and precipitation are presented in Figure 3.1.

Cultivars evaluated included some of each type of Engeland's classification: one Artichoke (Italian), two Asiatic (Tibetan Red and Tibétain Red), three Creole (Ajo Rojo, Creole, and Rose de Lautrec), eight Porcelain (Georgian Crystal, German White, Leningrad, Music, Northern Quebec, Susan Delafield, Ukrainian Mavnir, and Yugoslavian Porcelain), seven Marble Purple Stripe (Duganskij, Eureka Myrtis, Kostyn's Red Russian, Metechi, Northern Siberian, Siberian Red, and Wenger's Russian), two Purple Stripe (Chesnok Red and Persian Star), eight Rocambole (Baba Franchuk, French Rocambole, German Red, Italian Purple, Killarney Red, Marino, Mountain Top, and Penn Wonder), two Silverskin (Silver Rose and Silver White), two Turban (Maiskij and Portuguese) and one unclassified (Sweet Haven). Artichoke and Silverskin types are considered to be softneck cultivars, Creole, Asiatic and Turban can be either softneck or hardneck cultivars and all other cultivars evaluated are hardneck cultivars. Sweet Haven was not planted in S19, but all cultivars were planted in S20. In N19, cultivars Tibétain Red, Tibetan Red, Music, Susan Delafield, Kostyn's Red Russian, Northern Siberian, Killarney Red and German Red were not planted, while in N20, the cultivars Tibétain Red, Music, Susan Delafield, Northern Siberian, and German Red were not planted.

Each plot was planted in the fall and consisted of one row of 12 cloves in 2018 and 20 in 2019 planted 12 cm apart at a 5 to 7 cm depth. Interow spacing was of 30 cm. Each plot was separated in two, plants in the first half being grown using traditional management (*i.e.*, removing garlic flowers before they uncurl), while flowers were kept untouched until harvest in the second half of the plots. Plots were assigned to a randomized complete block design each treatment being replicated thrice.

Seeding date, irrigation schedule, soil cover used for overwintering, fertilization and curing were conducted using local practices prevailing in the region of concern, see Table 3.1 for details. Garlic was hand-harvested when a third to half of the leaves turned yellow. After curing, garlic outside bulb wrappers were cleaned, roots and shoot were cut. They were then stored in the dark at 20-24°C until analysis.

3.2.2 Phenotypic Data Collection

The phenotypic data was based on the criteria used in the European garlic classification system (CPVO, 2004), the Czech collection (IPGRI *et al.*, 2001) and the U.S. National Plant Germplasm System (USDA, 2019). At the beginning of the flowering stage which occurred in the last two weeks of June in the southern environments and in the first two weeks of July in the northern environments, leaf number, plant height, pseudostem diameter, pseudostem coloration and maximal number of turns of the flowering stem for each plant were recorded. At harvest which occurred at the end of July or beginning of August at the southern sites and later in August at the northern sites, spathe length was determined. After harvest, dried garlic and bulbils were sent to McGill University for analysis were dry bulb weight and size (*i.e.*, circumference, diameter, and height), clove number, bulbils weight and number were determined.

Visual assessment of bulbs, cloves and bulbils colors and shape was also performed. Garlic bulbs color patterns were assigned to the following groups: plain white, purple stripes or marbled purple. Their shapes were described using the stem plate position (inner, outside, or neutral) and the bulb longitudinal form (large, narrow, or round). Cloves were assessed for their color and assigned to the following groups: white, cream, pink or purple with or without stripes. Cloves' arrangement was described in terms of number of clove layers. Bulbils were assessed for their color and assigned to the following groups: with a continuous color or a marbled color of white, brown, purple or pink. Their shapes were described depending on their length:width ratio where 1 to 1.5 is a corn shape, between 1.5

and 2.5 is a drop shape and >2.5 is a rice shape. Finally, the presence/absence of a hard flower stem was assessed.

3.2.3 Statistics

Data from the four environments were analyzed separately as there were significant interactions between cultivars and sites for all variables (data not presented). All quantitative data were submitted to an analysis of variance with treatments considered fixed factors and replicates as random factors using R (R Core Team, 2020). When significant treatment effects were observed differences between cultivars were assessed using LSD ($P<0.05$). For bulbs related data, only the plants submitted to traditional management were analyzed, while the plants of the other half of the plots were used for data related to flowering. ~~.~~ Qualitative data were assessed using frequencies for each class of the phenotypic descriptors. Two cultivars, Metechi and Music, showed two different phenotypes even when grown at the same location. As a result, those two phenotypes were separated in the statistical analysis to better understand this pattern and are referred to as cultivars Metechi-2 and Music-2.

Using mean and mode for each cultivar in each field, a dissimilarity matrix was made using Gower's distance (Gower, 1971) with the *cluster* package in R (Maechler *et al.*, 2021). This distance allows using both qualitative and quantitative variables. Visualization of this distance was made using a dendrogram.

Table 3.1: Location and agricultural practices used to grow 36 garlic cultivars.

	City	Administrative region	Planting Year	Planting date	Harvest Date	Latitude (°)	Longitude (°)	Canada Hardiness Zone	Irrigation source	Cover	Nitrogen source	Drying technique
S19	Sainte-Anne-de-Bellevue	Montreal	2018	Early Nov.	End of July - Beginning Aug.	45.41006	-73.93838	5b	Rainfall	Leaves	Inorganic	Bulk of 8 complete plants - Hanging with air flow
S20	Sainte-Anne-de-Bellevue	Montreal	2019	Late Oct.	End of July - Beginning Aug.	45.40789	-73.93600	5b	Drip tape as needed starting mid-june	Leaves	Inorganic	Bulk of 8 complete plants - Hanging with air flow
N19	L'Ascension-de-Notre-Seigneur	Saguenay-Lac-St-Jean	2018	Early Nov.	August	48.67707	-71.61972	3a	Rainfall	None	Compost, Chicken Manure, bonemeal	Bulk of 8 complete plants - On heating ground
N20	Alma	Saguenay-Lac-St-Jean	2019	Early Nov.	Late August	48.63946	-71.74165	3a	Rainfall	None	None	Bags of 8 bulbs - Hanging with air flow

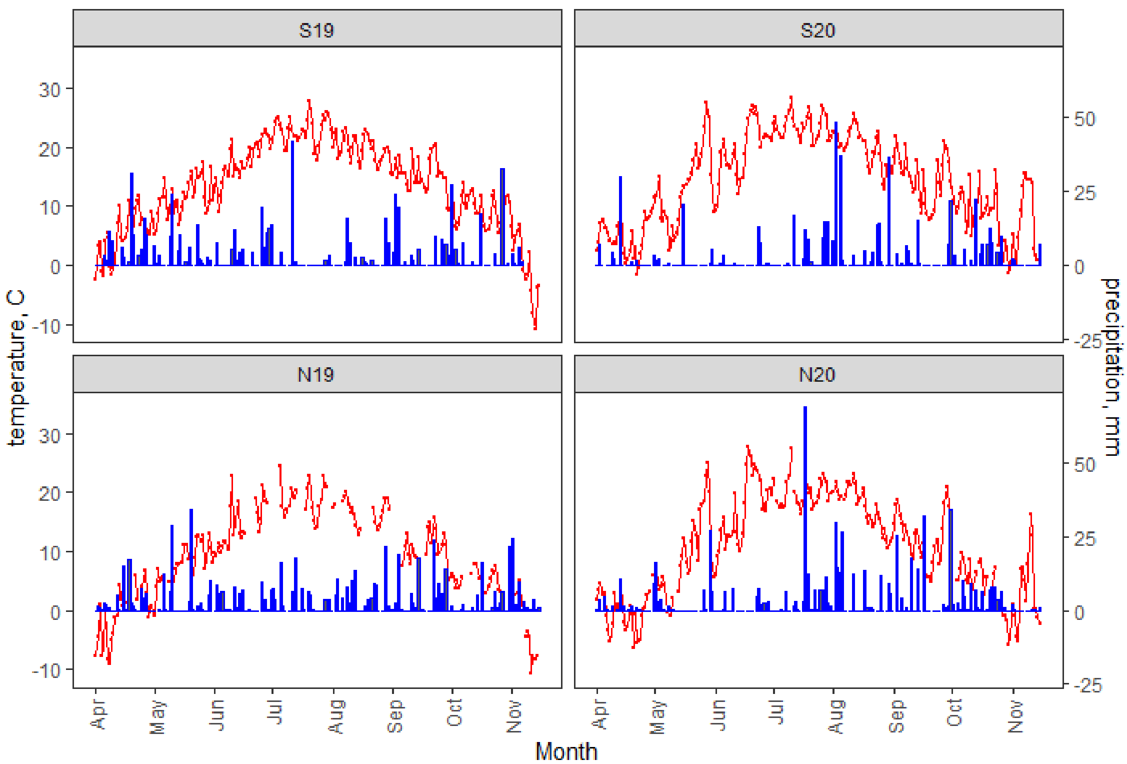


Fig. 3.1 Average daily temperature (red) and total precipitations (blue) during the growing season in 2019 and 2020 at Saint-Anne-de-Bellevue (S19, S20) and Lac St-Jean (N19, N20).

3.3 Result:

3.3.1 Yield and yield components

Differences in yield and yield components were observed between cultivars in each environment (Tables 3.2-3.5). The yields observed were highly variable since they were impacted strongly by the percentage of plant emergence. The only site with a consistently high emergence across most cultivars was S20, therefore this specific environment shows less intra-cultivar variation across replicates in terms of bulb yield per hectare. The overall yield was maximal in S20 and minimal in N20. Overall, across cultivars bulb yield in S20 was 20, 35, and 68% greater than in S19, N19 and N20, respectively. In the southern environments (*i.e*., S19 and S20), bulb yields in 2019 ranged between 1.4 and 12.1 ton/ha with an average of 6.3 ton/ha, while in 2020 yields ranged between 0.4 and 15.0 ton/ha with an average of 8.7 ton/ha. In the northern environments (*i.e*., N19 and N20), bulbs yield in 2019 ranged between 1.2 and 9.4 ton/ha with an average of 5.4 ton/ha, while they ranged between 1.1 and 6.2 ton/ha with an average of 2.3 ton/ha in 2020.

Specific cultivars had consistently high bulb yield across three out of four environments including Duganskij which is one of the highest yielding cultivars in S19, N19 and N20 (average of 8.6 ton/ha) and Eureka Myrtis and Wenger Russian in S19, S20, N20 (average of 9.0 and 9.8 ton/ha respectively) (Tables 3.2-3.5). Along with these cultivars, Leningrad had a relatively stable performance across all four environments producing above average yields (average of 8.4 ton/ha) and below average variation across replicates and environments (data not shown). The cultivar Music, which is the most commonly grown cultivar in Québec was among the highest yielding cultivars in the two environments in which it was grown in (9.3 ton/ha in S19 and 13.2 ton/ha in S20). The performance of the cultivar Kostyn Red Russian in S20 (12.2 ton/ha), the cultivars Mountain Top and Northern Siberian in S19 (average of 9.3 ton/ha) and the cultivar Ukrainian Mavnir in N19 (8.8 ton/ha) were also better than most in these specific environments.

Variation in average bulb weight in a given environment was less important than variation observed for bulb yield per hectare, however, it remained large and greatly impacted by the environment (Tables 3.2-3.5). As a result, none of the cultivar was among the ones with the greatest bulb weight in more than two out of four environments. Music and Wenger Russian were among the cultivars with the greatest average bulb weight in S19 and S20 (average of 47.1 g/bulb), like the cultivar Duganskij in S19 and N19 (average of 39.2g/bulb), and the cultivar Leningrad in S19 and N20 (average of 34.2g/bulb). Among these cultivars, Leningrad had a high stability across the four environments, along with the cultivars German White, Northern Siberian, and Susan Delafield which were only evaluated in the southern environments (*i.e.*, S19 and S20). Differences among cultivars and environments for bulb circumference were overall, following similar trends to the ones observed for average bulb weight (Tables 3.2-3.5).

Some cultivars had a poor performance in all environments in terms of bulb yield per hectare and yield components. Cultivars Creole and Ajo Rojo were the least productive producing the lowest yields (average of 1.5 ton/ha) and smallest bulbs (9.7 g/bulb) in all environments (Table 3.2-3.5). In some cases, the plants failed to emergence or were too small to produce a bulb at the time of harvest. Also, when bulbs were produced, they often presented abnormalities like a clove outside the bulb wrappers or one clove skin for many cloves. The cultivars Silver Rose and Siberian Red also did not grow in N20 even if 60 cloves were planted. The cultivars Marino, Silver Rose and German Red also tended to produce low yields and low average bulb weights at all sites (average of 4.4 ton/ha and 25.7 g/bulb at the southern sites and an average of 2.3 ton/ha and 14.8 g/bulb at the northern sites).

Table 3.2: Bulb yield and yield components of 36 garlic cultivars grown at Saint-Anne-de-Bellevue, QC, Canada in 2019 (S19). Means in a given column followed by different letters are significantly different ($LSD_{0.05}$).

	Yield (ton/ha)		Bulb Weight (g)		Bulb Circumference (mm)		Clove Number per Bulb		Clove Weight (g)		Bulbils Number per Spathe		Bulbils Weight (mg)	
Cultivar	Mean	sd	Mean	sd	Mean	sd	Mean	sd	Mean	sd	Mean	sd	Mean	sd
Artichoke														
Italian	8.1	± 1.5 a-d	35.6	± 10.4 b-d	14.9	± 1.6 ab	10.7	± 2.8 b	3.5	± 1.0 g-i	-	- -	-	- -
Asiatic														
Tibetan red	7.8	± 0.9 a-d	33.5	± 9.5 b-f	13.6	± 1.3 c-g	7.6	± 1.6 cd	4.4	± 1.2 g-i	20.9	± 7.3 i	243.8	± 126.6 de
Tibétain red	7.5	± 3.8 a-e	31.0	± 8.0 c-f	13.5	± 1.2 c-g	6.5	± 0.8 de	4.8	± 1.2 g-i	28.7	± 9.6 i	267.4	± 157.5 cd
Creole														
Creole Red	2.3	± 0.5 ef	15.4	± 15.2 g	9.8	± 3.1 h	8.9	± 3.3 c	2.6	± 4.2 i	131.0	± 0.1 e-g	4.0	± 0.1 h
Rose de Lautrec	6.0	± 2.5 a-f	28.8	± 11.0 d-g	12.7	± 2.2 fg	8.9	± 2.0 c	3.4	± 1.4 hi	108.7	± 54.5 fg	23.7	± 13.5 h
Marble Purple Stripe														
Duganskij	12.1	± 1.6 a	45.5	± 14.0 a	15.8	± 2.2 a	5.7	± 0.5 e-g	7.9	± 2.1 d-f	71.4	± 38.7 gh	104.4	± 64.2 fg
Eureka Myrtis	7.8	± 1.5 a-d	29.1	± 10 d-g	12.9	± 1.4 fg	3.0	± 1.4 i-k	11.1	± 4.9 c	133.4	± 30 ef	56.1	± 23.2 gh
Kostyn's Red Russian	5.7	± 4.2 a-f	26.5	± 12.1 fg	12.4	± 1.9 g	3.7	± 2.2 ij	9.9	± 6.6 cd	116.1	± 36.6 fg	56.7	± 14.1 gh
Metechi	5.6	± 2.8 a-f	29.2	± 10 d-g	13.3	± 2.1 d-g	4.5	± 1.0 g-i	6.8	± 3 e-g	79.5	± 6.6 gh	96.0	± 4.8 gh
Metechi(2)	5.6	± 4.2 a-f	36.8	± 12 a-d	14.3	± 1.5 a-e	9.0	± 1.3 c	4.1	± 1.4 g-i	203.8	± 79.0 bc	12.5	± 4.3 h
Northern Siberian	9.0	± 2.9 a-c	33.8	± 11.4 b-f	13.7	± 2.0 c-f	3.9	± 1.3 i	9.3	± 3.6 c-e	100.7	± 52.1 g	99.3	± 142.4 g
Siberian red	6.6	± 4.7 a-f	39.3	± 8.1 ab	14.7	± 1.3 a-c	4.4	± 0.7 g-i	9.2	± 2.4 c-e	78.8	± 24.3 gh	68.3	± 17.8 gh
Wenger's Russian	8.0	± 3.3 a-d	37.3	± 14.3 a-c	13.9	± 2.2 b-f	4.2	± 1.7 hi	9.7	± 4.1 cd	98.8	± 29.4 g	80.2	± 33.4 gh
Porcelain														
Georgian Crystal	7.4	± 1.4 a-f	34.4	± 6.4 b-f	13.6	± 1.1 c-g	3.0	± 0.8 i-k	12.5	± 5.2 a-c	235.8	± 76.7 b	27.0	± 4.1 h
German White	7.9	± 2.7 a-d	32.1	± 7.7 b-f	12.9	± 1.5 fg	2.3	± 0.6 k	14.1	± 3.5 ab	235.2	± 38.0 b	22.6	± 7.6 h
Leningrad	7.7	± 0.9 a-e	36.1	± 12.4 a-d	13.3	± 2.2 d-g	3.6	± 1.3 i-k	10.8	± 4.1 cd	296.4	± 56.5 a	23.4	± 3.5 h
Music	9.3	± 5.0 ab	43.5	± 17.0 a	15.5	± 2.1 a	3.1	± 0.9 i-k	14.1	± 4.7 ab	315.9	± 68.4 a	33.3	± 11.9 h
Music(2)	6.4	± 1.1 a-f	25.0	± 4.2 fg	12.7	± 0.1 fg	8.5	± 0.7 cd	2.9	± 0.3 hi	26.5	± 3.5 i	392.0	± 83.4 ab
Northen Quebec	5.1	± 4.3 b-f	37.3	± 9.6 a-d	13.6	± 1.5 c-g	2.6	± 0.9 jk	15.8	± 6.0 a	301.0	± 87.9 a	23.1	± 5.2 h
Susan Delafield	4.2	± 2.4 c-f	27.8	± 5.4 d-g	12.4	± 1.4 g	2.4	± 0.7 k	12.1	± 3.0 bc	170.3	± 79.4 cd	23.9	± 10.7 h
Ukrainian Mavniv	6.0	± 2.3 a-f	26.7	± 5.8 e-g	12.3	± 0.9 g	2.6	± 0.8 jk	10.8	± 3.4 c	211.2	± 57.3 b	19.2	± 4.2 h
Yugoslavian Porcelain	2.7	± 2.7 ef	34.3	± 5.8 b-f	13.1	± 1.4 d-g	2.2	± 0.4 k	15.6	± 2.5 a	182.5	± 83.3 b-d	18.8	± 5.9 h
Purple Stripe														
Chesnok Red	6.5	± 4.4 a-f	34.2	± 11.1 b-f	13.6	± 2.5 c-g	7.6	± 3.1 cd	5.8	± 3.6 fg	112.5	± 29.7 fg	32.3	± 18.8 h
Rocambole														
Baba Franchuk's	4.7	± 2.3 b-f	35.5	± 7.0 b-e	14.0	± 1.6 b-f	7.4	± 2.0 cd	5.0	± 1.3 f-i	24.8	± 7.4 i	284.9	± 133 b-d
French Rocambole	7.1	± 3.4 a-f	32.1	± 10.2 b-f	13.3	± 1.9 d-g	6.5	± 0.9 de	5.6	± 1.5 f-h	25.5	± 11.1 i	194.5	± 91.0 ef
German Red	1.4	± 0.2 f	29.6	± 3.4 d-g	13.6	± 1.3 c-g	9.0	± 0.1 bc	3.3	± 0.4 hi	29.3	± 12.7 hi	205.0	± 76.0 def
Italian Purple	5.6	± 3.3 a-f	39.2	± 15.2 a-c	14.6	± 2.2 a-d	8.1	± 1.1 cd	4.8	± 1.8 g-i	27.6	± 9.0 i	232.9	± 162.9 de
Killarney red	6.6	± 2.6 a-f	33.2	± 8.4 b-f	13.4	± 1.2 d-g	7.4	± 1.2 cd	4.6	± 1.3 g-i	15.3	± 6.8 i	438.1	± 194.4 a
Marino	3.3	± 3.0 d-f	23.1	± 9.4 fg	11.5	± 2.3 gh	6.4	± 1.7 d-f	3.5	± 1 g-i	12.8	± 3.8 i	220.2	± 71.6 de
Mountain top	9.5	± 0.9 ab	35.7	± 8.8 b-d	13.7	± 1.4 c-f	7.7	± 1.4 cd	4.7	± 1 g-i	27.7	± 8.2 i	279.7	± 91.7 b-d
Penn wonder	7.4	± 1.4 a-e	30.1	± 7.0 d-f	13.0	± 1.0 e-g	6.2	± 0.9 d-f	4.9	± 1.5 g-i	16.9	± 9.7 i	319.9	± 134.6 bc
Silverskin														
Silver Rose	3.6	± 2.9 d-f	24.9	± 7.9 fg	11.5	± 1.5 g	10.9	± 1.2 b	2.3	± 0.8 i	-	- -	-	- -
Silver White	6.2	± 2.9 a-f	31.5	± 15.1 c-f	12.4	± 2.4 g	12.3	± 1.4 a	2.5	± 1.3 i	-	- -	-	- -
Turban														
Maiskij	7.1	± 3.4 a-f	32.5	± 11.9 b-f	14.0	± 1.4 b-f	5.1	± 1.4 f-h	7.4	± 2.9 ef	102.5	± 44.0 g	43.0	± 22.5 gh
Portugese	7.5	± 1.1 a-e	29.5	± 7.2 d-g	12.9	± 1.4 fg	7.5	± 1.7 cd	4.0	± 0.7 g-i	150.4	± 53.1 de	15.1	± 5.3 h
Max	12.1		45.5		15.8		12.3		15.8		315.9		438.1	
Average	6.6		32.7		13.4		6.3		7.5		121.4		133.3	
Min	1.4		15.4		9.8		2.2		2.3		12.8		4.0	

Table 3.3: Bulb yield and yield components for 36 garlic cultivars grown at Saint-Anne-de-Bellevue, Qc, Canada in 2020 (S20). Means in a given column followed by different letters are significantly different ($LSD_{0.05}$).

Cultivar	Yield (ton/ha) Mean	Yield (ton/ha) sd	Bulb Weight (g) Mean	Bulb Weight (g) sd	Bulb Circumference (mm) Mean	Bulb Circumference (mm) sd	Clove Number per Bulb Mean	Clove Number per Bulb sd	Clove Weight (g) Mean	Clove Weight (g) sd	Bulbils Number per Spathe Mean	Bulbils Number per Spathe sd	Bulbils Weight (mg) Mean	Bulbils Weight (mg) sd
Artichoke														
Italian	7.2	± 0.9 k-p	27.9	± 10.5 j-l	13.1	± 2.0 g-j	10.1	± 2.8 cd	2.8	± 0.9 lm	6.4	± 2.9 n	536.6	± 93.1 a
Asiatic														
Tibetan red	6.5	± 1.4 l-q	26.1	± 10.8 k-n	12.5	± 2.1 i-l	7.4	± 1.2 g	3.7	± 1.1 jkl	29.1	± 9.6 n	249.9	± 83.3 d
Tibétain red	6.0	± 1.1 n-r	21.5	± 6.7 m-o	11.6	± 1.4 lm	6.3	± 1.3 h-j	3.5	± 1.1 jkl	32.5	± 8.2 mn	212.9	± 43.9 e
Creole														
Ajo Rojo	0.4	± 0.1 s	5.3	± 1.9 q	6.3	± 0.4 o	2.2	± 0.5 o	2.5	± 1.2 lm	-	- -	-	- -
Creole Red	2.1	± 0.7 s	12.3	± 8.6 pq	9.3	± 2.3 n	6.0	± 3.1 i-k	2.6	± 2.7 lm	10.1	± 35.2 n	2.1	± 6.7 n
Rose de Lautrec	8.7	± 1.1 gl	33.5	± 15.1 f-i	13.5	± 2.6 e-i	9.2	± 2.0 e	3.6	± 1.4 jkl	87.4	± 34.2 gh	23	± 6.7 mn
Marble Purple Stripe														
Duganskij	10.4	± 0.8 dg	36.5	± 15.4 e-g	14.4	± 2.4 c-e	5.3	± 1.2 k-m	7.1	± 3.1 h	39	± 17.3 l-n	124.7	± 53.0 hi
Eureka Myrtis	14.4	± 1.6 a	49.1	± 12.3 bc	16.5	± 1.8 ab	5.3	± 0.8 k-m	9.3	± 2.1 ef	99.9	± 39.2 g	88.7	± 41.5 jk
Kostyn's Red Russian	12.9	± 2.5 a-c	54.1	± 17 ab	17.4	± 2.2 a	5.2	± 1.1 k-m	11.1	± 4.6 cd	66.9	± 31.1 h-k	136.9	± 38.5 gh
Metechi	11.0	± 1.8 c-f	39.8	± 12.8 c-f	14.8	± 1.8 cd	5.8	± 1.0 i-l	7.0	± 2.0 h	71.8	± 55.3 i-l	124	± 68.3 g-i
Metechi-2	10.0	± 2.0 e-i	35.2	± 10.3 e-i	13.9	± 1.8 d-h	10.6	± 1.7 bc	3.4	± 1.2 j-m	152.4	± 68.9 de	54	± 69.4 k-m
Northern Siberian	10.3	± 0.8 d-g	36.3	± 9.5 e-g	14.7	± 1.6 cd	4.6	± 1.2 mn	8.9	± 4.6 fg	77.5	± 46.1 hi	83.2	± 25.5 j-l
Siberian red	12.2	± 0.6 b-d	42.9	± 14.1 cd	15.4	± 1.9 c	5.3	± 1.1 k-m	7.9	± 2.2 gh	62.1	± 20.5 i-k	99.3	± 28.7 ij
Wenger's Russian	15.0	± 2.0 a	53.1	± 15.9 ab	16.8	± 2.3 ab	5.6	± 1.2 j-l	10.0	± 4.1 d-f	70.9	± 43.8 h-j	126.3	± 53.9 hi
Georgian Crystal	8.7	± 0.8 g-l	31.4	± 5.8 g-k	13.6	± 1.0 e-i	3.2	± 0.9 o	10.7	± 3.5 c-e	198.1	± 51.2 c	38.9	± 9.0 mn
Porcelain														
German White	9.4	± 2.0 f-j	35.9	± 6.8 e-h	14.4	± 1.1 c-e	3.4	± 0.7 o	10.8	± 2.4 cd	186.6	± 51.6 cd	42.3	± 13.6 l-n
Leningrad	11.9	± 1.0 b-e	40.5	± 7.5 c-e	14.8	± 1.1 cd	3.4	± 0.9 o	12.6	± 2.5 b	240.8	± 72.2 b	52.8	± 13.5 lm
Music	13.2	± 0.9 ab	54.5	± 13.3 a	16.3	± 1.5 b	3.6	± 0.7 o	15.4	± 3.8 a	269	± 71.4 a	57.4	± 16.8 lm
Music-2	13.4	± 1.2 ab	51.2	± 18.5 a-c	15.9	± 2.3 bc	8.2	± 1.0 fg	6.4	± 2.9 hi	39.5	± 6.4 k-n	334.5	± 58.1 c
Northen Quebec	9.4	± 0.6 f-j	32.8	± 8.1 f-j	13.8	± 1.3 d-h	3.3	± 0.8 o	10.2	± 2.7 d-f	237.4	± 61.8 b	46.1	± 14.1 l-n
Susan Delafield	10.2	± 1.7 d-h	36.9	± 8.2 d-e	14.4	± 1.4 c-e	3.1	± 0.7 o	11.9	± 2.3 bc	181.3	± 59.9 cd	46.9	± 9.4 l-n
Ukrainian Mavniv	8.1	± 1.1 h-n	27.2	± 6.9 j-m	12.8	± 1.2 h-k	3.7	± 0.6 no	7.5	± 1.8 h	192.7	± 35.1 c	39.4	± 8.2 mn
Yugoslavian Porcelain	8.6	± 1.2 g-m	36.0	± 12.2 e-h	14.1	± 2.1 d-g	3.6	± 1.5 o	11.3	± 5.1 b-d	198.9	± 51.2 c	39.3	± 9.0 mn
Purple Stripe														
Chesnok Red	6.5	± 1.0 l-q	23.9	± 6.1 l-o	12.4	± 1.3 i-l	11.7	± 1.4 b	2.1	± 0.5 m	139.4	± 32.5 ef	26.1	± 5.5 mn
Persian star	7.2	± 0.1 k-p	27.1	± 10.1 j-n	12.8	± 1.7 h-k	9.3	± 1.9 de	2.9	± 1.0 k-m	129.7	± 53.8 f	53.3	± 20.2 lm
Rocambole														
Baba Franchuk's	7.8	± 1.3 j-p	28.9	± 9.6 i-l	13.1	± 1.5 g-j	8.0	± 1.2 fg	3.6	± 1.0 j-l	34.3	± 10.6 l-n	197.1	± 71.0 ef
French Rocambole	8.4	± 1.2 g-m	29.8	± 6.7 h-k	13.1	± 1.2 g-j	7.1	± 0.7 gh	4.2	± 1.0 ij	28.4	± 10.0 n	171.5	± 56.5 f
German Red	5.7	± 1.3 o-r	24.4	± 5.9 k-o	12.0	± 1.2 k-m	7.9	± 1.1 fg	3.2	± 1.0 j-m	33.1	± 11.3 l-n	198.7	± 61.9 ef
Italian Purple	5.0	± 1.2 qr	20.9	± 6.7 no	11.9	± 1.6 k-m	8.4	± 2.1 ef	2.6	± 0.9 lm	19.6	± 9.1 n	259.8	± 115.7 d
Killarney red	7.8	± 1.6 i-o	28.6	± 6.8 i-l	13.1	± 1.1 g-j	8.4	± 1.0 f	3.5	± 0.9 j-l	20.7	± 15.0 n	326.2	± 148.8 c
Marino	5.7	± 0.6 p-r	24.1	± 7.9 k-o	12.4	± 1.4 j-l	6.6	± 1.3 g-i	3.7	± 1.2 j-l	20.9	± 7.5 n	191.2	± 77.4 ef
Mountain top	8.5	± 0.5 g-m	31.2	± 9.1 g-k	13.4	± 1.3 e-i	7.6	± 1.3 fg	4.1	± 0.9 i-k	31.7	± 9.9 mn	276.6	± 90.1 cd
Penn wonder	7.9	± 1.7 i-o	29.2	± 6.7 i-l	13.3	± 1.2 f-j	7.1	± 1.1 gh	4.1	± 0.8 i-k	20	± 7.1 n	273.3	± 70.5 cd
Silverskin														
Silver Rose	6.5	± 0.9 m-q	27.8	± 11.4 j-l	12.5	± 1.9 i-l	14.7	± 3.1 a	1.9	± 0.6 m	-	- -	-	- -
Silver White	10.0	± 0.7 e-j	33.6	± 9.2 f-i	13.7	± 1.4 e-h	14.8	± 2.4 a	2.3	± 0.6 m	41	± 19.2 k-n	171.1	± 91.7 fg
Turban														
Maiskij	9.3	± 2.6 f-k	34.4	± 15.8 f-i	14.1	± 2.5 d-f	5.0	± 1.3 lm	7.1	± 3.2 h	51.3	± 15.8 j-m	124.8	± 51.3 hi
Portugese	8.8	± 1.0 f-k	30.9	± 7.0 g-k	13.3	± 1.2 f-j	10.9	± 1.4 b	2.9	± 0.7 lm	149.7	± 52.5 e	23	± 7.0 mn
Unclassified														
Sweet Haven	4.3	± 1.4 r	18.2	± 7.0 op	10.8	± 1.6 m	6.5	± 1.4 g-j	3.0	± 1.1 j-m	10.9	± 2.4 n	398.9	± 141.3 b
Max	15.0		54.5		17.4		14.8		15.4		269		536.6	
Average	8.7		32.4		13.5		6.8		6.1		91.1		145.9	
Min	0.4		5.3		6.3		2.2		1.9		6.4		2.1	

Table 3.4: Bulb yield and yield components of 36 garlic cultivars grown at L'Ascension-de-Notre-Seigneur, Qc, Canada in 2019 (N19). Means in a given column followed by different letters are significantly different ($LSD_{0.05}$).

Cultivar	Yield (ton/ha) Mean	sd	Bulb Weight (g) Mean	sd	Bulb Circumference (mm) Mean	sd	Clove Number per Bulb Mean	sd	Clove Weight (g) Mean	sd	Bulbils Number per Spathe Mean	sd	Bulbils Weight (mg) Mean	sd
Artichoke														
Italian	2.0	± 0.3 fg	10.4	± 6.4 h-j	8.5	± 2.1 k-m	5.9	± 3.1 bc	1.8	± 0.8 jk	-	- -	-	- -
Creole														
Ajo Rojo	1.2	± 0.2 g	7.3	± 1.9 j	8.1	± 0.9 m	7.9	± 1.9 a	1.0	± 0.3 k	-	- -	-	- -
Creole Red	1.7	± 0.4 g	8.6	± 2.9 ij	8.3	± 1.5 lm	7.7	± 3.7 a	1.2	± 0.4 k	-	- -	-	- -
Rose de Lautrec	2.6	± 1.1 fg	14.2	± 5.2 f-i	9.6	± 1.5 h-l	6.0	± 2.7 b	3.0	± 2.2 i-k	77.8	± 30.6 de	30.7	± 3.6 f-h
Marble Purple Stripe														
Duganskij	9.4	± 0.5 a	32.9	± 7.3 ab	14.2	± 1.3 a	3.8	± 1.6 d-g	9.8	± 3.8 ef	70.7	± 21.5 e	69.8	± 21.0 d-h
Eureka Myrtis	6.4	± 1.1 a-f	28.5	± 8.4 bc	12.6	± 1.2 bc	2.9	± 1.1 f-i	10.6	± 3.4 def	98.4	± 30.3 c-e	78.2	± 22.0 d-h
Kostyn's Red Russian	7.2	± 0.5 a-e	25.5	± 7.7 cd	11.6	± 1.4 c-g	2.1	± 0.3 i	12.2	± 3.3 a-d	78.2	± 34.7 de	92.5	± 26.7 d-g
Metechi-2	2.1	± 0.1 fg	12.7	± 2.8 f-j	9.0	± 0.9 i-m	5.8	± 0.5 b-d	2.2	± 0.6 i-k	95.0	± 5.6 c-e	18.3	± 1.2 gh
Siberian red	8.2	± 6.8 a-d	31.9	± 20.6 bc	12.6	± 4 bc	3.1	± 1.4 f-i	9.8	± 4.9 ef	65.1	± 27.3 ef	109.0	± 39.4 de
Wenger's Russian	6.8	± 2.2 a-f	26.3	± 8.7 bc	12.2	± 1.9 cd	2.4	± 0.7 g-i	11.0	± 2.6 de	106.0	± 72.5 cd	86.7	± 25.7 d-g
Porcelain														
Georgian Crystal	7.8	± 0.3 a-d	30.8	± 11.2 bc	12.8	± 2.2 a-c	2.8	± 0.7 f-i	11.2	± 3.9 c-e	245.1	± 36.0 a	29.4	± 8.3 f-h
German White	8.2	± 1.3 a-d	39.6	± 11.0 a	14.1	± 1.7 ab	2.9	± 0.6 f-i	13.8	± 2.4 a-c	223.1	± 50.8 a	44.5	± 11.0 e-h
Leningrad	8.6	± 1.3 a-c	31.0	± 7.6 bc	12.7	± 1.4 abc	2.8	± 0.7 f-i	11.6	± 2.8 b-e	173.5	± 60.2 b	40.0	± 6.5 e-h
Northen Quebec	7.2	± 1.6 a-f	28.0	± 7.8 bc	11.9	± 1.6 c-g	2.7	± 0.8 f-i	10.6	± 3.2 d-f	225.1	± 51.4 a	36.1	± 8.1 e-h
Ukrainian Mavniv	8.8	± 4.1 ab	32.4	± 11.1 ab	12.5	± 2.1 bc	2.4	± 0.7 g-i	14.4	± 6.5 a	163.4	± 55.6 b	43.7	± 16.8 e-h
Yugoslavian Porcelain	6.6	± 2.2 a-f	27.8	± 10.6 bc	12.0	± 1.9 c-e	2.0	± 0.1 i	13.9	± 5.3 ab	182.0	± 35.6 b	36.4	± 6.9 e-h
Purple Stripe														
Chesnok Red	4.5	± 3.8 c-g	19.8	± 8.5 d-f	10.8	± 1.9 d-h	7.5	± 1.5 a	2.6	± 0.8 i-k	111.0	± 49.5 c	14.9	± 5.0 h
Rocambole														
Baba Franchuk's	4.8	± 1.5 b-g	16.7	± 6.3 f-h	10.4	± 1.4 f-i	4.7	± 1 b-e	3.6	± 1.0 ij	27.9	± 14.0 g	271.7	± 158.1 bc
French Rocambole	7.2	± 0.4 a-f	25.2	± 6.5 c-e	12.0	± 1.2 c-f	4.1	± 1.1 d-f	6.8	± 4 gh	32.8	± 6.1 fg	232.8	± 55.6 c
Italian Purple	4.2	± 2.2 d-g	16.0	± 7.7 f-h	9.7	± 1.9 h-k	3.7	± 1.6 e-h	6.0	± 5 gh	37.8	± 18.7 fg	131.9	± 56.1 d
Marino	2.6	± 1.7 fg	13.7	± 8.3 f-i	9.7	± 2.3 h-l	5.1	± 1.2 b-d	2.6	± 1.3 i-k	28.5	± 12.2 g	102.6	± 70.6 d-f
Mountain top	4.3	± 0.5 d-g	15.2	± 4.5 f-h	10.0	± 1.3 h-j	4.6	± 0.9 c-e	3.3	± 0.8 ij	27.5	± 6.3 g	264.2	± 99.4 c
Penn wonder	5.4	± 0.5 a-g	14.5	± 6.1 f-h	9.7	± 1.8 h-k	5.0	± 0.8 b-d	2.9	± 1.3 i-k	20.2	± 8.5 g	331.5	± 210.5 ab
Silverskin														
Silver Rose	3.3	± 1.0 e-g	12.6	± 4.0 g-j	8.9	± 1.2 j-m	7.5	± 2.8 a	1.8	± 0.5 jk	-	- -	-	- -
Silver White	4.5	± 2.1 c-g	18.5	± 6.7 e-g	10.2	± 1.6 g-j	7.7	± 1.1 a	2.4	± 0.7 i-k	-	- -	-	- -
Turban														
Maiskij	5.2	± 0.5 a-g	18.1	± 3.7 e-g	10.6	± 1.1 e-i	2.2	± 0.7 hi	8.2	± 1.1 fg	99.7	± 17.1 c-e	62.2	± 22.7 d-h
Portugese	4.3	± 0.1 d-g	15.0	± 5.3 f-h	9.5	± 1.4 i-l	3.5	± 1.2 e-h	4.4	± 1.0 hi	123.9	± 29.1 c	24.8	± 6.0 gh
Unclassified														
Sweet Haven	5.0	± 0.7 b-g	17.5	± 4.6 fg	10.6	± 1.2 e-i	5.1	± 0.7 b-d	3.4	± 0.6 ij	22.6	± 6.5 g	375.0	± 149.3 a
Max	9.4		39.6		14.2		7.9		14.4		245.1		375.0	
Average	5.4		21.1		10.9		4.4		6.6		101.5		109.9	
Min	1.2		7.3		8.1		2.0		1.0		20.2		14.9	

Table 3.5: Bulb yield and yield components of 36 garlic cultivars grown at Alma, Qc, Canada in 2020 (N20). Means in a given column followed by different letters are significantly different ($LSD_{0.05}$).

	Yield (ton/ha)		Bulb Weight (g)		Bulb Circumference (mm)		Clove Number per Bulb		Clove Weight (g)		Bulbils Number per Spathe		Bulbils Weight (mg)	
Cultivar	Mean	sd	Mean	sd	Mean	sd	Mean	sd	Mean	sd	Mean	sd	Mean	sd
Artichoke														
Italian	1.3	± 0.1 ef	18.6	± 0.5 bc	11.9	± 0.5 bc	4.0	± 0.1 c-e	4.7	± 0.1 d-g	-	- -	-	- -
Asiatic														
Tibetan red	3.4	± 1.4 b-f	19.0	± 6.5 bc	11.6	± 1.7 bc	3.8	± 0.9 c-e	4.9	± 1.3 d-f	15.6	± 11.2 hi	360.2	± 246.4 de
Creole														
Rose de Lautrec	3.9	± 0.5 a-f	34.5	± 15.7 a	14.2	± 2.7 a	10.2	± 2.7 a	3.3	± 0.9 fg	101.5	± 57.5 ab	46.3	± 25.6 k
Marble Purple Stripe														
Duganskij	4.5	± 0.9 a-d	23.1	± 7.1 b	12.6	± 1.5 b	4.3	± 1.0 cd	5.5	± 1.7 de	26.7	± 12.7 g-i	225.0	± 101.7 g
Eureka Myrtis	4.7	± 4.0 a-c	23.0	± 10 b	12.5	± 2.4 b	4.2	± 1.0 cd	5.8	± 3.0 de	40.5	± 20.3 fg	222.2	± 101.6 gh
Kostyn's Red Russian	3.1	± 1.2 b-f	20.5	± 9.6 bc	12.1	± 2.0 b	3.6	± 1.2 de	5.8	± 1.9 de	60.5	± 43.6 d-f	168.0	± 73.3 g-i
Metechi	2.3	± 0.1 c-f	14.2	± 5.0 c	10.1	± 1.3 c	3.4	± 1.3 de	4.6	± 1.9 d-g	43.2	± 14.0 e-g	146.0	± 55.9 h-j
Wenger's Russian	6.2	± 0.6 a	26.8	± 11.2 b	13.0	± 2.3 ab	3.7	± 1.2 de	7.8	± 3.4 bc	39.7	± 17.0 fg	248.4	± 92.5 fg
Porcelain														
Georgian Crystal	2.9	± 3.2 b-f	15.5	± 7.1 c	10.3	± 2.2 c	2.5	± 1.0 ef	6.9	± 4.2 b-d	110.6	± 70.5 ab	62.0	± 22.7 i-k
German White	2.5	± 0.4 b-f	20.5	± 7 bc	11.3	± 1.7 bc	2.1	± 0.4 f	9.6	± 3.2 ab	92.0	± 40.9 bc	75.6	± 34.7 i-k
Leningrad	5.2	± 1.6 ab	27.8	± 7.4 ab	13.0	± 1.1 ab	2.3	± 0.6 ef	12.0	± 2.0 a	98.4	± 49.9 a-c	63.2	± 13.8 i-k
Northen Quebec	4.2	± 1.9 a-e	19.7	± 8.4 bc	11.2	± 2.2 bc	2.2	± 0.6 f	9.0	± 3.9 ab	124.4	± 39.8 a	53.1	± 17.6 k
Ukrainian Mavniv	3.2	± 1.6 b-f	20.5	± 7.6 bc	11.1	± 1.7 bc	2.4	± 0.6 ef	8.8	± 3.2 b	129.0	± 50.8 a	76.1	± 34.2 i-k
Yugoslavian Porcelain	1.6	± 0.9 ef	15.6	± 3.1 c	10.3	± 0.6 c	2.7	± 0.9 ef	6.2	± 1.7 cd	94.5	± 48.5 bc	52.6	± 12.9 k
Purple Stripe														
Chesnok Red	1.3	± 0.6 f	8.6	± 0.5 c	8.5	± 0.5 c	6.0	± 0.5 bc	1.4	± 0.3 g	83.6	± 45.5 b-d	57.6	± 19.3 jk
Persian star	2.4	± 1.4 b-f	16.8	± 5.0 c	10.9	± 1.0 bc	5.9	± 1.2 c	2.9	± 0.8 g	68.7	± 25.8 c-e	52.4	± 10.0 k
Rocambole														
Baba Franchuk's	3.2	± 1.7 b-f	19.5	± 11.8 bc	11.4	± 2.3 bc	4.8	± 0.8 c	4.1	± 2.4 e-g	20.0	± 1.9 g-i	492.8	± 47.5 bc
French Rocambole	3.3	± 0.8 b-f	16.6	± 6.2 c	11.2	± 1.4 bc	4.2	± 1.2 cd	4.1	± 1.4 e-g	8.5	± 5.1 i	434.7	± 164.5 cd
Italian Purple	1.9	± 1.5 d-f	13.2	± 2.9 c	10.3	± 0.9 c	4.6	± 0.9 c	2.9	± 0.9 fg	14.1	± 8.4 hi	183.5	± 75.3 g-i
Killarney red	1.3	± 0.2 ef	11.8	± 2.2 c	10.5	± 0.7 bc	5.5	± 0.7 c	2.1	± 0.1 g	14.5	± 11 hi	322.6	± 138.5 ef
Marino	1.1	± 0.5 f	16.9	± 2.8 c	10.7	± 0.4 bc	4.5	± 0.7 cd	3.8	± 1.2 e-g	12.4	± 4.7 hi	399.0	± 188.6 c-e
Mountain top	3.0	± 0.8 b-f	20.6	± 5.3 bc	11.9	± 1.3 bc	4.2	± 0.9 cd	5.0	± 1.1 d-f	11.4	± 4.2 hi	539.6	± 212.1 ab
Penn wonder	2.4	± 0.5 c-f	16.1	± 4.2 c	10.9	± 1.0 bc	3.2	± 0.8 de	5.2	± 1.5 d-f	8.4	± 4.4 i	572.6	± 157.3 ab
Silverskin														
Silver White	1.2	± 0.1 f	13.9	± 0.1 c	9.8	± 0.1 c	8.0	± 0.1 b	1.7	± 0.1 g	-	- -	-	- -
Turban														
Maiskij	2.4	± 0.6 c-f	18.1	± 4.5 bc	11.5	± 1.1 bc	3.7	± 0.8 c-e	4.9	± 0.9 d-f	36.7	± 15.6 f-h	188.0	± 114.5 gh
Portugese	3.4	± 1.2 b-f	16.9	± 5.0 c	10.4	± 1.4 c	5.9	± 1.5 c	2.9	± 0.7 g	72.1	± 30.0 cd	46.7	± 17.2 k
Unclassified														
Sweet Haven	1.4	± 0.4 ef	19.0	± 2.5 bc	11.4	± 0.5 bc	4.0	± 0.1 c-e	4.8	± 0.6 d-g	6.5	± 2.1 i	690.0	± 328.1 a
Max	6.2		34.5		14.2		10.2		12.0		129.0		690.0	
Average	2.9		18.8		11.3		4.3		5.2		53.3		231.1	
Min	1.1		8.6		8.5		2.1		1.4		6.5		46.3	

3.3.2 Bulb color

In all environments, cultivars of the Porcelain, Creole and Silverskin types had predominantly white bulbs (Fig. 3.2). The Asiatic, Marble Purple Stripe, Purple Stripe, Turban types and the unclassified cultivar Sweet Haven showed a purple stipe motif on an average of 80%of their bulb wrappers. However, Metechi-2 showed less coloration than the rest of Marble Purple Stripe cultivars. If bulb pattern colors were relatively stable across environments for these types, some differences across environments were observed for others. The Artichoke type cultivar Italian produced a majority of white bulbs in the two southern environments (*i.e.*, S19 and S20) while 50 and 100% of the bulbs presented a stripe pattern in N19 and N20, respectively. Other specific cultivars presented a different coloration pattern in a specific environment, for example, the cultivars Chesnok Red, Duganskij, Metechi-2 and Portuguese were predominantly white when grown in N19. Also, the cultivars German Red and Italian Purple showed less than 25% of stripes on their bulbs at S19. In general, Rocambole type had a paler motif than Purple Stripe and Marble Purple Stripe types (data not shown). The environment in which certain types and cultivars are grown thus can impact bulb color.

A

B

Fig 3.2. Bulb wrapper coloration of 36 garlic cultivars and their corresponding type grown in four environments in Québec (Canada) [Saint-Anne-de-Bellevue in 2019 (S19) and 2020 (S20); L'Ascension-de-Notre-Seigneur (N19) and Alma (N20)]. B) Example of Bulb wrapper coloration.

3.3.3 Clove characteristics

Overall, across environments and cultivars, when more cloves were present in a bulb the less they weighted, clove weight ranging between 0.5 and 28 g/clove (Tables 3.2-3.5). In general, cultivars grown in the southern environments (*i.e.*, S19 and S20) tended to produce more cloves per bulbs and more layers of cloves per bulb than the ones grown in the northern environments, except for cultivars of the Porcelain and Marble Purple Stripe types that were more stable across environments and the cultivars Metechi-2 and Music-2 (Table 3.2-3.5, Fig. 3.3). As a matter of fact, cultivars of Porcelain type produced between two and four cloves per bulbs in all environments with the exception of the cultivar Music-2 that produced an average of eight cloves per bulbs. The cultivars Silver White and Silver Rose produced the largest number of cloves at all site with a maximum at 15 cloves per bulb in S20 but only eight cloves per bulb in N19 and N20. Similarly, the Italian cultivar of the Artichoke type produced an average of ten cloves per bulb in the south environments but only half of that in the north. Rocambole type cultivars varied between six to nine cloves per bulb in the south environments to three to five in the north environments, Marble Purple Stripe type varied from three to six cloves in the south to between two and four cloves in the north with the exception of the cultivar Metechi-2 that varied like the Purple Stripe type (8-12 cloves in the south environments, 6-7 cloves in the north ones). In the Turban type, the cultivar Maiskij produced fewer cloves (4-5 cloves in the south, 2-3 in the north) than the cultivar Portuguese (7-12 cloves in the south and 4-6 in the north). Cultivars Creole Red and Rose de Lautrec produced between six and ten cloves per bulbs in all sites while the cultivar Ajo Rojo performed more poorly at S20 (two cloves per bulb) than at N19 (eight cloves per bulb).

Clove arrangement was stable for cultivars of the Porcelain type all having only one layer of cloves in all sites with the exception of Music-2, which had up to two layers (Fig. 3.3). Between 25 and 50% of the Rocambole type cultivars produced two layers of cloves per bulb in the south environments while no two-layer bulbs were produced in the north environments. The Marble Purple Stripe type cultivars also tended to produce bulbs with one layer of cloves but the cultivar Metechi-2 had more than 60% of two-layer bulbs when grown in southern environments. The cultivar Chesnok Red had 40 and 95% of two-layer bulbs in S19 and S20, respectively, while the cultivar Persian Star produced 90% of two layers-bulb at S20. Clove arrangement also switched from more than 75% of three- or four-layer bulbs at S19 and S20 to less than 50% for the cultivar Silver White in the northern environments. The cultivar Ajo Rojo is the only cultivar that had more clove layers when grown in the North compared to South, however, bulbs were often having undesirable cloves with one clove skin for many cloves.

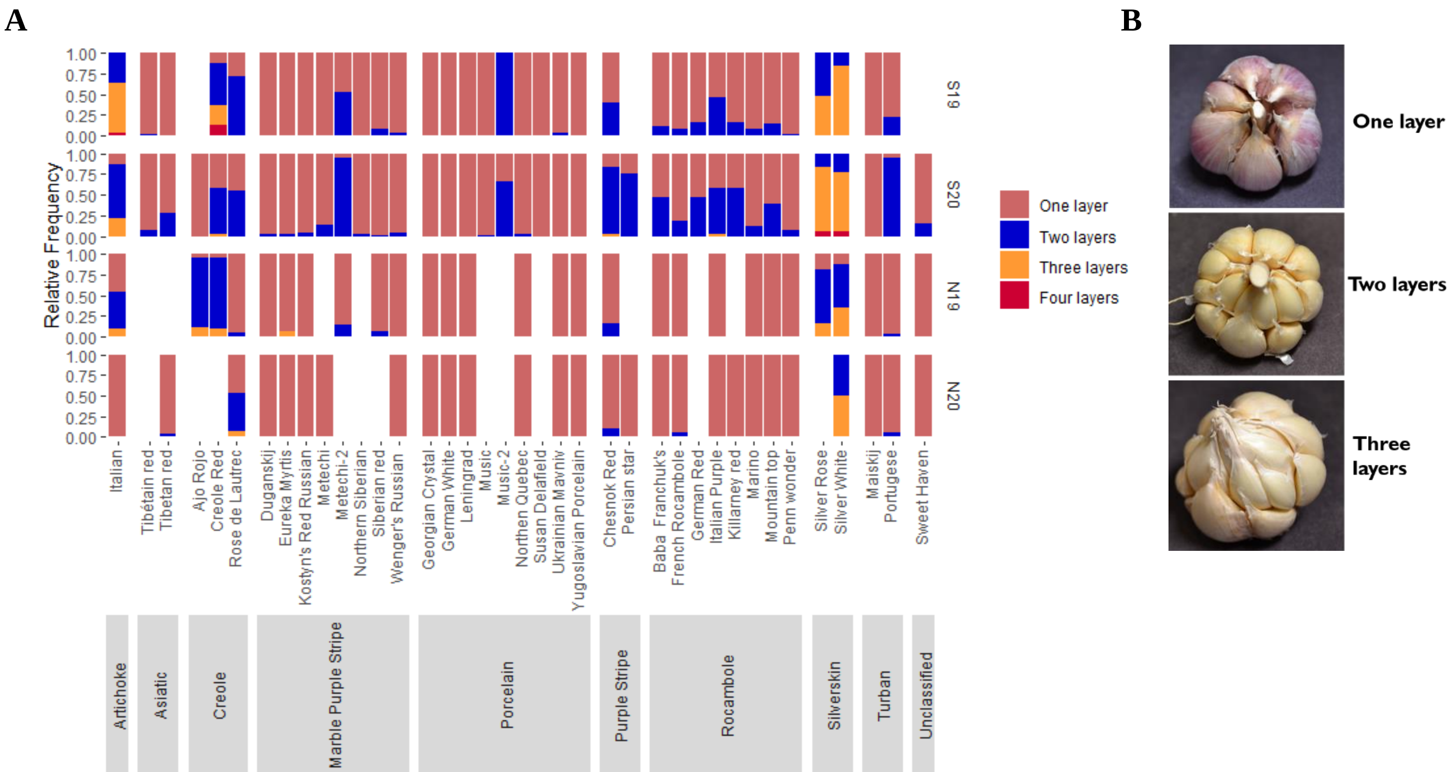


Fig. 3.3 A) Clove arrangement of 36 garlic cultivars and their corresponding type grown in four environments in Québec (Canada) [Saint-Anne-de-Bellevue in 2019 (S19) and 2020 (S20); L'Ascension-de-Notre-Seigneur (N19) and Alma (N20)]. B) Example of clove layers in a garlic bulb

3.3.4 Clove color

Clove color was relatively stable across environments, except for cultivars of the Porcelain and Marble Purple Stripe types (Fig. 3.4). Cultivars of the Rocambole and Asiatic types as well as the unclassified cultivar Sweet Haven all had more than 85% of their bulbs with cream cloves in all environments except for the cultivars French Rocambole and Baba Franchnuk that had a slightly lower proportion of cream color cloves (70%) in the North environments. Cultivars of the Porcelain type produced a majority of light pink cloves but in the southern environments approximately 40% of the bulb had darker cloves (pink or purple) while in the northern environments around 30% of the bulbs had paler cloves (white).Specifically, in the southern environments the cultivars Georgian Crystal, German White, Leningrad and Music had more than 75% of white or light pink cloves, the cultivars Northern Quebec, Susan Delafield and Ukrainian Mavnir had between 25 and 50% of light color cloves, while Music-2 and Yugoslovian Porcelain had (50 to 75%) of darker cloves. Cultivars of the Marble Purple Stripe type, namely the cultivars Duganskij, Eureka Myrtis, Kostyn's Red Russian, and Siberian Red tended to be darker in the northern environments compared to southern ones with nearly 50% of the cloves being purple rather than pink. The cultivar Wenger's Russian showed this pattern only in N19. The cultivar Metechi had 75% of light pink clove at S19 while Metechi-2 showed that pattern in S20. The cultivars Chesnok Red and Persian Star had less than 25 % of darker cloves in all environments. The softneck cultivars (*i.e*. Artichoke and Silverskin type) produced white cloves but the cultivar Silver White often produced a light pink coloration in its middle cloves in S19 and N20. The cultivar Rose de Lautrec had more than 90% of bulbs with light pink cloves while the cultivars Creole Red and Ajo Rojo had darker pink cloves.

Fig. 3.4 A) Clove color of 36 garlic cultivars and their corresponding type grown in four environments in Québec (Canada) [Saint-Anne-de-Bellevue in 2019 (S19) and 2020 (S20); L'Ascension-de-Notre-Seigneur (N19) and Alma (N20)]. B) Example of clove color

3.3.5 Scape structure and bulbils characteristics

Scape production was a very stable characteristic except for the softneck cultivars Silver White and Italian that each produced a scape and bulbils on 10 plants out of 60 in S20 (data not shown). Also, in that specific environment 20 plants of the cultivar Italian produced bulbils through the pseudostem. In the weakly bolting Creole type, the cultivar Rose de Lautrec acted like an hardneck cultivar while cultivars Ajo Rojo and Creole Red had unclear patterns. The cultivar Ajo Rojo did not produce any bulbils but showed signs of a vestigial hard stem in their bulbs. The cultivar Creole Red showed the same pattern except for one plant out of 34 in S20 that produced a complete scape and underdeveloped bulbils and 2 plants in S19 out of 10.

In general, there was an inverse relationship between bulbils number and bulbil weight, the more bulbils being produced the smaller their individual weight was (Table 3.2-3.5). Overall, the Porcelain types produced the highest number of bulbils and Rocambole the lowest. The number of bulbils produced was consistent between environments with the exception of N20. In that environment, bulbils production was approximately 50% lower except for the cultivar Rose de Lautrec that produced a similar number of bulbils when compared to other environments. Overall, with the exception of N20, Porcelain type cultivars produced more than 150 bulbils per spathe, Marble Purple Stripe type cultivars had between 50 and 100 bulbils and Rocambole and Asiatic types and the unclassified Sweet Haven cultivar less than 40. The cultivars Chesnok Red, Metechi-2 and Persian Star produced between 110 and 150 bulbils. Also, Music-2 produced fewer bulbils than other Porcelain type cultivars (*i.e.*, 25-40 bulbils). In the Turban type, the cultivar Maiskij produced a similar number of bulbils than Marble Purple Stripe type while the Portuguese cultivar produced a comparable number of bulbils as Purple Stripe type. When cultivars in the Creole type produced a complete flower stem, the bulbils number ranged between 75 and 120 per spathe.

The bulbils shape was consistent between environments (Fig. 3.5). Porcelain type with the exception of the cultivar Music-2 and Purple Stripe type as well as the cultivar Portuguese (Turban type), Rose de Lautrec (Creole type) and Metechi-2 (Marble Purple

Stripe type) produced rice shape bulbils on more than 80% of their plants. The cultivar Music-2 had corn shape bulbils similar to Marble Purple Stripe type and the cultivar Maiskij (Turban type) on more than 80% of their plants. Asiatic and Rocambole types varied between a drop shape and a corn shape. In N19 and N20, bulbils tended to have a drop shape (average of 80% drop shape) compared to S20 and N19 where less 40% of the plants produced drop shaped bulbils.

Bulbils coloration was less stable between environment than bulbils shape with a more important difference in N20 where bulbils tended to be darker (Fig. 3.6). In general, Porcelain type and the cultivar Rose de Lautrec had the palest bulbils being mostly white (average of 80%) at S19 and N19 and with a light pink coloration at the tip in S20 and N20 (average of 70%). In contrast, the cultivar Music-2 only had darker bulbils either purple in S19 or pink in S20. Marble Purple stripe type tended to have the darkest bulbils of all types with an average of 75% of purple bulbils with the exception of Metechi-2 that had a paler purple coloration similar to Purple Stripe type on 80% of its plants. Cultivars of Rocambole type had a bulbil coloration varying between pink and purple being generally paler in S19 (less than 30% of purple) and darker in N20 (more than 50% of purple). However, in this type the cultivar Penn Wonder produced purple bulbils in all environments. The cultivars Sweet Haven and Maiskij as well as cultivars of the Asiatic type had similar coloration patterns as cultivars of the Rocambole type. Not enough plants from the typically softneck garlic produced bulbils to observe a clear trend in terms of the characteristic of their bulbils.

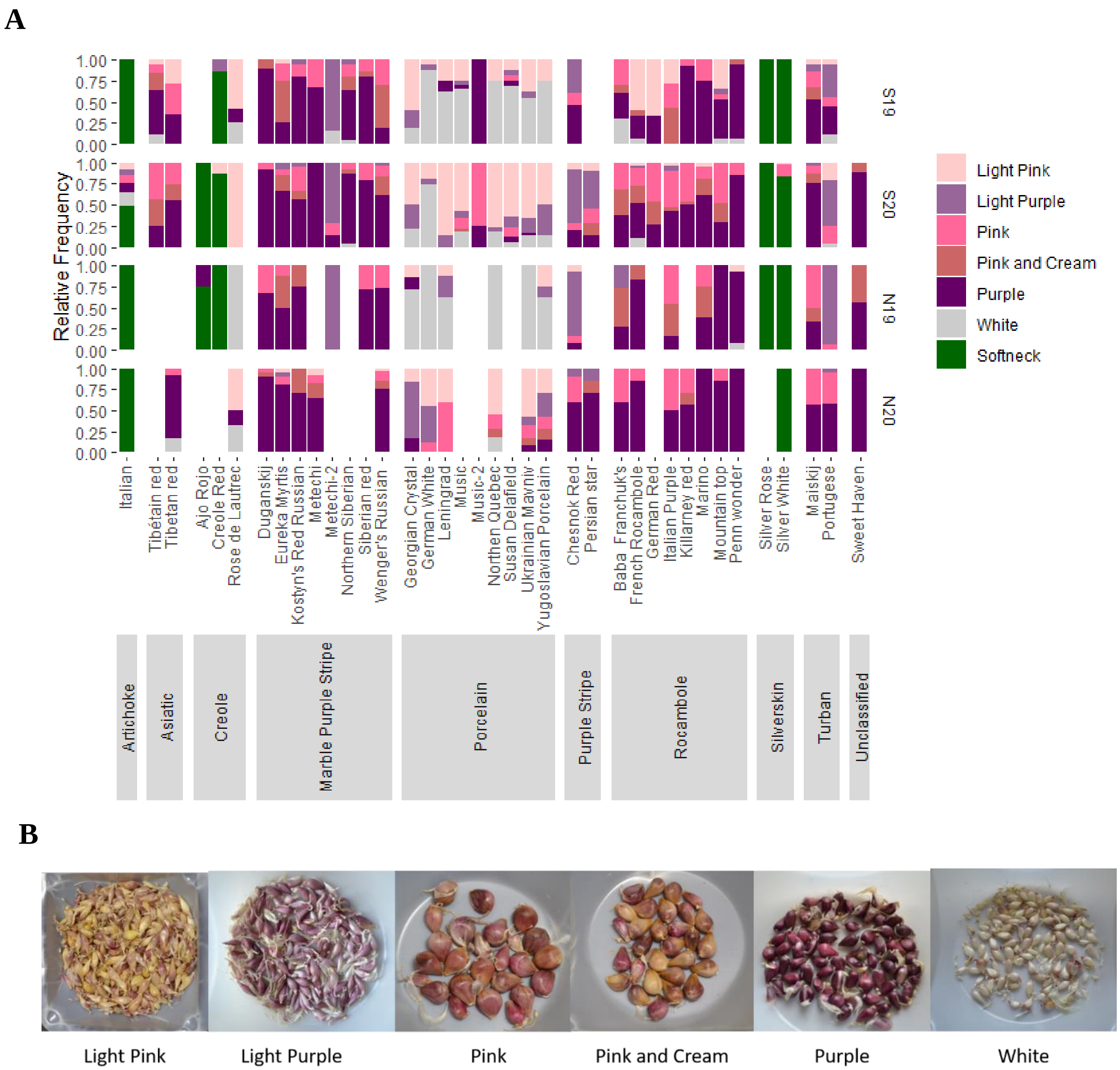
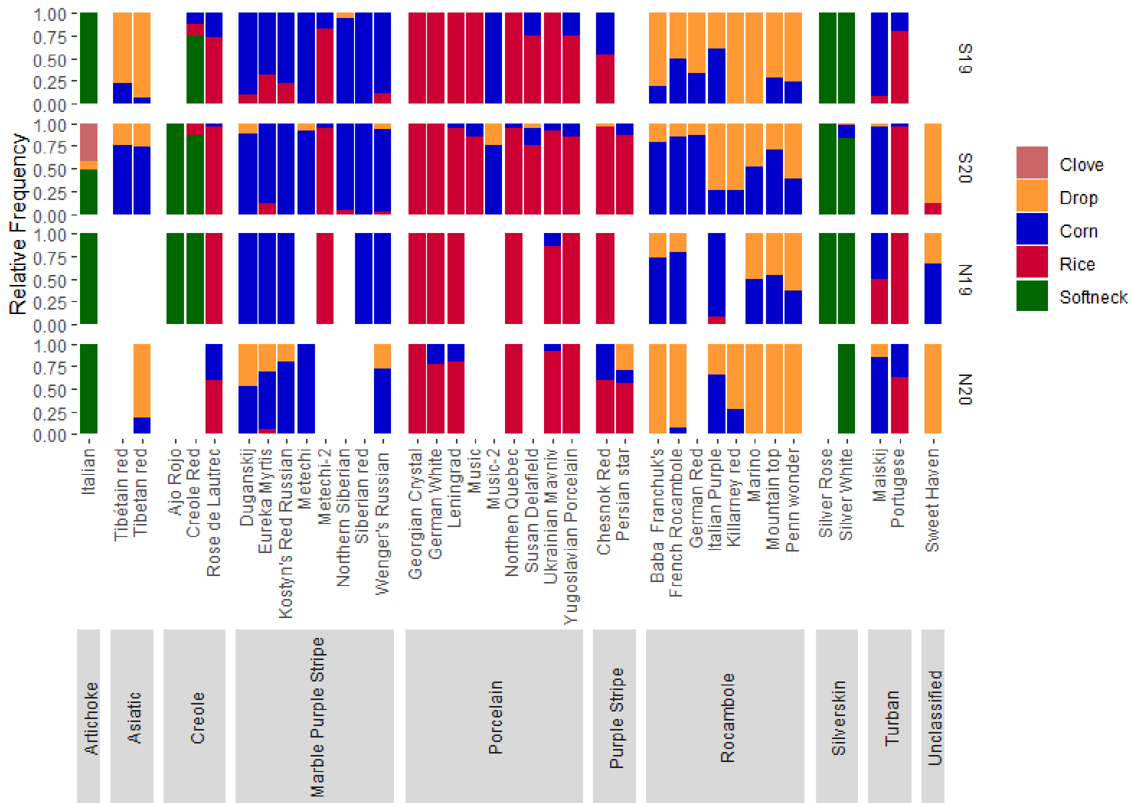


Fig. 3.5 A) Bulbils coloration of 36 garlic cultivars and their corresponding type grown in four environments in Québec (Canada) [Saint-Anne-de-Bellevue in 2019 (S19) and 2020 (S20); L'Ascension-de-Notre-Seigneur (N19) and Alma (N20)]. B) Example of bulbils color. Note: Softneck cultivars predominantly failed to produce scape and thus bulbils. For these cultivars the green color refers to plants having not produced bulbils.

A



B

Fig. 3.6 A) Bulbils shape of 36 garlic cultivars and their corresponding type grown in four environments in Québec (Canada) [Saint-Anne-de-Bellevue in 2019 (S19) and 2020 (S20); L'Ascension-de-Notre-Seigneur (N19) and Alma (N20)]. B) Example of bulbils shape. Note: Softneck cultivars predominantly failed to produce scape and thus bulbils. For these cultivars the green color refers to plants having not produced bulbils.

3.3.6 Plant characteristics

In general, plants from the Porcelain type were taller with a larger pseudostem diameter and a longer scape and spathe than the other types (Supplemental Tables 1-4). Marble Purple Stripe type had slightly smaller plants than Porcelain type. The other types had similar plant measurements with the exception of the cultivars Creole Red and Ajo Rojo of the Creole type that were significantly smaller.

The number of leaves per plant was stable between 6 and 8 with the exception of cultivars from the Silverskin type that produced more (*i.e*., up to 10) (Supplemental Tables 1-4). Leaves were generally more flexible and erect for Artichoke, Asiatic, Rocambole type and the cultivars Silver White, Sweet Haven, Ajo Rojo and Creole Red, while the other types had semi-erect rigid leaves. In terms of pseudostem coloration, Porcelain type as well as the cultivars Chesnok Red, Metechi-2, Persian Star and Portuguese had no anthocyanin coloration (Supplemental Tables 5-8). In contrast, the cultivars Italian, Maiskij, Sweet Haven and the Asiatic type had coloration in more than 90% of their pseudostem. Rocambole and Silverskin types had coloration on 60% of their pseudostem. The overall flowering pattern showed that Rocambole and Porcelain type had flowers that curled more than Purple Stripe and Marble Purple Stripe types (Supplemental Figures 1-4). The form of the bulb and the position of the stem base was not consistent nor precise enough to detect variation among cultivars or groups (data not show).

3.3.7 Cluster analysis

A cluster analysis based on all morphological characteristics (n= 28) observed in this study was conducted to determine the relationship between cultivars. Upon analysis, cultivars grouped into five clusters in S19 while six clusters were observed in the three other environments (*i.e.*, N19, S20 and N20) (Fig. 3.7). Overall, Porcelain type always cluster together. Asiatic type and the unclassified cultivar Sweet Haven clustered with cultivars of the Rocambole type. The cultivar Maiskij from the Turban type was closer to the Marble Purple Stripe type but the cultivar Portuguese is closer to cultivars of the Purple Stripe type and Metechi-2. However, in S19 the cultivar Chesnok Red clustered with the Marble Purple Stripe type and at N19 Maiskij clustered with the Rocambole type. Softneck cultivars grouped together and were close to the cultivars Creole and Ajo Rojo. Rose de Lautrec as well as Music-2 clustered differently in each environment. Overall, the types described by Engeland (1991) were almost always clustering together.

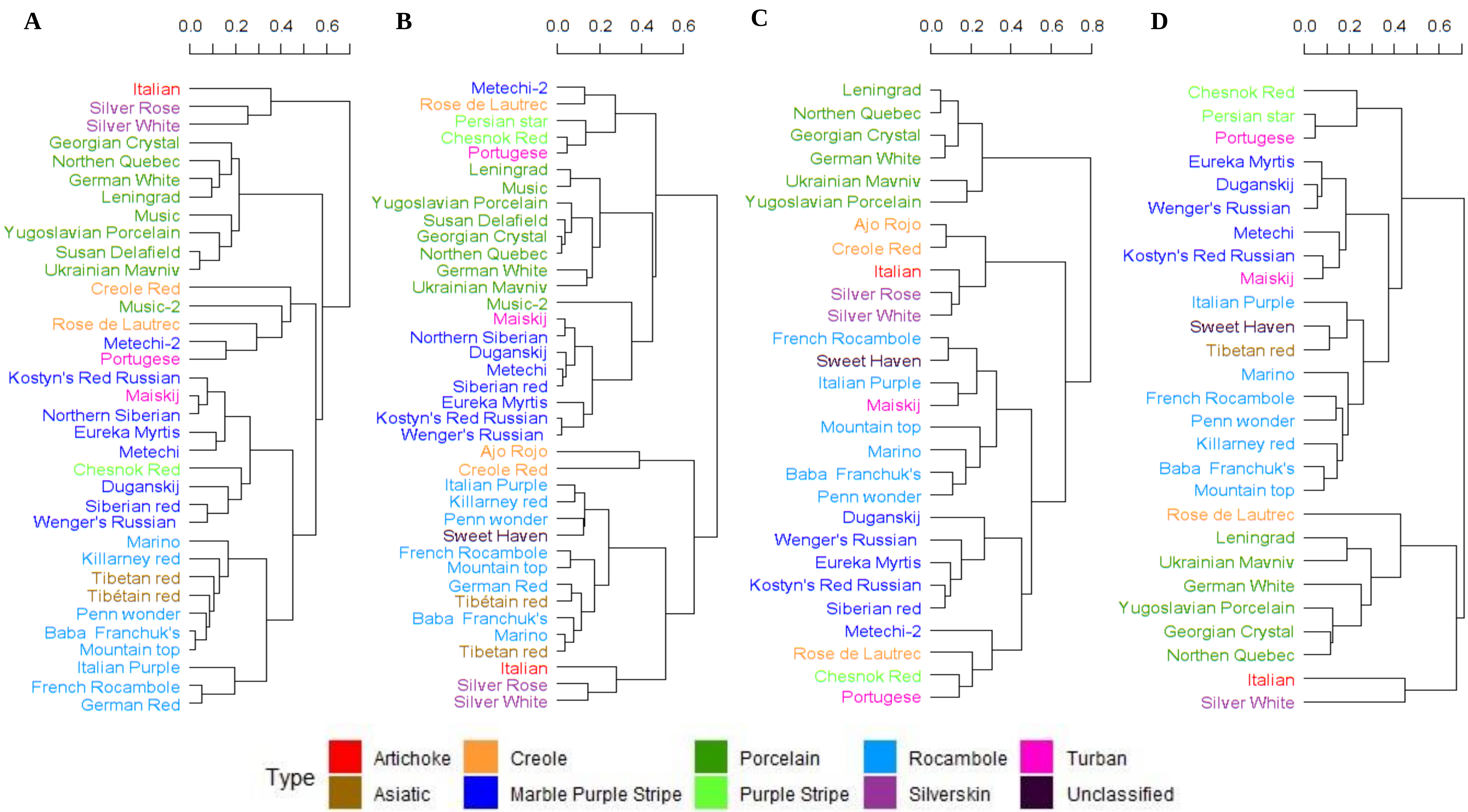


Fig 3.7. Classification using Gower's distance on 28 morphological characteristics for 36 garlic cultivars colored by type growing in Québec (Canada) A) in 2019 at Saint-Anne-de-Bellevue (S19), B) in 2020 at Saint-Anne-de-Bellevue, C) in 2019 at L'Ascension-de-Notre-Seigneur (N19) and D) in 2020 at Alma (N20).

3.4 Discussion:

3.4.1. Performance of cultivars

In the last decades, the cultivar Music has been the most commonly grown by garlic producers in Québec due to its production of consistently high yields and its adaptation to most regions of the province (Landry and Khanizadeh, 1994; Beaudoin, 2019). In recent years, several producers, however, have expressed their interest in expanding their production to include other cultivars in order to gain a bigger share of the local market by expending the harvest range and conservation capacity of their production, but also to meet consumer demand for a greater diversity in garlic characteristics. Because of the dominance of the cultivar Music locally, there is limited knowledge on the other cultivars named in the Seed of Diversity Catalogue in Canada (2020). This lack of information on adaptation, and agronomic as well as morphological characteristics is one key limitation to a greater diversification of the local garlic market. Garlic phenotypic and genotypic variability has been reported in several studies conducted in many areas of the world (Panthee *et al.*, 2006; Sandhu *et al.*, 2015; Wang *et al.*, 2016; Singh *et al.*, 2018). However, many of the studies investigating phenotypic variability of a range of garlic cultivars have been conducted in a limited number of environments, often even in a single environment. Such studies provide limited information on the stability of cultivars' performance and their phenotypes given the large phenotypic plasticity and genotype by environment interaction reported for this species (Hoogerheide *et al.*, 2017).

In this study, garlic was grown in four contrasting environments, differing geographically and environmentally. Geographically, environments from the south of the province used in the present study have a warmer winter with a small snow cover and early spring, compared to the northern environments used. Environmentally, the 2019 growing season was characterized by greater precipitations than the 30-yr average, while the beginning of the 2020 season was characterized by the occurrence of a drought (Environment Canada, 2021). The environments included in the present study thus varied considerably and large overall differences in the performance of the cultivars evaluated in the different environments were observed. Not only were overall differences between environments observed but also variation between replicates in each of the environments, except for S20 (Tables 2-5). Bulb weight at harvest can be affected by the initial planting stock size and this variable was not controlled in this study (Castellanos *et al.*, 2004). Bulb yield, bulb weight and circumference are among the phenotypic characters that varied the most between environments in accordance to the finding of Panthee *et al.* (2006), Volk and Stern (2009), de Lucena *et al.* (2016). The drought we observed in 2020 could in part explain the overall poor bulb yield and bulb weight observed in N20 when compared to S20, given that plots in this later environment were irrigated thus reducing the impact of the prevailing drought. Lack of precipitation was previously reported by others to result in plant loss and decreased bulb size (Kamenetsky, 2007). The large variation in yield and yield components observed within a site has limited our capacity to differentiate cultivars solely on those traits, except at S20.

Even if often large differences were observed in the performance of cultivars across environments, some consistently performed well in most environments. The cultivars Duganskij, Wenger Russian, Eureka Myrtis, and Leningrad were among the ones with the greatest bulb weight and bulb yield in all environments (Tables 2-5). In the two southern environments (*i.e.*, S19 and S20), the performances of these four cultivars were comparable to that of the most commonly grown cultivar Music as they presented an average of less than 5 % difference in yield and 15% in bulb weight when compared to this later cultivar. Not only the cultivars Duganskij, Wenger Russian and Eureka Myrtis have demonstrated a good stable performance across environments, but in addition, they distinguished themselves from the cultivar Music in that they also present a purple motif on their bulb wrappers, which could be more appealing for some consumers. Their utilization in combination with the cultivar Music, already used by most growers, could be an attractive option for diversification of the offering of locally produced garlic. Also, since they are not from the same type according to Engeland classification, they are more likely to taste different than the cultivar Music. Indeed, it was previously reported by Monta *et al.* (2011) that different ecophysiological group of cultivars can offer different sensory characteristics. Among garlic of the Rocambole type, the cultivar French Rocambole was one of the best performers along with the cultivar Mountain Top (Tables 2-5) with bulb weight at an average of 29.4 g/bulb in three environments out of four. However, they dropped to 15g/bulb in N19 for Mountain Top and in N20 for French Rocambole. Softneck garlic had a similar pattern where they had average performers in the southern environments (average of 30.3 g/bulb, 6.9 ton/ha) but poor performance in the northern environments (average of 14.8 g/bulb, 2.7 ton/ha). The cultivar Silver Rose had lower emergence than the other softneck cultivars (*i.e* Silver White and Italian). However, planting softneck could represent a good option if a producer uses their own cloves to plant and wants to rapidly expand its stock since it produced more cloves than the cultivar Music. It is especially true in the southern environment where the clove number and bulb weight were higher. The last advantage of softneck cultivars is their absence of flower stem that would reduce field management need, the flower typically having to be cut down upon flowering.

If some cultivars did not seem adapted to the northern environments of Quebec, others were not adapted to the four environments included in the present study. As a matter of fact, the cultivars Ajo Rojo and Creole Red had low emergence and weak plants that produced irregular bulbs in all four environments. Their average bulb weight represented only 27% of the average overall bulb weight.

3.4.2 Stability of traits across environments

Some traits varied less between environments and within environments than bulb yield. Clove color, clove number, bulbils color, shape and number were relatively stable across environment similarly with the finding of Volk and Stern (2009). The only environment that showed lower values of most quantitative traits compared to other environments was N20, probably due to the poor growing conditions in that environment (*i.e*., heavy drought at emergence, no fertilisation) that strongly affected overall growth of most cultivars. The minor changes observed in terms of clove and bulbils colors across sites are more linked to color intensity (*i.e*., dark pink to purple clove color) than major changes in opposition to the switch from colored to plain bulb wrapper color or pseudostem coloration often observed between environments. Those major changes in some cultivars have also been observed in the US for bulb wrappers (Volk and Stern, 2009) and in Greece for anthocyanin coloration of pseudostem, cloves and bulbs as well as foliage erectness and clove arrangement (Polyzos *et al.*, 2019) The description of a collection of garlic cultivars has to be based on traits that will have no or little variation between environments. Even if bulbils are not used by many growers to produce garlic, their stable characteristics make them the primary choice to differentiate garlic types along with clove color and number.

3.4.3 Classification

In all four environments, using all 28 phenotypic traits we monitored in the present study, cultivars overall clustered following a pattern which was very similar to the garlic types defined by Engeland (1991) (Fig. 3.7). Some notable exceptions across environments included the cultivar Metechi-2 from the Marble Purple Stripe type and the cultivar Portuguese form the Turban type which both generally clustered along with cultivars of Purple Stripe type. The cultivar Maiskij, the other cultivar of the Turban type, clustered with cultivars of the Marble Purple Stripe type. Therefore, garlic producers have to be careful before using the type of garlic as their main source of information on cultivars since some can be either wrongly classified or diverge from the typical traits of a type. Aa a result, using only the type of garlic instead of the cultivar's name to sell garlic on small market could lead to confusion for the consumers if a garlic cultivar does not present phenotypic characteristics of the type it is assigned to.

The phenotypic traits which discriminated cultivars in distinct clusters and garlic types included the presence of a hard stem, number of bulbils and their shape, number of cloves and their colors. However, the distance between clusters were modified by bulbil color, foliage rigidity, pseudostem coloration and bulb color. Other studies have found that the presence of a hard stem is a key trait contributing to the separation of cultivars in distinct clusters (Etoh and Simon, 2002). However, in our study, if cultivars that did not produce scape clustered together, it was not always the main divider of the classification.

3.4.4 Differentiation of cultivars

Despite obtaining garlic from reliable providers and keeping records of the identification, the purity of the bulb stock appeared to be compromised for two cultivars: Metechi and Music, as the two presented two phenotypes that clustered in different groups (Fig. 3.7). Generally, the cultivar Metechi produced an average of five purple cloves per bulb with few dark colored bulbils, while Metechi-2 produced more cloves of a pinkish color and more bulbils with a paler purple coloration. The cultivar Metechi presented characteristics which were common to the majority of Marble Purple Stripe type cultivars while Metechi-2 presented characteristics similar to the cultivar Portuguese and other cultivars of the Purple Stripe type. The cultivar Music like most Porcelain type cultivars had between three and five large whitish cloves with numerous whitish bulbils. In contrast, Music-2 produced eight to twelve dark colored cloves with few dark bulbils. Its traits presented similarity to the Porcelain type in terms of bulb size, to the Marble Purple Stripe in terms of appearances of bulbils and to the Purple Stripe type for the appearance of its clove, although the size and number of bulbils and cloves were more similar to the Rocambole type. It could be the reason why Music-2 clustered differently at S19 than S20 since small difference in frequency could have highlighted its similarity with a different type. An analysis on more individuals could result in a better understanding of its behaviour.

For a garlic producer having a stock of mixed cultivars could result in losses as they can reach maturity at different stages. Also, the lack of uniformity could come in conflict with retailers and consumers acceptance. Creation of a garlic index similar to the one used in France (Messiaen *et al.*, 1993) could be an alternative to avoid this issue. However, many cultivars used in this study had limited morphological differences both in terms yield components and qualitative traits. The morphological redundancy of garlic cultivars could be an indication that this set of cultivars may contain duplicates. In the US, Volk *et al.* (2004) have found duplicate for 41 % of the commercial garlic and 64% of the Western Regional Plant Introduction Station (WRPIS) collection while Barboza *et al.* (2020) found duplicates for 74% of the Argentine cultivars tested. It highlights the importance of using more than just morphological traits to describe garlic cultivars before being able to produce a garlic catalogue for producers, retailers and consumers. The use of genetic information such as genetic markers would be a good alternative method to investigate which cultivar is truly different from another since the phenotype had limited capacity to separate cultivars.

3.5 Conclusion

We observed that some garlic cultivars overall performed better than others but the yield and yield component were highly variable across the environments included in our study. The cultivars Duganskij, Wenger Russian, Leningrad and Music appeared to have potential for use across a range of environments in the Province of Quebec while the cultivars Ajo Rojo and Creole Red appear not to be adapted locally. In general, clove color, bulbils number, color and shape were among the most stable phenotypic traits across environments and thus could be used as indicators of stock purity.

3.6 Literature cited

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Connecting text for chapter 4

In chapter 3, garlic cultivars grown in contrasting environments of Quebec were comprehensively described and compared using phenotypic traits. Many cultivars within a type were found to be close morphologically, sometimes too close to find unique characteristics to differentiate them. This, together with the presence of distinct phenotypes for a uniquely named cultivar, highlights the necessity to find more accurate ways to differentiate garlic. Therefore, more investigation is needed to ascertain the identity of those cultivars. In chapter 4, the genetic identity of the 36 cultivars evaluated in chapter 3 was investigated and a comparison between classification obtained by studying morphological traits and genetic features was made.

4. Genetic diversity among garlic cultivars (*Allium sativum* L.) grown in Québec, Canada using microsatellite markers

Abstract

Garlic is propagated asexually, but still presents hundreds of lines or cultivars each exhibiting distinct valuable agronomical features. However, through repeated rounds of human manipulation the origin of several cultivars was lost as many got renamed. Genetic studies are thus essential to determine if cultivars are truly different from another and to better guide stock diversification efforts of Québec garlic growers. In this study, 45 samples coming from 37 cultivars named differently were analyzed using 21 SSR markers. A total of 81 different alleles were found for these SSR markers with a mean PIC value of 0.5. The set of samples was composed of 18 unique genotypes with no more than three genotypes per garlic type. The cultivars Metechi and Music respectively had two and three different genotypes, which concord with the morphological differences observed in Chapter 3. Overall, the genetic groups obtained with this analysis were similar to the garlic type described by Engeland (1991) and the morphological classification in Québec obtained in Chapter 3. This study could serve as a cornerstone for further genetic analyses of Québec grown garlic.

4.1 Introduction

Garlic originates from Central Asia and has been grown for consumption and medicinal purposes for thousands of years. The earliest recorded use of garlic was by the Egyptian civilization nearly 5,000 years ago (Etoh and Simon, 2002). Nowadays, commercial garlic is produced all around the world. In the process of domestication, garlic has lost its fertility, but multiple spontaneous mutations have allowed producers to select lines with traits of agronomical interests. As a result, hundreds of different cultivars with a wide range of morphological and genetic characteristics are now available to producers.

At a smaller scale, the heavy volume of seed exchanges between farmers and the garlic capacity to adapt to its environment has led to a multiplication of names for several cultivars. For example, farmers have renamed cultivars to fit best with new growing locations, consumer preferences or new features. Consequently, in the US, 64% of the Western Regional Plant Introduction Station (WRPIS) collection and 41% of the commercial U.S. garlic cultivars was found identical genetically (Volk *et al.*, 2004). In the Seed of Diversity Catalogue in Canada (2021), 200 garlic cultivars are named but many have similar names and others are already known as possible duplicate such as Persian Star and Duganskij (M-P Beaudoin, Agr, Personal Communication).

These name duplications have become so extensive that some cultivars have over five secondary names, which can often be sold as being part of different garlic types (Meredith, 2008). For example, Italian Purple is currently sold as a Rocambole type (https://www.groworganic.com), as a Purple Stripe type (https://garlicseed.ca/), and even as a Turban type (https://www.australiangarlic.net.au). Many traits such as clove number, clove color, presence of a hard stem, taste and conservation show extreme variations between providers for this cultivar.

The difficulty to access the real traits of garlic as led farmers to restrain their utilization of new cultivars. Diversification of their stock could result in disadvantage like growing unadapted cultivars or duplicate cultivars. Therefore, in Quebec, Music is mainly used since it is well adapted to this climate (Landry and Khanizadeh, 1994). However, to secure a bigger part of the current garlic market in Quebec, producers must work with more cultivars to increase availability and meet more consumer preferences in terms of taste, look and ease of use. To resolve this problem, in France, a certification program assesses that garlic seeds are coming from pure well-characterized disease-free lines (Messiaen *et al.*, 1993). This easiest access to quality seed stock allows producers to increase their production more rapidly and to promote their cultivars to retailers, chef and the transformation industry more easily. In Quebec, like most of the world, such a system does not exist. One of the main steps towards certifying garlic will be to determine if cultivars are different from another.

Since morphological characteristics tend to vary between environments due to the high phenotypic plasticity of garlic, differentiation of cultivars remains difficult. However, studies have shown a great level of genetic variability between cultivars making molecular markers a useful tool to characterized garlic lines. Recently, isoenzymes (Pooler and Simon, 1993; Lallemand *et al.*, 1994), Sequence Related Amplified Polymorphism (SRAP) (Chen, Zhou, *et al.*, 2013), Randomly Amplified Polymorphic DNA (RAPD) (Bradley *et al.*, 1996; Al-Zahim *et al.*, 1997), Amplified Fragment Length Polymorphism (AFLP) (Ipek *et al.*, 2003; Volk *et al.*, 2004; Wang *et al.*, 2016), Single Nucleotide Polymorphism (SNPs) (Egea *et al.*, 2017) and microsatellites (Jo *et al.*, 2012; Cunha *et al.*, 2014; Kumar *et al.*, 2019; Barboza *et al.*, 2020) were used for this purpose. Microsatellites or simple sequence repeats (SSR) are a precise, informative, fast, low-cost technique that is gaining popularity to differentiate population. One of the main uses of these codominant markers is to fingerprint cultivars (Vieira *et al.*, 2016).

Therefore, the aim of this chapter was to use microsatellite to differentiate garlic cultivars from one another and investigate their relationship. Specifically, the objectives of this chapter are to i) select a set of microsatellites to differentiate garlic cultivars of chapter 3, ii) evaluate the genetic variability in garlic cultivars grown in Quebec and iii) compare the genetic diversity of garlic cultivars mapped in this chapter to the morphological classification obtained in chapter 3.

4.2 Materials and methods

4.2.1 Plant material

Bulbs from thirty-seven cultivars were purchased from agricultural producers in Canada. Cultivars from different sources or having large phenotypic dissimilarities, but having the same name were labelled with an appended number (e.g., name-1, name-2) resulting in a total of 45 samples. Garlic bulbs that showed important divergence in phenotype were analyzed separately. Cultivars used in this study and their main morphological characteristic when grown in Sainte-Anne-de-Bellevue, Qc in 2020 (45.410789°, -73.93600°) are described in Table 4.1 with the addition of bulbs coming from the same stock but grown in Alma in 2019 (*i.e.* Metechi-4), in L'Ascension-de-Notre-Seigneur in 2019 (*i.e.* Metechi-5) and in Napierville in 2019 (*i.e.* Music-2) to better understand the origin of the phenotypically mixed cultivars. In addition, the cultivar Messidor, a French imported cultivar grown in Québec was used as a potential outlawyer.

4.2.2 SSR genotyping

Foliage leaf primordia of each cultivar were collected from two cloves for DNA analysis. DNA extraction was made using Qiagen DNeasy Plant extraction kit (Qiagen Inc., Toronto, ON, Canada). Amplification of DNA was made with an additional fluorescent primer similar to the technique developed by Schuelke (2000). Briefly, a M13

Tag labelled with a fluorescent dye (FAM) (5'-CCT TTG TCG ATA CTG GT-3') was designed and added to the PCR reaction. This additional primer will bind to the forward primer of each region making the amplicon fluorescent without designing specific fluorescent markers for each region. This technique is effective and reduce the cost of each amplification (Schuelke, 2000).

Therefore, PCR reactions were performed using a modified Qiagen HotStarTaq Plus protocol (Qiagen Inc., Toronto, ON, Canada) in a final volume of 20 ul and containing 15.8 µl of water, 2 µl 10×PCR buffer (contains 15mM $MgCl_2$), 0.4 µl dNTPs (200 uM each), 0.2 µl of each primer at 0.1 µM, 0.2 µl of M13 fluorescent primer at 0.1 µM, 0.2 µl DNA polymerase at 5 units/µl, and 1 µl of genomic DNA (10–20 ng). The PCR program had an initial denaturation of 5 min at 95°C, followed by 30 cycles of 95°C (30s), Ta (60s), 72°C (60s), followed by 4 cycles of 94°C (30s), 50°C (30s), 72°C (60s) and a final extension at 72°C for 12 min.

A total of 32 regions were selected in the literature to screen the cultivars set (Ma *et al.*, 2009; Cunha *et al.*, 2012; Ipek *et al.*, 2015; Barboza *et al.*, 2018). Each primer is presented in table X. Confirmation of amplification was obtained on a 1% agarose gel and fragment lengths were estimated by capillary electrophoresis on an ABI 3730xl Genetic Analyzer at Plateforme de séquençage et de génotypage des génomes of Centre Hospitalié Universitaire (CHU) de Québec - Université Laval using a GeneScan 600 LIZ dye Size Standard. Allele sizes were analyzed using Geneious microsatellite plugin (Geneious Prime, 2021.0.3).

4.2.3 Statistical analysis

Each locus was evaluated using allele frequencies, Polymorphic Information Content (PIC) (Botstein *et al.*, 1980) calculated using R *POLYSAT* packages v. 1.7-4 (Clark and Jasieniuk, 2011). PIC characterizes a marker depending on its ability to detect polymorphisms in a population with 0 having an identical allele frequency on all individuals and 1 having multiple alleles of similar frequencies. Overall, loci with a PIC value of more than 0.5 are really informative (Botstein *et al.*, 1980).

To evaluate the genetic distance between cultivars, Bruvo's distance was used. Bruvo's distance is based on a stepwise mutation model design to take into account the mutation in microsatellite region since it depends on the length of the repeat units of each region (Bruvo *et al.*, 2004). Distance between individuals were shown using a principal component analysis (PCA) analysis using R *POLYSAT* packages v. 1.7-4 (Clark and Jasieniuk, 2011), and a minimum spinning tree with 100 bootstraps using R *poppr* packages v. 2.9.0 (Kamvar *et al.*, 2014).

Table 4.1 : Main morphological characteristics of 45 garlic genotyped samples

Cultivar	Type	type	Clove arrangment	Bulbils no.	Bulbils shape	Bulbils colors	Clove no.	Clove Weight (g)	Clove color	Bulb color
Italian	Artichoke	SN	2-3	-	-	-	7-12	2-3.5	White	White
Tibétain Red	Asiatic	HN	1	20-30	Big drop	Pink fading to cream	6-7	2.6-3.8	Cream	Thin purple stripes
Tibetan Red		HN	1	20-30	Big drop	Purple/pink fading to cream	7-9	3-4.3	Cream	Thin purple stripes
Ajo Rojo	Creole	WB	1-2	-	-	-	2-6	1.7-2.4	Purple/red to pink cloves with purple/red stripes	White
Creole Red		WB	1-2	30-100	-	-	3-8	1.2-2.3	Purple/pink cloves with stripes	White
Rose de Lautrec		WB	1-2	65-100	Rice	White or light pink	8-10	2-3.8	White with small upper pink stripes	White
Messidor	French	SN	2	-	-	-	10-15	-	White	White
Duganskij	Marble Purple Stripe	HN	1	30-50	Drop	Dark purple	4-6	4.4-7.7	Pink/purple	Purple stripes and spots
Eureka Myrtis		HN	1	80-110	Drop	Dark purple tip fading to cream or pink	4-6	7.3-9.9	Light Pink to Pink	Purple stripes and spots
Kostyn's Red Russian		HN	1	45-75	Drop	Dark purple	4-6	7.4-13.3	Pink	Large purple stripes
Metechi-1		HN	1	30-80	Drop	Purple	4-6	6-8.5	Pink	Large purple stripes
Metechi-2		HN	1	30-80	Drop	Purple	4-6	6-8.5	Pink	Large purple stripes
Metechi-3		HN	1	90-120	Rice	Pale Purple	6-8	2.2-3.2	Pink	White
Metechi-4		HN	1	30-60	Drop	Dark Purple	3-5	4-6	Dark purple	Marble purple
Metechi-5		HN	2	100-200	Rice	Pale purple	10-12	2.2-3.2	Light pink with small pink stripes	Thin pale purple stripe
Northern Siberian		HN	1	45-100	Drop	Dark Purple	3-5	6-9	Pink with darker spots	Large purple stripes and spots
Siberian		HN	1	45-75	Drop	Purple	4-6	6.3-9.3	Pink	Large purple stripes and spots
Wenger's Russian		HN	1	45-80	Drop	Purple	5-6	7.3-11.3	Pale pink	Large purple stripes, often entierly purple

Table 4.1 (Continued) : Main morphological characteristics of 45 garlic genotyped samples

Cultivar	Type	type	Clove arrangment	Bulbils no.	Bulbils shape	Bulbils colors	Clove no.	Clove Weight (g)	Clove color	Bulb color
German White	Porcelain	HN	1	150-210	Rice	White, some with a light pink tip	3-4	8-12	Light cream background with pink spots	White
Gerogian Crystal		HN	1	170-250	Rice	Ligth pink tip, white body	2-4	7.8-11.9	Light cream with upper pink spots	White
Leningrad		HN	1	180-300	Rice	White and light pink	4-6	5-8.4	Light pink border going pale in the center	White
Music-1		HN	1	280-350	Rice	Ligth pink tip, white body	3-4	12-17	light pink	White
Music-2		HN	1	230-330	Rice	Ligth pink tip, white body	3-5	12-20	light pink	White
Music-3		HN	1	200-300	Rice	Ligth pink tip, white body	3-4	12-20	Pink going to light cream	White
Music-4		HN	2	30-40	Drop	Pink	8	3-4	Pink	White
Music-5		HN	2	30-40	Drop	Dark Purple	8-9	3-5	Purple with white stripe	White
Northern Quebec		HN	1	190-280	Rice	Ligth pink tip, white body	2-4	8.6-11	Pink with center band light cream	White
Susan Delafield's		HN	1	145-200	Rice	Ligth pink tip, white body	2-4	7.2-12.6	Marble of pink and light cream	White
Ukrainian Mavnir		HN	1	170-210	Rice	Ligth pink tip, white body	3-4	5-7.4	Marble of pink and light cream with pink stripes	White
Yugoslavian Porcelain		HN	1	175-215	Rice	Ligth pink tip, white body	2-4	6.2-13	Marble of pink and light cream	White
Chesnok Red	Purple Stripe	HN	1-2	115-160	Rice	Pale purple	10-12	1.5-2.1	Pink with small purple stripes	Thin light purple stripes
Persian Star		HN	1-2	95-160	Rice	Pale purple/pink	8-11	1.7-3.3	Pink-purple at the border going paler in the center, pink stripes	Thin light purple stripe
Baba Franchuk's	Rocambole	HN	1-2	30-40	Big drop	Pink tip fading to cream, pink stripes	7-9	2.6-3.9	Cream	Thin light purple stripes

Table 4.1 (Continued) : Main morphological characteristics of 45 garlic genotyped samples

Cultivar	Type	type	Clove arrangment	Bulbils no.	Bulbils shape	Bulbils colors	Clove no.	Clove Weight (g)	Clove color	Bulb color
French Rocambole	Rocambole	HN	1-2	20-40	Big drop	Pink tip fading to cream, pink stripes	6-8	3.3-4.5	Cream	Thin light purple stripe
German Red		HN	1-2	25-40	Big drop	Purple/pink	7-9	2.3-3.3	Cream	Thin light purple stripes
Italian Purple		HN	1-2	15-25	Corn	Pink with purple stripes	8-10	2-2.8	Cream	Thin light purple stripes
Killarney Red		HN	1-2	15-25	Corn	Purple/pink	8-9	2.7-3.6	Cream	Thin light purple stripes, pale spots
Marino		HN	1	10-30	Corn	Purple/pink	6-7	2.6-4.3	Cream	Thin light purple stripe, pale spots
Mountain Top		HN	1-2	25-40	Corn	Pink tip fading to cream	7-8	3-4.3	Cream	Thin light purple stripe, pale marble
Penn Wonder		HN	1	15-30	Corn	Purple	6-8	3.2-4.4	Cream	Thin light purple stripe, pale marble
Silver Rose	Silverskin	SN	2-4	-	-	-	13-17	1.3-2.3	White	White
Silver White		SN	2-3	-	-	-	13-16	1.8-2.7	White	White
Maiskij	Turban	HN	1	40-65	Drop	Purple	4-6	5-8.4	Pink-Purple with purple spots	Marble purple
Portuguese		HN	1-2	125-190	Rice	Pale purple	10-12	2.4-3.4	Pink marble Purple	Thin light purple stripe
Sweet Haven	Unclassified	HN	1	10-15	Corn	Purple	5-7	2.6-3.9	Cream	Thin light purple stripe

Table 4.2 Characteristics of 32 microsatellites used for screening 36 garlic grown in Québec

Code	Reference	Primer name	Motif	SRR type	Primer sequence (5'-3')	Ta (°C)	Expected size	Number of allele	Expected PIC
1	Barboza et al. 2018	AsESSR-107	(TCC)4acct cc(TCT)4	c	F: **GGAAACAGCTATGACC**AATTCCACTTAACCCCCAAATC	59	276	2	0.35
					R: CGCAGGTGAAGTGAGTAGGAG				
2	Ma et al. 2009	GB-ASM-035	(GCC)3, (TCC)3	c	F: **GGAAACAGCTATGACC**ATTGGACTGAATTCTGAATACCT	60	288–302	6	0.34
					R: GGGTGTGTGGTTCAAGGA				
3	Ipek et al. 2015	AS11065	(GA)12	p2	F: **GGAAACAGCTATGACC**AAACAGTCGAAAGCGTGGATTG	57	189-219	11	0.87
					R: TACGGCTTGCTACCAAAGAC				
4	Ipek et al. 2015	AS392	(AC)10	p2	F: **GGAAACAGCTATGACC**ATTTCAACAGCATCAGTTTGTAGA	57	294-302	4	0.71
					R: CCTTCACCATCAACCTACATTG				
6	Ipek et al. 2015	AS926	(TA)21	p2	F: **GGAAACAGCTATGACC**AGCCTTGCTTGTCTACAACAC	58	110-112	2	0.42
					R: CCCTTTTACTTATACACGACTTAC				
7	Cunha et al. 2012	Asa06	(TG) 5	p2	F: **GGAAACAGCTATGACC**AGGGGTGTTACATTCTCCCCT	57	192	1	.
					R: ACCGCCTGATTTTGCATTAG				
8	Cunha et al. 2012	Asa07	(TG) 7	p2	F: **GGAAACAGCTATGACC**ACTCGGAACCAACCAGCATA	58	229-235	3	0.398
					R: CCCAAACAAGGTAGGTCAGC				
9	Ma et al. 2009	GB-ASM-078	(GT)12	p2	F: **GGAAACAGCTATGACC**ATGTTCCAACCAGATTTAATGC	60	194–236	6	0.69
					R: AAGTGGCGGTTGTGTCTG				
10	Ipek et al. 2015	AS211	(CAG)7	p3	F: **GGAAACAGCTATGACC**AAGAACATGAACCGGGATAGA	57	146-164	4	0.7
					R: GAGGTTGCTGTTGCTGC				
11	Ipek et al. 2015	AS352	(CTT)6	p3	F: **GGAAACAGCTATGACC**AGAAATGATCACAGCCCATTAC	59	289-292	2	0.24
	Ipek et al. 2015	AS352	(CTT)6	p3	R: AGGAGATGGAGTAGATCTGGC				
12	Ipek et al. 2015	AS981	(AAG)7	p3	F: **GGAAACAGCTATGACC**AAACATGCCCACCAACAGTC	59	209-215	3	0.59
					R: GAGATTGGTTGCGCTTAGAT				
13	Barboza et al. 2018	AsESSR-012	(TTG)9	p3	F: **GGAAACAGCTATGACC**AGCCAATCAGTTGATTACGCTTT	60	297	3	0.41
					R: AGCGTAAAAATGCAAGAGTGC				
14	Barboza et al. 2018	AsESSR-027	(GCT)6	p3	F: **GGAAACAGCTATGACC**AGTGACACACCCCATACATGC	59	246	3	0.34
					R: GCTGGAGAAGATGTCAAGGAA				
15	Barboza et al. 2018	AsESSR-030	(AGC)5	p3	F: **GGAAACAGCTATGACC**AGCAGCAGTAGAAGAACCTGCT	59	273	3	0.34
					R: AACCTCTTTTGGTGCCTCCT				
16	Barboza et al. 2018	AsESSR-038	(AAG)5	p3	F: **GGAAACAGCTATGACC**AGGTTTGAGACCAGCGTTACCT	60	267	2	0.34
					R: CGCTTTGTTGGGTGATGTAGT				
17	Barboza et al. 2018	AsESSR-055	(TCT)6	p3	F: **GGAAACAGCTATGACC**AGAAGCCTTGATCAGAGAAGCA	59	385	2	.
					R: TGCATTGATCTCCTCTTCCAT				
18	Ma et al. 2009	GB-ASM-080	(CCG)5	p3	F: **GGAAACAGCTATGACC**AAATCTCCCTCCAAAGTCCC	60	171–174	2	0.19
					R: CCTGTATTTTGTGTAAAGCATCA				
19	Ipek et al. 2015	AS614	(AAAT)5	p4	F: **GGAAACAGCTATGACC**AAATTCAATGCGCTTCACAGC	59	194-198	2	0.24
					R: AGCAGGTGCAATCAAACTGG				
20	Ipek et al. 2015	AS6389	(AGCCTG) 5	p6	F: **GGAAACAGCTATGACC**AGGCAGAAAACACCGAGAATG	60	118-136	4	0.69
					R: GCTGCTCCCCTTATATCGTTC				
21	Barboza et al. 2018	AsESSR-091	(CAAATC) 4	p6	F: **GGAAACAGCTATGACC**AGTTCTCCGTTGCGTCAATC	59	294	3	0.61
					R: GAATTTGCATCTTTCCCCTTC				
22	Barboza et al. 2018	AsESSR-096	(TGGATC) 3	p6	F: **GGAAACAGCTATGACC**AATTGATTTCCGTCGCTTAGAC	59	421	1	.
					R: GCAATCTGGATCCTGTTCTCA				
30	Barboza et al. 2018	AsESSR-14	(AGG)8	p3	F: **GGAAACAGCTATGACC**ACCCCTTCGGTTGTTTTTCTT	60	289-298	4	0.35
					R: CTGGGTACGGTCGTTATTGG				
31	Barboza et al. 2018	AsESSR-78	(CTTT)3	p4	F: **GGAAACAGCTATGACC**ATCTGACAGACGACCTGGAGAT	60	269-284	3	0.3

Table 4.2 (Continued) Characteristics of 32 microsatellites used for screening 36 garlic cultivars grown in Quebec

Code	Reference	Primer name	Motif	SRR type	Primer sequence (5'-3')	Ta (°C)	Expected size	Number of allele	Expected PIC
					R: ATCACTGCCTCTCCACAAGAA				
32	Barboza et al. 2018	AsESSR-83	(CATCTC)5	p6	F: **GGAAACAGCTATGACCA**CCAAAGCTCCCATCTTCATC	59	207-225	4	0.41
					R: CGTCGGCTCTCTTATTTTGC				
33	Barboza et al. 2018	AsESSR-102	(TCC)5acctcctc(CTT)4	c	F: **GGAAACAGCTATGACCA**TTATCTCCATCGACCCTTTCC	60	273-282	4	0.35
					R: GAGGAGAGAGAGGAGATCATGC				
34	Barboza et al. 2018	AsESSR-103	(TCC)6(TTC)5	c	F: **GGAAACAGCTATGACCA**TTATCTCCGTCGACCCTTTC	59	270-290	5	0.38
					R: GAACGGAGGAGAGAGAGGAGA				
35	Kulh et al. 2004	ACM086	(TAG)5	p3	F: **GGAAACAGCTATGACCA**GCGGATTGGATCATCAGATT	57	158-180	5	0.41
					R: TTCTTGATTCCTCCGTTTGG				
36	Ipek et al. 2015	AS2655	(AGAAA)5	p5	F: **GGAAACAGCTATGACCA**AACTCAATGCATGACAGAAGG	57	242-262	5	0.77
					R: AGGAGGAGGAGAATGCTGAA				
37	Ipek et al. 2015	AS437	(AGA)8	p3	F: **GGAAACAGCTATGACCA**TCGTCTGGCGTTGCATTATC	60	332-341	4	0.66
					R: CGCTTGTAATCGTTGATGACG				
38	Ipek et al. 2015	AS739	(AGC)10	p3	F: **GGAAACAGCTATGACCA**AACAGGGATCTTTGCTTCAGC	59	201-227	7	0.76
					R: GATCTGTTGTGGTTGGATGTTC				
39	Ipek et al. 2015	AS987	(AAT)6	p3	F: **GGAAACAGCTATGACCA**GTACCAACTCTTTCCTAACGC	57	216-231	5	0.72
					R: TCCAATAGTTGTGATGACAGG				

4.3 Results

To analyze the genetic diversity present in garlic cultivars grown in Québec, a set of 32 SSR markers were selected. Of those, two were monomorphic (SSR19, SSR22), four did not produce amplicons on more than half of the cultivars (SSR1, SSR6, SSR7, SSR10) and four had unclear or unstable pic patterns (SSR3, SSR8, SSR9, SSR17, SSR35). The remaining 21 SSR markers produced constant amplicons on at least two replicates per cultivar. In total, two to nine alleles were found per locus with a mean of 3.9 for a total of 81 alleles (Table 4.3). Only four alleles had a frequency below 5% and were therefore considered as rare. The loci selected for the analysis had a mean PIC value of 0.5 (range of 0.04 to 0.71) with nine loci considered as highly informative (PIC>0.6). This indicates that the selected SSR markers can differentiate garlic cultivars used in this study.

Using a PCA analysis, cultivars were separated into five main groups (Fig. 4.1). First, the softneck cultivars of the Silverskin and Creole type clustered apart from most hardneck cultivars. Then the hardneck cultivars were separated into four groups, the first being mainly from the Rocambole type, the second composed of Porcelain and Purple Stripe types, the third group included Music-4 and Music-5, while the fourth group was composed of the Marble Purple Stripe type. The latter group was closer to the softneck group. Two cultivars (*i.e*. Italian and Messidor), each being the only representative of their types did not cluster with any other cultivars.

To provide more information on the link between the cultivars, a minimum spanning tree was constructed (Fig. 4.1). The analysis revealed a separation between seven groups. The groups obtained were similar to the ones identified with the PCA analysis apart from the cultivars Italian and Messidor, which clustered together. In addition, the cultivars of the Purple Stripe type were separated from those of the Porcelain type using this method. Separation between groups was clear with a mean distance of 0.255 between groups. This genetic separation fit the phenotypical types obtained previously (Fig. 3.7) for most cultivars.

The high distance between groups can be explained by the fact that most of the SSR markers used in this study separated our set of cultivars in similar ways. As a matter of fact, most of the loci used could only differentiate groups of cultivars and not individual cultivars (Table 4.5). Most of the cultivars of the Porcelain type were differentiated by SSR12, SSR14, SSR15, SSR16, SSR31 and SSR37 while the cultivars Music-4 and Music-5 could be separated using SSR4, SSR14, SSR15, SSR20, SSR21. SSR30 and SR34. Cultivars of the Marble Purple Stripe type, with the exception of Metechi-3 and Metechi-5, were differentiated using SSR4, SSR12, SSR14, SSR20, SSR32, SSR33, SSR34, SSR36, SSR37, SSR38 and SSR39. The rest of Marble Purple Stripe type (*i.e* Metechi-3, Metechi-5) clustered with Purple Stripe type because of SSR12, SSR14, SSR15 and SSR21. Rocambole and Asiatic type were separated from the other using SSR5, SSR13, SSR30, SSR33, SSR38 and SSR39. The Creole and Silverskin types were separated from the other types by loci: SSR4, SSR5, SSR15, SSR20, SSR32, SSR33, SSR34, SSR36, SSR38 and SSR39. The Silverskin type could only be differentiated from the Creole type using SSR16 and SSR37. Finally, the cultivars Italian and Messidor clustered together at most loci and could only be differentiated using SSR2, SSR4 and SSR14. This indicates that the SSR markers used in this study were capable of separating garlic types, but provided much less definition between the cultivars of a given type.

In addition, for most of the types of garlic studied, genetically identical cultivars were found (Fig. 4.1, Table 4.4). For example, only two different genotypes were found for six differently named cultivars in the Marble Purple Stripe type, while the eight cultivars of the Rocambole type were divided into two genotypes. In addition, the eight cultivars of the Porcelain type clustered into four genotypes while the three of the Creole type into only one genotype. The two cultivars of the Asiatic type were identical to most of those of the Rocambole type. Finally, the cultivar Portuguese had the same genotype as the outgroup composed of the cultivars of the Marble Purple Stripe type (*i.e.*, Metechi-3, Metechi-5). Overall, only 18 unique genotypes were obtained on the 45 analyzed with 13 only represented by one cultivar. In general, cultivars with the same genotypes were shown to be closed phenotypically with the exception of the cultivar Rose de Lautrec that has distinct characteristic to the rest of the Creole type (Table 4.1)

Cultivars with samples coming from different sources (*i.e.*, Music and Metechi) presented more than one genotype. The cultivar Metechi clustered into two different and highly distant genotypes (0.379), one composed of Metechi-1, Metechi-2 and Metechi-4 and the second composed of Metechi-3 and Metechi-5. The cultivar Music clustered into three genotypes, one being Music-1, Music-2 and Music-3, while the two others were Music-4 and Music-5 respectively with distances averaging 0.287. Music-4 and Music-5 clustered closely together with a distance of 0.059. Because of its high genetic distance between genotypes, it is possible that cultivars of different origin have been mixed. In opposition, the two Asiatic cultivars, that were closely named (*i.e*. Tibetan Red and Tibétain Red) had identical genotype and were probably cultivars renamed to appeal to French markets.

Table 4.3 Main characteristics of the 21 loci used to analyze 37 garlic cultivars grown in Québec, Canada

Locus	Allele No.	Allele Size (bp)	Allele frequency	Overall PIC
SSR2	2	287	0.27	0.34
		296	0.73	
SSR4	3	309	0.35	0.57
		313	0.52	
		317	0.13	
SSR5	9	185	0.21	0.71
		189	0.01	
		191	0.13	
		203	0.18	
		205	0.02	
		207	0.13	
		213	0.01	
		221	0.30	
		223	0.01	
SSR11	2	320	0.10	0.06
		323	0.90	
SSR12	3	265	0.14	0.55
		268	0.57	
		271	0.29	
SSR13	3	244	0.75	0.41
		307	0.14	
		316	0.11	
SSR14	3	253	0.64	0.49
		256	0.24	
		259	0.11	
SSR15	5	290	0.04	0.68
		293	0.20	
		296	0.47	
		299	0.09	
		302	0.20	
SSR16	3	280	0.05	0.29
		283	0.20	
		286	0.76	
SSR18	2	170	0.09	0.04
		173	0.91	
SSR20	4	124	0.07	0.51
		130	0.24	
		136	0.60	
		142	0.09	
SSR21	3	309	0.33	0.5
		315	0.34	
		321	0.33	
SSR30	4	302	0.19	0.64
		305	0.23	
		308	0.31	
		311	0.27	

Table 4.3 (Continued) : Main characteristics of the 21 loci used to analyze 37 garlic cultivars grown in Québec, Canada

Locus	Allele No.	Allele Size (bp)	Allele frequency	Overall PIC
SSR31	2	284	0.15	0.22
		296	0.85	
SSR32	4	222	0.11	0.63
		225	0.30	
		228	0.53	
		240	0.06	
SSR33	5	289	0.33	0.71
		295	0.09	
		298	0.15	
		301	0.29	
		304	0.14	
SSR34	5	294	0.34	0.71
		300	0.09	
		303	0.17	
		306	0.26	
		309	0.14	
SSR36	4	255	0.09	0.54
		265	0.26	
		270	0.27	
		275	0.38	
SSR37	4	349	0.24	0.65
		352	0.24	
		355	0.11	
		358	0.41	
SSR38	6	215	0.38	0.67
		218	0.10	
		221	0.18	
		224	0.18	
		227	0.06	
		242	0.09	
SSR39	5	230	0.06	0.70
		236	0.34	
		239	0.24	
		242	0.27	
		245	0.09	
Mean	3.9	-	0.26	0.51

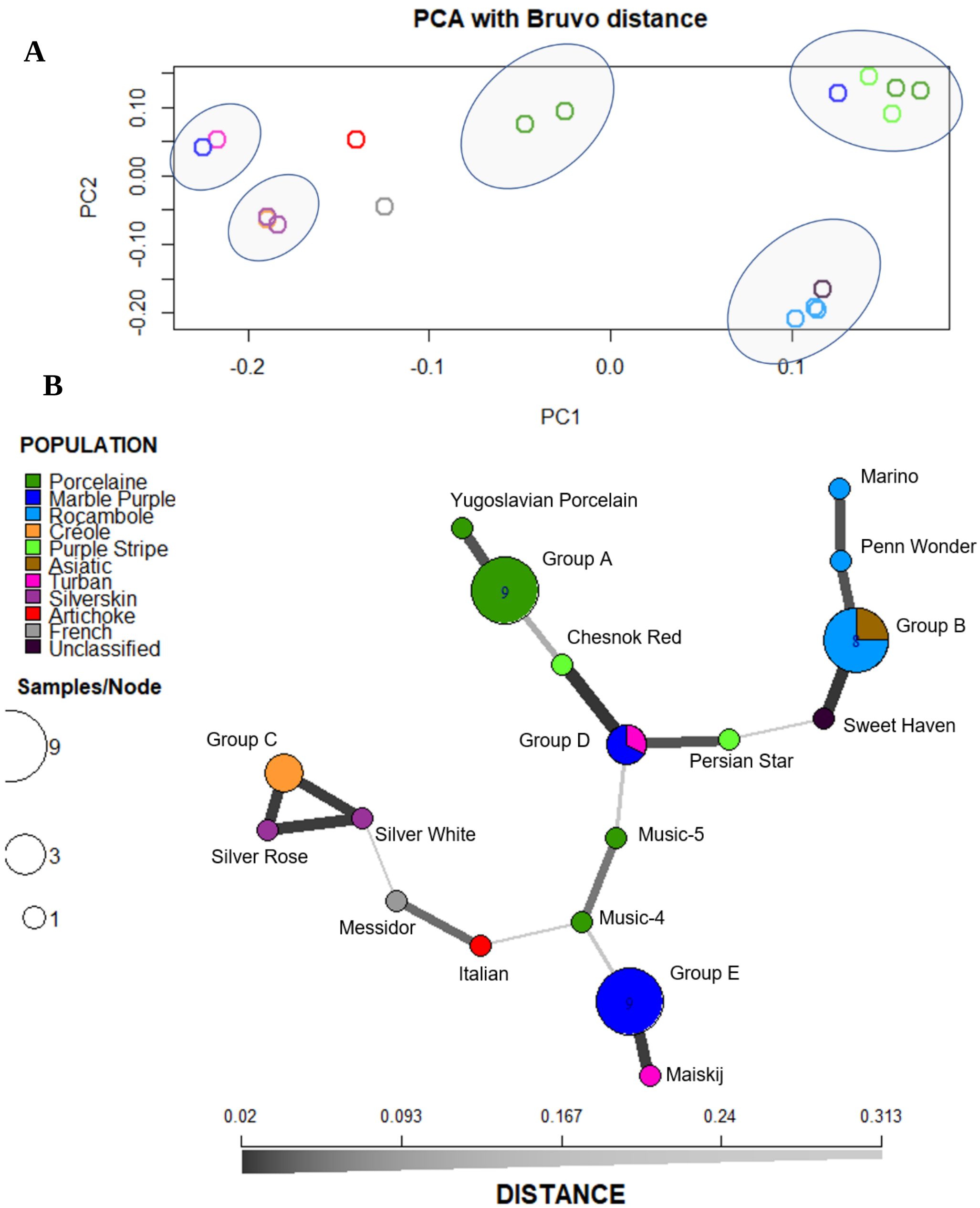

Fig. 4.1 Genetic relationship between 45 samples representing 37 garlic cultivars grown in Québec. A) PCA analysis using Bruvo's distance B) Minimum Spanning network (MSN) using Bruvo's Distance. Node colors indicate cultivar types. The thicker and darker the link between nodes is, the less important is the genetic distance between nodes. Nodes including more than one cultivar are labelled with a letter and described in table 4.4.

Table 4.4 Description of the group identify in the minimum spanning network (Fig. 4.1) composed of genetically identical garlic cultivars on 21 microsatellite regions.

Group A	Group B	Group C	Group D	Group E
Georgian Crystal	Baba Franchnuk	Ajo Rojo	Metechi-3	Duganskij
German White	French Rocambole	Creole Red	Metechi-5	Eureka Myrtis
Leningrad	German Red	Rose de Lautrec	Portuguese	Kostyn Red Russian
Music-1	Italian Purple			Metechi-1
Music-2	Killarney Red			Metechi-2
Music-3	Mountain Top			Metechi-4
Northern Quebec	Tibetan Red			Northern Siberian
Susan Delafield	Tibétain Red			Siberian Red
Ukrainian Mavnir				Wenger Russian

Table 4.5 Allele size obtained for 45 samples representing 37 garlic cultivars grown in Québec at 21 different loci

Type	Cultivar	SSR 2	SSR 4	SSR 5	SSR 11	SSR 12	SSR 13	SSR 14	SSR 15	SSR 16	SSR 18	SSR 20	SSR 21	SSR 30	SSR 31	SSR 32	SSR 33	SSR 34	SSR 36	SSR 37	SSR 38	SSR 39
Artichoke	Italian	287	309	185	320	268	244	253	296	283	170	130	315	302	284	225	289	294	270	349	218	236
		296	313	203	323	268	244	259	299	286	173	136	321	302	296	228	289	294	270	358	242	236
Asiatic	Tibétain Red	296	313	191	323	268	244	253	296	286	173	136	309	311	296	225	301	306	265	352	215	242
		296	313	207	323	271	307	253	302	286	173	136	315	311	296	228	304	309	275	358	215	242
	Tibetan Red	296	313	191	323	268	244	253	296	286	173	136	309	311	296	225	301	306	265	352	215	242
		296	313	207	323	271	307	253	302	286	173	136	315	311	296	228	304	309	275	358	215	242
Creole	Ajo Rojo	296	309	203	323	268	244	253	296	280	173	130	309	308	296	228	289	294	255	349	221	236
		296	309	221	323	268	244	253	296	286	173	142	321	308	296	228	295	300	270	355	221	245
	Creole Red	296	309	203	323	268	244	253	296	280	173	130	309	308	296	228	289	294	255	349	221	236
		296	309	221	323	268	244	253	296	286	173	142	321	308	296	228	295	300	270	355	221	245
	Rose de Lautrec	296	309	203	323	268	244	253	296	280	173	130	309	308	296	228	289	294	255	349	221	236
		296	309	221	323	268	244	253	296	286	173	142	321	308	296	228	295	300	270	355	221	245
French	Messidor	296	313	185	320	268	244	253	296	283	170	130	315	302	284	225	289	294	270	349	218	236
		296	313	203	323	268	244	256	299	286	173	136	321	302	296	228	289	294	270	358	242	236
Marble Purple Stripe	Duganskij	287	317	185	323	265	244	253	296	286	173	124	309	308	296	225	289	294	275	349	224	230
		296	317	221	323	268	244	259	302	286	173	130	315	308	296	240	298	303	275	358	227	236
	Eureka Myrtis	287	317	185	323	265	244	253	296	286	173	124	309	308	296	225	289	294	275	349	224	230
		296	317	221	323	268	244	259	302	286	173	130	315	308	296	240	298	303	275	358	227	236
	Kostyn's Red Russian	287	317	185	323	265	244	253	296	286	173	124	309	308	296	225	289	294	275	349	224	230
		296	317	221	323	268	244	259	302	286	173	130	315	308	296	240	298	303	275	358	227	236
	Metechi-1	287	317	185	323	265	244	253	296	286	173	124	309	308	296	225	289	294	275	349	224	230
		296	317	221	323	268	244	259	302	286	173	130	315	308	296	240	298	303	275	358	227	236
	Metechi-2	287	317	185	323	265	244	253	296	286	173	124	309	308	296	225	289	294	275	349	224	230
		296	317	221	323	268	244	259	302	286	173	130	315	308	296	240	298	303	275	358	227	236
	Metechi-3	287	309	185	323	265	244	256	293	286	173	136	321	305	296	222	298	303	265	352	215	239
		296	313	221	323	271	316	256	293	286	173	136	321	305	296	228	301	306	275	358	224	239
	Metechi-4	287	317	185	323	265	244	253	296	286	173	124	309	308	296	225	289	294	275	349	224	230
		296	317	221	323	268	244	259	302	286	173	130	315	308	296	240	298	303	275	358	227	236
	Metechi-5	287	309	185	323	265	244	256	293	286	173	136	321	305	296	222	298	303	265	352	215	239
		296	313	221	323	271	316	256	293	286	173	136	321	305	296	228	301	306	275	358	224	239
	Northern Siberian	287	317	185	323	265	244	253	296	286	173	124	309	308	296	225	289	294	275	349	224	230
		296	317	221	323	268	244	259	302	286	173	130	315	308	296	240	298	303	275	358	227	236
	Siberian	287	317	185	323	265	244	253	296	286	173	124	309	308	296	225	289	294	275	349	224	230
		296	317	221	323	268	244	259	302	286	173	130	315	308	296	240	298	303	275	358	227	236
	Wenger's Russian	287	317	185	323	265	244	253	296	286	173	124	309	308	296	225	289	294	275	349	224	230
		296	317	221	323	268	244	259	302	286	173	130	315	308	296	240	298	303	275	358	227	236
Porcelain	German White	287	309	221	323	271	244	253	290	283	173	136	309	305	284	222	298	303	265	355	215	239
		296	313	221	323	271	316	256	293	286	173	136	315	305	296	228	301	306	275	358	224	239
	Gerogian Crystal	287	309	221	323	271	244	253	290	283	173	136	309	305	284	222	298	303	265	355	215	239
		296	313	221	323	271	316	256	293	286	173	136	315	305	296	228	301	306	275	358	224	239

Table 4.5 (Continued): Allele size obtained for 45 samples representing 37 garlic cultivars grown in Québec at 21 different loci

Type	Cultivar	SSR 2	SSR 4	SSR 5	SSR 11	SSR 12	SSR 13	SSR 14	SSR 15	SSR 16	SSR 18	SSR 20	SSR 21	SSR 30	SSR 31	SSR 32	SSR 33	SSR 34	SSR 36	SSR 37	SSR 38	SSR 39
Porcelain	Leningrad	287	309	221	323	271	244	253	290	283	173	136	309	305	284	222	298	303	265	355	215	239
		296	313	221	323	271	316	256	293	286	173	136	315	305	296	228	301	306	275	358	224	239
	Music-1	287	309	221	323	271	244	253	290	283	173	136	309	305	284	222	298	303	265	355	215	239
		296	313	221	323	271	316	256	293	286	173	136	315	305	296	228	301	306	275	358	224	239
	Music-2	287	309	221	323	271	244	253	290	283	173	136	309	305	284	222	298	303	265	355	215	239
		296	313	221	323	271	316	256	293	286	173	136	315	305	296	228	301	306	275	358	224	239
	Music-3	287	309	221	323	271	244	253	290	283	173	136	309	305	284	222	298	303	265	355	215	239
		296	313	221	323	271	316	256	293	286	173	136	315	305	296	228	301	306	275	358	224	239
	Music-4	287	309	185	323	268	244	256	293	286	173	124	315	302	296	225	301	294	265	358	215	236
		296	317	221	323	268	244	259	302	286	173	136	315	308	296	228	301	306	275	358	224	236
	Music-5	287	309	185	323	265	244	256	293	286	173	124	315	302	296	225	301	294	265	355	215	239
		296	317	221	323	268	244	259	302	286	173	136	315	308	296	225	301	306	275	358	224	239
	Northern Quebec	287	309	221	323	271	244	253	290	283	173	136	309	305	284	222	298	303	265	355	215	239
		296	313	221	323	271	316	256	293	286	173	136	315	305	296	228	301	306	275	358	224	239
	Susan Delafield's	287	309	221	323	271	244	253	290	283	173	136	309	305	284	222	298	303	265	355	215	239
		296	313	221	323	271	316	256	293	286	173	136	315	305	296	228	301	306	275	358	224	239
	Ukrainian Mavnir	287	309	221	323	271	244	253	290	283	173	136	309	305	284	222	298	303	265	355	215	239
		296	313	221	323	271	316	256	293	286	173	136	315	305	296	228	301	306	275	358	224	239
	Yugoslavian Porcelain	287	309	223	323	271	244	253	290	283	173	136	309	305	284	222	298	303	265	355	215	239
		296	313	223	323	271	316	256	293	286	173	136	315	305	296	228	301	306	275	358	224	239
Purple Stripe	Chesnok Red	287	309	185	323	265	244	256	293	286	173	136	321	305	284	222	298	303	265	352	215	239
		296	313	221	323	271	316	256	293	286	173	136	321	305	296	228	301	306	275	358	224	239
	Persian Star	287	309	185	323	265	244	256	293	286	173	136	321	305	296	222	301	303	265	352	215	239
		296	313	205	323	271	316	256	293	286	173	136	321	305	296	228	301	306	275	358	224	239
Rocambole	Baba Franchuk's	296	313	191	323	268	244	253	296	286	173	136	309	311	296	225	301	306	265	352	215	242
		296	313	207	323	271	307	253	302	286	173	136	315	311	296	228	304	309	275	358	215	242
	French Rocambole	296	313	191	323	268	244	253	296	286	173	136	309	311	296	225	301	306	265	352	215	242
		296	313	207	323	271	307	253	302	286	173	136	315	311	296	228	304	309	275	358	215	242
	German Red	296	313	191	323	268	244	253	296	286	173	136	309	311	296	225	301	306	265	352	215	242
		296	313	207	323	271	307	253	302	286	173	136	315	311	296	228	304	309	275	358	215	242
	Italian Purple	296	313	191	323	268	244	253	296	286	173	136	309	311	296	225	301	306	265	352	215	242
		296	313	207	323	271	307	253	302	286	173	136	315	311	296	228	304	309	275	358	215	242
	Killarney Red	296	313	191	323	268	244	253	296	286	173	136	309	311	296	225	301	306	265	352	215	242
		296	313	207	323	271	307	253	302	286	173	136	315	311	296	228	304	309	275	358	215	242
	Marino	296	313	189	320	268	244	253	296	286	173	136	309	311	296	225	301	306	265	352	218	242
		296	313	213	323	271	307	253	302	286	173	136	315	311	296	228	304	309	275	358	218	242
	Mountain Top	296	313	191	323	268	244	253	296	286	173	136	309	311	296	225	301	306	265	352	215	242
		296	313	207	323	271	307	253	302	286	173	136	315	311	296	228	304	309	275	358	215	242
	Penn Wonder	296	313	189	323	268	244	253	296	286	173	136	309	311	296	225	301	306	265	352	215	242
		296	313	213	323	271	307	253	302	286	173	136	315	311	296	228	304	309	275	358	215	242

Table 4.5 (Continued): Allele size obtained for 45 samples representing 37 garlic cultivars grown in Québec at 21 different loci

Type	Cultivar	SSR 2	SSR 4	SSR 5	SSR 11	SSR 12	SSR 13	SSR 14	SSR 15	SSR 16	SSR 18	SSR 20	SSR 21	SSR 30	SSR 31	SSR 32	SSR 33	SSR 34	SSR 36	SSR 37	SSR 38	SSR 39
Silverskin	Silver Rose	296	309	203	323	268	244	253	296	283	173	130	309	308	296	228	289	294	255	349	221	236
		296	309	221	323	268	244	253	296	283	173	142	321	308	296	228	295	300	270	355	221	245
	Silver White	296	309	203	323	268	244	253	296	283	173	130	309	308	296	228	289	294	255	349	221	236
		296	309	221	323	268	244	253	296	286	173	142	321	308	296	228	295	300	270	352	221	245
Turban	Maiskij	287	317	221	323	265	244	253	296	286	173	124	309	308	296	225	289	294	275	349	224	230
		296	317	221	323	268	244	259	302	286	173	130	315	308	296	240	298	303	275	358	227	236
	Portuguese	287	309	185	323	265	244	256	293	286	173	136	321	305	296	222	298	303	265	352	215	239
		296	313	221	323	271	316	256	293	286	173	136	321	305	296	228	301	306	275	358	224	239
Unclassified	Sweet Haven	287	313	191	323	268	244	253	296	286	173	136	309	311	296	225	301	306	265	352	215	242
		296	313	207	323	271	307	253	302	286	173	136	315	311	296	228	304	309	275	358	215	242

4.4 Discussion:

Over centuries of domestication, hundreds of different garlic lines have been isolated. Since garlic is propagated asexually most of the selected traits are due to spontaneous mutation or phenotypic plasticity. However, the distinctive characteristics of individual lines have not been clearly recorded and cultivar names have been modified resulting in a high level of redundancy of garlic seed in collections all over the world (Volk *et al.*, 2004; Cunha *et al.*, 2014). Since garlic morphological traits vary between environments, genetic is the only trustable way to assess the unicity of the cultivars used. In recent years, microsatellites have been one of the main technique used in the field of genetic markers used for cultivars fingerprinting (Vieira *et al.*, 2016). As a result, even without the garlic genome being sequenced, over 300 SSR markers have been tested on garlic cultivars in the literature (Ma *et al.*, 2009; Cunha *et al.*, 2012; Ipek *et al.*, 2015; Liu *et al.*, 2015; Barboza *et al.*, 2018).

Since only a few of those markers have been tested on North American garlic cultivars, the set used in this study comprised markers previously characterized in multiple studies (Ma *et al.*, 2009; Cunha *et al.*, 2014; Ipek *et al.*, 2015; Barboza *et al.*, 2018). The PIC values of the SSR markers used were similar or higher when compared to the values obtained by Ma *et al.* (2009) and Barboza *et al.* (2018) with their set of cutivars. The higher PIC values obtained in our study is probably due to a better spread in garlic types than the one of Barboza *et al.* (2018, 2020), a study that included proportionally more softneck cultivars than our set. Softneck cultivars are known to be relatively less diverse than hardneck cultivars (Volk *et al.*, 2004). In contrast, the SSR markers of the study of Ipek *et al.* (2015) obtained lower PIC values in our study. The lower polymorphic content observed in our set could come from the fact that the set of cultivars used by Ipek *et al.* (2015) was selected to contain maximum genetic diversity and minimal redundancy as some cultivars of their set had already been characterized with other types of genetic markers (Ipek *et al.*, 2003). Overall, our set of SSR markers offered an average PIC value of 0.5, which represent a good discrimination power (Botstein *et al.*, 1980).

Even with this informative set of SSR markers, 61% of the cultivar shared their genotype with at least one other cultivar. Finding duplicates in a garlic set is common as the habit of renaming cultivars is well spread creating genetic replicates (Volk *et al.*, 2004; Cunha *et al.*, 2014). The number of duplicates obtained in this study is lower than the one (72%) obtained from Argentinian cultivars also using SSR markers (Barboza *et al.*, 2020) but higher than other studies which have shown between 39 and 50% of duplicates (Ma *et al.*, 2009; Cunha *et al.*, 2014; Ipek *et al.*, 2015). Other types of markers have revealed similar proportions of duplicates. Two collections of garlic cultivars in the USA showed 41% and 68% of redundancy using AFLP (Volk *et al.* 2004) while Ipek *et al.* found 33% of duplicates in various cultivars with the same technique.

At the end, the main difference found in our set of cultivars can be attributed to the garlic types and not the cultivars in themselves, as most cultivars within a type were found to be genetically identical. Although, no individual SSR marker was able to discriminate all types by itself. SSR12 was the one marker closer to achieving such differentiation, but it did not cluster softneck cultivars apart from Creole type. Also, contrary to what was reported by Barboza *et al.* (2020), SSR15 did not show a unique allele for all softneck cultivars. In this study, this unique allele was shared by the cultivar Italian of the Artichoke type and the cultivar Messidor, a French garlic variety, but not by cultivars of the Silverskin type. No SSR marker used in this study were capable of separating the softneck from the hardneck cultivar as the Silverskin type nearly always clustered with the Creole type composed of weakly bolting cultivars. Overall, because the selected SSR markers differentiated cultivars in similar groups, the same classification could be obtained using only 7 SSR markers (*i.e*. SSR2, SSR5, SSR14, SSR16, SSR20, SSR31 and SSR37) instead of 21

The bolting capacity is one of the main classification factors used for garlic both phenotypically (Etoh and Simon, 2002; Meredith, 2008) and genotypically (Al-Zahim *et al.*, 1997; Barboza *et al.*, 2020). However, similarly to the finding of Chen *et al.* (2014), weakly bolting cultivars (*i.e*. Creole type) did not cluster apart from softneck cultivars. Also, in opposition to Engeland classification, but in concordance with the finding of Volk *et al.* (2004), cultivars in the Asiatic type were not softneck cultivars as they always showed strong bolting and clustered genetically with hardneck cultivars, in this case those of the Rocambole type. However, only a few cultivars of Asiatic and Creole types were used in this study, which might reduce its classification power.

By comparing the genetic classification (Fig. 4.1) and the phenotypic classification obtained in Chapter 3 (Fig 3.7), similarities were found. In both classifications, cultivars clustered close to cultivars of the same type as described by Engeland (1991). The similarity between cultivars of the Asiatic and the Rocambole types was confirmed as they were found to be genetically identical. The cultivar Sweet Haven, an unclassified cultivar, also clustered genetically and phenotypically with the Rocambole type. Its identity as a different cultivar was confirmed since it had an average distance of 0,050 from the Rocambole genotypes. Finally, the cultivars of the Turban type did not cluster together in both classifications. Even with the morphological variation observed between and within sites some characteristics were robust enough to fit to the major genetic differences.

Some differences between classifications were also found. Phenotypically the Silverskin type clustered closer to the cultivar Italian, but genotypically cultivars of this type were closer to the Creole type. Also, the cultivar Rose de Lautrec had a different morphological classification depending on the environments it was grown in, but was found to be genetically identical to the rest of the Creole type. It is possible that not enough microsatellites were used to differentiate this cultivar since other studies have used up to 130 alleles to differentiate their cultivar set (Ipek *et al.*, 2015). Also, the phenotypic divergence observed in the Creole type could be the result of the poor performance of the cultivars Ajo Rojo and Creole Red. Those cultivars did not develop full-size plants like the other cultivars. The trait obtained can be affected by an incomplete growth or stress-related factors leading to dissimilarities with the cultivar Rose de Lautrec. In addition, some morphological differences that seemed important and often stable through years, like yield, did not translate into genetic difference using SSR markers in other studies (Cunha *et al.*, 2014). Epigenetic changes can happen in garlic when grown in a specific environment for a long time, thus creating small change in its morphology (Gimenez and Lampasona, 2018). This could be the reason why the cultivar Music, the most commonly grown cultivar in Québec, has higher yield than other cultivars sharing its genotype.

The stock of the cultivars Metechi and Music used in the chapter 3 were composed of more than one cultivar as divergence occurred both genetically and phenotypically. For the cultivar Metechi, the main phenotype (*i.e*. genotype Metechi-1, Metechi-2, Metechi-4) had the same genotype as the rest of Marble Purple Stripe type while the secondary phenotype (*i.e* genotype Metechi-3, Metechi-5) was identical to the cultivar Portuguese of the Turban type. Phenotypically, the secondary phenotype of the cultivar Metechi is similar to the one of the Portuguese cultivar, but with generally paler cloves color. The cultivar Music diverged into three genotypes. The first one (*i.e*.. sample Music-1, Music-2, Music-3) had a similar phenotype than the rest of the Porcelain type cultivars and was found to be identical genetically to them. However, Music-4 and Music-5 were different genetically and phenotypically from every cultivar and type analyzed in this study. Since they clustered closer genetically to Marble Purple Stripe and Purple Stripe types, and had morphological traits close to those groups (*i.e*., clove and bulbils appearances), it could be possible that they can be part of the Glazed Purple Stripe type. In its first attempt of classification, Engeland put cultivars of these three groups together. It is only later, after genetics analysis, that they have been separated (Meredith, 2008). In this study no representative of this type were analyzed.

With the recent release of the garlic genome (Sun *et al.*, 2020), more advanced genetic methods could be used more easily to assess the differences between garlic cultivars. One of them is genotyping-by-sequencing, it is a low genome coverage sequencing technique that is useful for single nucleotide polymorphism (SNP) discovery in large and complex genomes. Egea *et al.* (2017) have use this technique to find 14,982 SNPs on 417 accessions before having a reference genome. Even with this extensive work, 30% of their samples were genetically redundant. Also, with the decrease in cost whole genome sequencing could be used to better understand the specific mutation that created the different lines. With more information on both the genetic and epigenetic of garlic, a selection of cultivars more adapted to Quebec climate could be done. For growers, it would be the opportunity to select cultivars with known different characteristics that could diversify their production and thus access a bigger place on the local market.

4.5 Conclusion

In conclusion, the set of microsatellites used was able to differentiate the garlic types but not the cultivars. This absence of differentiation could be caused by duplicates as genetically identical cultivars were generally close phenotypically. However, further genetic analyzes are needed to know if garlic cultivars grown in Québec are as diverse as their names.

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5. General conclusion

Garlic is a crop of which the popularity is increasing in Québec. However, the lack of knowledge about the genotypes and phenotypes of garlic cultivars grown in this province hinders the sustainable development of this industry. As such, the first part of this study tried to determine the potential of 36 garlic cultivars for production in Québec and their main traits. While yield and yield-related traits have varied between environments, cultivars from the Porcelain and the Marbled Purple Stripe types performed similarly to the most popular cultivar grown in Québec, Music. On the other hand, the cultivars Ajo Rojo and Creole Red performed poorly and therefore cannot be recommended for Québec production. In general, the performance of cultivars was higher in the southern environment than the northern ones. The traits linked to the bulbils as well as clove color and number seemed to vary less between environments. However, the cultivars within a type showed small differences in regard to the traits analyzed in this study. As a result, the description obtained for each cultivar were generic, but could still be the base for a classification system.

The second part of this project was to analyze genetically the 36 cultivars using 35 microsatellites markers in order to confirm their true identity. 61% of the cultivars analyzed were found to be genetically identical using 21 SSR markers. In general, cultivars of a given type were genetically identical. However, despite having the same names, two and three distinct genotypes were respectively found for the Metechi and Music cultivars. In addition, those distinct genotypes displayed divergent phenotypic traits. With the exception of these, the other cultivars analyzed clustered similarly both phenotypically and genotypically in accordance with the Engeland classification.

Finally, the results obtained partially confirmed our hypotheses. As a matter of fact, we could not find a unique set of morphological or genetic traits to differentiate all the cultivars grow in this study. Similarities between cultivars of a given type were too high to truly separate them morphologically. However, in accordance with our second hypothesis, most cultivars clustered close to Engeland (1991) types both genetically and phenotypically, but many were genetically identical.

5.1 Future directions

Those findings have highlighted the importance of the characterisation both phenotypically and genotypically of garlic cultivars to ensure that growers can have access to pure-line stocks. Further genetic analyzes are needed to ensure the identity of the cultivars grown in Québec, which will become easier since the garlic genome is now available. With stocks of pure, well-described cultivars, growers will likely increase their production and meet retailer and consumer demands for a high-quality product. Moreover, studies link to the taste and the pathogen resistance are also needed for those pure garlic line to offer an even better classification/certification system. This work and future research will help the creation of a garlic cultivar catalogue for growers to ensure the sustainability of the garlic production sector of the province of Québec.

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Suppl. Table 1: Plant characteristics of 36 garlic cultivars grown at Sainte-Anne-de-Bellevue in 2019 (S19). Means in a given column followed by different letters are significantly different ($LSD_{0.05}$).

	Leaves Number		Plant Height (cm)		Stem Diameter (mm)		Flower turns		Flowering stem height (cm)		Spathe Length (cm)	
Cultivar	Mean	sd	Mean	sd	Mean	sd	Mean	sd	Mean	sd	Mean	sd
Artichoke												
Italian	9.0	± 0.7 a	25.6	± 2.8 hi	11.9	± 1.8 c-e	9.0	± 12.0 a	-	- -	-	- -
Asiatic												
Tibetan red	8.1	± 1.2 c	27.5	± 3.6 f-h	10.9	± 1.7 d-f	1.6	± 0.6 ef	83.6	± 10.2 e-g	11.5	± 2.4 g
Tibétain red	7.2	± 0.8 ef	28.9	± 3.3 f	10.7	± 2.1 ef	1.6	± 0.5 e-g	88.6	± 8 d-f	12.2	± 2.8 g
Creole												
Creole Red	5.1	± 0.6 j	23.2	± 9.2 i	7.2	± 3.2 g	7.3	± 3.4 b	64.0	± 0.1 h	13.0	± 0 fg
Rose de Lautrec	6.6	± 0.6 f-h	29.1	± 4 f	13.3	± 2.7 bc	1.1	± 1.5 g-i	80.0	± 16.2 f-h	10.6	± 1.9 g
Porcelain												
Duganskij	7.2	± 0.6 ef	30.2	± 3.1 ef	15.2	± 1.8 a	1.1	± 0.4 g-i	80.3	± 8.8 f-h	15.1	± 2.6 ef
Eureka Myrtis	6.1	± 0.8 hi	28.6	± 4.2 fg	12.8	± 2 c	1.0	± 0.3 i	83.6	± 8.9 e-g	17.5	± 2.3 b-e
Kostyn's Red Russian	5.7	± 0.7 i	27.7	± 3.8 fg	11.5	± 1.8 c-e	0.8	± 0.3 i	80.4	± 13.3 f-h	17.8	± 2.4 b-e
Metechi	6.7	± 0.5 f-h	31.0	± 4.1 d-f	13.8	± 1.2 a-c	0.8	± 0.3 i	87.5	± 4.0 d-g	17.1	± 0.2 b-e
Metechi-2	6.7	± 0.7 fg	33.6	± 4.4 cd	13.8	± 1.5 a-c	0.9	± 0.3 i	89.6	± 6.6 d-f	21.5	± 1.8 a
Northern Siberian	6.3	± 0.6 f-h	28.7	± 2.4 fg	12.8	± 2.1 c	0.9	± 0.3 i	83.0	± 7.6 e-g	16.1	± 3.1 e
Siberian red	7.2	± 0.9 ef	30.6	± 3.6 ef	13.3	± 1.9 bc	1.4	± 1.5 e-i	78.4	± 7.0 f-h	15.2	± 2.0 ef
Wenger's Russian	6.7	± 0.7 fg	30.7	± 4.8 ef	13.2	± 2.4 bc	1.0	± 0.2 hi	89.1	± 8.0 d-f	16.7	± 2.4 c-e
Georgian Crystal	7.6	± 0.5 c-e	37.0	± 3.7 b	13.1	± 2.4 bc	1.0	± 0.3 g-i	116.3	± 9.5 bc	20.1	± 4.7 ab
Purple Stripe												
German White	6.9	± 0.6 f	35.8	± 4 bc	13.7	± 2.3 bc	1.4	± 0.4 e-g	118.4	± 9.8 b	21.0	± 2.1 a
Leningrad	6.6	± 1.1 f-h	35.9	± 7 bc	13.5	± 2.1 bc	1.7	± 0.3 c-e	123.5	± 15.9 ab	22.0	± 7.3 a
Music	7.1	± 0.7 ef	42.2	± 7.3 a	14.0	± 2.6 a-c	2.0	± 0.4 cd	130.9	± 33.5 a	22.2	± 6.0 a
Music-2	7.5	± 0.7 d-f	45.2	± 2.5 a	14.0	± 1.4 a-c	1.2	± 0.4 f-i	105.5	± 6.4 cd	18.2	± 4.6 b-e
Northen Quebec	6.9	± 0.7 f	38.4	± 3.3 b	14.3	± 1.9 ab	1.6	± 0.3 e-g	119.9	± 13.9 b	19.4	± 5.1 a-c
Susan Delafield	6.6	± 0.6 f-h	32.2	± 8 de	11.5	± 2.9 c-e	1.4	± 0.4 e-h	104.6	± 18.1 cd	19.6	± 3.4 ab
Ukrainian Mavniv	6.5	± 0.6 f-h	31.9	± 4 de	14.2	± 1.8 ab	1.4	± 0.3 e-g	105.8	± 11.0 c	15.5	± 3.1 ef
Yugoslavian Porcelain	6.1	± 0.3 g-i	38.5	± 3.9 b	13.4	± 2.3 bc	1.3	± 0.3 e-i	111.0	± 13.0 bc	22.9	± 3.0 a
Chesnok Red	7.2	± 0.9 ef	28.8	± 4 fg	13.2	± 2.6 bc	0.8	± 0.3 i	78.9	± 10.0 f-h	16.2	± 3.1 de
Rocambole												
Baba Franchuk's	7.3	± 0.6 d-f	26.5	± 3.4 g-i	10.2	± 2.2 ef	2.1	± 1.8 cd	85.2	± 9.5 e-g	11.3	± 2.8 g
French Rocambole	7.7	± 1.1 cd	27.7	± 3 fg	11.3	± 1.8 c-e	1.2	± 0.4 g-i	84.4	± 8.7 e-g	13.3	± 3.8 fg
German Red	7.2	± 1.3 ef	25.2	± 2.9 hi	12.0	± 2.1 c-e	1.4	± 0.2 e-i	82.5	± 1.3 e-h	13.3	± 4.9 e-g
Italian Purple	7.3	± 1 d-f	28.4	± 3.2 fg	12.1	± 1.9 cd	1.7	± 0.3 c-e	86.6	± 5.7 d-g	12.1	± 2.5 g
Killarney red	8.0	± 1 c	30.8	± 2.9 ef	11.1	± 1.8 c-e	2.1	± 1.9 c	90.5	± 5.7 de	11.1	± 1.9 g
Marino	7.6	± 1.1 c-e	26.1	± 5.8 g-i	9.9	± 2.6 f	1.2	± 0.5 f-i	70.5	± 2.1 gh	9.0	± 0.9 g
Mountain top	7.5	± 0.9 de	28.0	± 3.1 fg	11.3	± 1.6 c-e	1.7	± 0.4 de	90.5	± 4.4 de	12.3	± 1.6 g
Penn wonder	8.0	± 0.9 c	28.1	± 2.8 fg	11.3	± 1.8 c-e	1.3	± 0.4 f-i	81.9	± 9.1 e-h	9.5	± 1.8 g
Silverskin												
Silver Rose	7.7	± 1.2 cd	24.0	± 5.8 i	9.9	± 2.6 f	-	- -	-	- -	-	- -
Silver White	8.5	± 1.2 b	29.8	± 6.2 ef	10.4	± 2.5 ef	-	- -	-	- -	-	- -
Turban												
Maiskij	6.5	± 0.8 f-h	25.1	± 3.6 i	12.3	± 1.9 c	0.9	± 0.2 i	69.0	± 16.5 h	18.1	± 4.2 b-e
Portugese	6.0	± 0.7 hi	29.9	± 3.7 ef	12.6	± 1.7 c	1.0	± 0.2 i	80.6	± 15.8 f-h	18.3	± 3.8 b-d
Max	9.0		45.2		15.2		9.0		130.9		22.9	
Average	7.1		30.6		12.3		1.7		91.4		15.9	
Min	5.1		23.2		7.2		0.8		64.0		9.0	

Suppl. Table 2: Plant characteristics of 36 garlic cultivars grown at Sainte-Anne-de-Bellevue in 2020 (S20). Means in a given column followed by different letters are significantly different ($LSD_{0.05}$).

	Leaves Number		Plant Height (cm)		Stem Diameter (mm)		Flower curl		Flowering stem height (cm)		Spathe Length (cm)	
Cultivar	Mean	sd	Mean	sd	Mean	sd	Mean	sd	Mean	sd	Mean	sd
Artichoke												
Italian	7.4	± 1.1 bc	22.8	± 3.1 r	9.2	± 1.8 k-m	-	- -	35.8	± 9.1 p	22.1	± 7.2 a
Asiatic												
Tibetan red	6.0	± 0.7 ij	27.6	± 4.2 k-m	9.9	± 1.4 i-k	1.3	± 0.4 f-h	80.6	± 4.3 e-g	10.4	± 1.4 i
Tibétain red	6.2	± 0.7 hi	25.8	± 3.5 n-q	8.8	± 1.3 mn	1.6	± 0.4 bc	81.3	± 8.6 e-g	9.9	± 1.4 ij
Creole												
Ajo Rojo	3.9	± 0.6 m	10.0	± 1.9 t	3.1	± 1.3 q	-	- -	-	- -	-	- -
Creole Red	5.0	± 0.9 l	17.0	± 7.1 s	5.5	± 3.1 p	0.8	± 0.7 o	47.8	± 12.7 p	12.2	± 1.8 hi
Rose de Lautrec	5.4	± 0.7 k	29.7	± 5.0 ij	11.4	± 2.5 e-h	1.0	± 0.4 l-o	73.3	± 10 i-l	9.3	± 2.1 i-k
Porcelain												
Duganskij	6.6	± 0.9 d-f	25.8	± 4.6 n-q	11.3	± 2.6 f-i	1.1	± 0.4 i-m	64.5	± 8.5 o	10.9	± 2.6 i
Eureka Myrtis	7.0	± 1.0 d	28.7	± 2.9 jk	14.4	± 1.9 a	1.0	± 0.2 l-o	77.7	± 5.4 g-j	16.0	± 1.9 d-f
Kostyn's Red Russian	7.0	± 0.8 d	31.2	± 4.7 hi	13.2	± 2.5 bc	1.3	± 0.3 g-i	78.1	± 7.3 f-i	13.8	± 1.9 f-h
Metechi	6.7	± 0.7 d-f	31.4	± 3.9 g-i	11.8	± 1.8 d-g	1.4	± 0.3 c-g	77.0	± 12.2 g-k	13.2	± 1.6 gh
Metechi-2	6.5	± 0.9 d-g	31.9	± 4 f-h	11.2	± 1.6 f-i	1.1	± 0.3 j-o	85.6	± 8.3 e	14.7	± 2.8 fg
Northern Siberian	6.7	± 0.9 d-f	28.6	± 3.7 jk	11.9	± 1.9 d-f	1.1	± 0.4 i-m	73.3	± 8.4 i-l	13.0	± 2.4 h
Siberian red	6.7	± 0.7 de	29.8	± 3.7 ij	11.8	± 2.2 d-g	1.1	± 0.3 i-l	71.7	± 10.2 k-m	13.6	± 2.5 gh
Wenger's Russian	7.4	± 0.7 bc	30.9	± 4.9 hi	13.5	± 2.3 b	1.1	± 0.4 j-n	74.5	± 6.9 h-l	13.5	± 2.5 gh
Georgian Crystal	7.0	± 0.6 d	39.1	± 3.7 b-d	11.7	± 1.3 d-h	1.3	± 0.4 e-h	112.0	± 7.1 c	18.2	± 1.8 bc
Purple Stripe												
German White	7.1	± 0.7 d	40.0	± 3.6 a-c	12.4	± 1.5 cd	1.4	± 0.4 d-h	119.4	± 9.1 b	18.4	± 2.5 bc
Leningrad	7.2	± 0.6 cd	40.6	± 4.2 ab	12.2	± 1.7 de	1.7	± 0.4 ab	124.8	± 8.5 b	18.3	± 3.9 bc
Music	7.0	± 0.7 d	41.2	± 6.0 a	13.3	± 1.9 b	1.9	± 0.6 a	132.1	± 10.4 a	19.6	± 3.5 b
Music-2	7.2	± 0.4 b-d	38.8	± 5.6 b-d	11.4	± 1.3 e-i	1.8	± 0.6 ab	88.9	± 4.7 e	16.5	± 2.1 c-f
Northen Quebec	6.8	± 0.5 de	39.2	± 3.5 b-d	11.8	± 1.5 d-g	1.6	± 0.4 bc	120.2	± 7.5 b	18.2	± 3.5 bc
Susan Delafield	6.8	± 0.8 d	38.2	± 4.4 cd	11.8	± 1.6 d-g	1.5	± 0.3 b-e	106.1	± 25.3 cd	16.1	± 4.1 d-f
Ukrainian Mavniv	6.6	± 0.8 d-g	34.0	± 4.3 e	10.6	± 1.8 i	1.5	± 0.4 b-f	104.3	± 8.6 d	17.3	± 2.9 cd
Yugoslavian Porcelain	6.7	± 0.8 d-f	37.3	± 8.2 d	10.6	± 2.6 i	1.5	± 0.4 b-f	110.8	± 15.4 c	19.6	± 4.4 b
Chesnok Red	6.0	± 0.7 ij	28.6	± 3.4 jk	9.4	± 1.3 j-m	1.0	± 0.3 m-o	77.0	± 6.5 g-j	12.8	± 2.4 h
Persian star	5.5	± 0.8 k	30.1	± 6.3 h-j	9.3	± 2.5 k-m	1.3	± 0.4 g-i	80.7	± 14.6 e-g	10.4	± 2.2 i
Rocambole												
Baba Franchuk's	6.4	± 0.7 f-h	26.2	± 3.2 m-p	9.7	± 1.2 jkl	1.4	± 0.4 c-f	79.7	± 6.2 e-g	10.0	± 1.7 ij
French Rocambole	6.7	± 0.6 de	26.9	± 3 l-n	9.6	± 1.4 j-l	1.2	± 0.3 h-j	78.5	± 7.8 f-h	11.0	± 1.6 i
German Red	6.2	± 0.7 g-i	25.2	± 2.7 n-q	9.3	± 1.3 j-m	1.4	± 0.4 d-h	81.6	± 3.9 e-g	11.0	± 1.1 i
Italian Purple	6.5	± 0.7 e-g	24.4	± 3.6 qr	8.2	± 1.7 no	1.5	± 0.4 b-d	67.8	± 8.7 m-o	8.4	± 2.1 k
Killarney red	6.9	± 0.7 d	26.3	± 3.0 l-p	9.1	± 1.7 lm	1.5	± 0.5 b-e	72.0	± 7.7 k-m	9.1	± 1.4 jk
Marino	7.0	± 1.1 d	25.2	± 3 opq	9.4	± 1.1 j-m	1.2	± 0.3 h-k	72.6	± 7.9 j-m	8.7	± 1.3 jk
Mountain top	6.5	± 0.7 d-g	26.7	± 3.4 l-o	9.9	± 1.2 i-k	1.5	± 0.5 b-e	82.8	± 7.5 ef	10.9	± 2.1 i
Penn wonder	6.9	± 0.6 d	24.9	± 3.6 pq	9.7	± 1.5 j-l	1.2	± 0.3 h-j	70.4	± 8.8 l-n	8.6	± 1.4 jk
Silverskin												
Silver Rose	7.7	± 0.9 ab	25.8	± 3.7 n-q	10.0	± 2.2 ij	0.7	± 0.6 o	-	- -	-	- -
Silver White	8.0	± 0.9 a	33.4	± 4.7 ef	10.9	± 1.8 hi	1.1	± 0.7 j-o	76.3	± 8.3 g-l	16.6	± 5.7 c-e
Turban												
Maiskij	6.1	± 1.0 i	27.8	± 3.8 kl	11.3	± 2.6 f-i	1.0	± 0.2 no	65.9	± 5.1 no	13.9	± 2.1 f-h
Portugese	6.1	± 0.7 i	32.9	± 3.8 e-g	11.1	± 1.3 g-i	1.1	± 0.3 k-o	81.3	± 11.9 e-g	15.3	± 3.4 ef
Unclassified												
Sweet Haven	5.7	± 0.7 jk	24.5	± 5.3 p-r	7.4	± 1.6 o	1.6	± 0.4 bc	70.8	± 8.2 k-n	9.5	± 1.5 i-k
Max	8.0		41.2		14.4		1.9		132.1		22.1	
Average	6.6		29.7		10.5		1.3		83.3		13.6	
Min	3.9		10.0		3.1		0.7		35.8		8.4	

Suppl. Table 3: Plant characteristics of 36 garlic cultivars grown at L'Ascension-de-Notre-Seigneur in 2019 (N19**).** Means in a given column followed by different letters are significantly different ($LSD_{0.05}$).

	Leaves per Plant			Plant Height (cm)			Stem Diameter (mm)			Flower turns			Flowering stem height (cm)			Spathe Length (cm)		
Cultivar	Mean		sd	Mean		sd	Mean		sd	Mean		sd	Mean		sd	Mean		sd
Artichoke																		
Italian	6.3	±	1.2 h	16.0	±	4.4 kl	7.3	±	2.3 hi	-	-	-	-	-	-	-	-	-
Creole																		
Ajo Rojo	5.6	±	0.8 jk	13.8	±	3.1 m	6.6	±	1.2 ij	-	-	-	-	-	-	-	-	-
Creole Red	5.2	±	0.9 k	13.0	±	3.0 m	6.8	±	2.4 ij	-	-	-	-	-	-	-	-	-
Rose de Lautrec	6.1	±	1.0 hi	23.1	±	5.8 ef	10.0	±	2.2 d-f	1.1	±	0.5 j	64.8	±	14.9 k	11.3	±	4.1 f-i
Porcelain																		
Duganskij	8.1	±	1.0 a-c	23.0	±	3.1 f	12.3	±	1.4 c	1.1	±	0.3 ij	74.2	±	8.2 h-k	14.7	±	2.3 b-d
Eureka Myrtis	7.0	±	1.2 e-g	23.8	±	5 ef	10.5	±	2.5 d	1.4	±	0.5 e-h	85.2	±	17.7 f-h	15.1	±	2.1 bc
Kostyn's Red Russian	6.9	±	0.9 fg	22.4	±	4.2 fg	9.8	±	1.5 d-f	1.6	±	0.4 c-e	81.2	±	14.7 g-i	13.6	±	2.7 c-e
Metechi-2	6.5	±	1.0 gh	19.2	±	1.7 h-k	10.0	±	1.7 d-f	1.2	±	0.3 g-j	68.5	±	5.5 i-k	15.0	±	0.8 b-d
Siberian red	7.2	±	1.1 d-g	22.5	±	5.1 fg	10.6	±	2.9 d	1.2	±	0.3 h-j	70.6	±	27.6 i-k	13.3	±	1.7 c-f
Wenger's Russian	7.5	±	1.0 de	23.1	±	2.6 ef	10.2	±	1.8 de	1.5	±	0.3 d-g	84.0	±	7.1 f-h	13.3	±	1.7 c-f
Purple Stripe																		
Georgian Crystal	7.5	±	0.5 c-e	31.7	±	2.7 ab	15.1	±	1.5 a	1.5	±	0.5 c-g	117.7	±	5.1 a-c	20.4	±	2.2 a
German White	7.8	±	0.6 b-d	32.4	±	4.4 a	14.8	±	1.6 ab	1.5	±	0.5 e-h	130.4	±	6.6 a	19.6	±	1.1 a
Leningrad	7.2	±	0.5 d-g	29.4	±	3.5 bc	14.9	±	1.9 a	1.2	±	0.3 ij	107.9	±	14.6 cd	19.9	±	3.5 a
Northen Quebec	7.4	±	0.6 d-f	27.5	±	6.9 cd	14.2	±	1.4 ab	1.3	±	0.4 f-j	108.7	±	8.8 b-d	19.6	±	1.4 a
Ukrainian Mavniv	7.7	±	0.5 b-d	31.0	±	2.8 ab	13.7	±	1.6 b	1.8	±	0.4 a-c	121.7	±	7.9 ab	16.7	±	2.6 b
Yugoslavian Porcelain	7.5	±	0.5 de	30.2	±	5.2 ab	14.0	±	1.5 ab	1.3	±	0.5 g-j	113.0	±	8.3 bc	19.9	±	3.0 a
Chesnok Red	6.9	±	1.1 fg	19.9	±	5.3 h-j	9.7	±	2.2 d-f	1.1	±	0.2 ij	76.2	±	11.2 h-j	15.2	±	3.3 bc
Rocambole																		
Baba Franchuk's	7.3	±	0.7 d-f	23.2	±	4.8 ef	9.4	±	1.6 ef	1.7	±	0.4 a-d	89.4	±	19.3 e-g	12.2	±	3.5 e-h
French Rocambole	8.5	±	0.8 a	24.9	±	2.3 d-f	10.8	±	1.0 d	1.8	±	0.3 a-d	98.8	±	6.6 de	14.2	±	0.8 b-e
Italian Purple	6.8	±	1.0 g	20.7	±	2.9 g-i	8.9	±	1.8 f	1.6	±	0.5 c-f	81.6	±	7.0 gh	11.2	±	3.3 g-i
Marino	6.8	±	1.0 fg	18.6	±	3.0 i-k	7.9	±	1.3 gh	1.6	±	0.5 b-e	67.8	±	11.9 jk	10.2	±	1 hi
Mountain top	7.1	±	0.7 e-g	23.0	±	3.3 f	9.1	±	1.6 f	1.9	±	0.2 a	92.2	±	8.8 ef	12.6	±	2 d-g
Penn wonder	7.0	±	1.0 e-g	18.4	±	3.7 jk	9.3	±	2.1 ef	1.9	±	0.3 a	87.3	±	12.3 e-g	9.8	±	1 i
Silverskin																		
Silver Rose	5.7	±	0.6 ij	15.0	±	2.4 lm	6.0	±	1.1 j	-	-	-	-	-	-	-	-	-
Silver White	5.7	±	0.8 ij	18.1	±	3.0 jk	7.2	±	1.6 hi	-	-	-	-	-	-	-	-	-
Turban																		
Maiskij	7.4	±	0.9 d-f	22.1	±	1.1 f-h	9.9	±	1.5 d-f	1.4	±	0.3 e-i	75.3	±	4.4 h-k	15.1	±	0.7 bc
Portugese	6.2	±	0.9 h	20.2	±	3.1 h-j	8.7	±	1.6 fg	1.5	±	0.3 e-g	85.6	±	12.2 fg	13.7	±	2.7 c-e
Unclassified																		
Sweet Haven	8.3	±	0.9 ab	25.3	±	3.3 de	10.6	±	1.6 d	1.8	±	0.3 ab	92.0	±	8.6 e-g	12.0	±	2.1 e-i
Max	8.5			32.4			15.1			1.9			130.4			20.4		
Average	7.0			22.6			10.3			1.5			90.2			14.7		
Min	5.2			13.0			6.0			1.1			64.8			9.8		

Suppl. Table 4: Plant characteristics of 36 garlic cultivars grown at Alma, Qc, Canada in 2020 (N20). Means in a given column followed by different letters are significantly different ($LSD_{0.05}$).

Cultivar	Leaves Number		Plant Height (cm)		Stem Diameter (mm)		Flower turns		Flowering stem height (cm)		Spathe Length (cm)	
	Mean	sd	Mean	sd	Mean	sd	Mean	sd	Mean	sd	Mean	sd
Artichoke												
Italian	8.3	± 0.6 a	26.0	± 1.7 b-e	10.3	± 2.9 ab	-	- -	-	- -	-	- -
Asiatic												
Tibetan red	6.0	± 0.7 c-f	24.1	± 4.7 de	8.8	± 2.2 b-d	1.7	± 0.5 ab	91.0	± 15.2 de	8.8	± 2.3 d-f
Creole												
Rose de Lautrec	6.1	± 1.1 b-e	28.5	± 5.7 ab	10.5	± 2.5 a	0.9	± 0.6 h	88.0	± 10.5 d-f	9.2	± 1.5 d-f
Porcelain												
Duganskij	6.4	± 0.6 b	26.5	± 4.4 bc	9.0	± 1.3 bc	1.5	± 0.4 a-d	77.6	± 10.5 fg	9.1	± 1.7 d-f
Eureka Myrtis	5.9	± 0.6 c-g	25.6	± 4.3 c-e	9.7	± 2.0 ab	1.3	± 0.3 ef	78.5	± 11.7 fg	10.0	± 2.5 c-e
Kostyn's Red Russian	5.8	± 0.8 d-i	23.9	± 4.8 de	9.0	± 2.1 bc	1.1	± 0.2 gh	86.3	± 10.3 ef	12.0	± 2.3 a-c
Metechi	5.7	± 0.7 e-i	21.5	± 4.4 e-g	8.7	± 1.7 b-d	1.1	± 0.2 f-h	73.4	± 13.9 g	8.9	± 2.3 d-f
Wenger's Russian	6.2	± 0.7 b-d	27.5	± 4.4 b	9.9	± 2.2 ab	1.3	± 0.3 ef	84.7	± 8.6 e-g	10.0	± 2.2 c-e
Georgian Crystal	6.2	± 0.6 b-d	28.5	± 3.7 ab	8.9	± 2.3 b-d	1.7	± 0.6 a-d	104.8	± 11.5 a-c	12.3	± 2.5 ab
Purple Stripe												
German White	5.9	± 0.8 c-h	25.8	± 6.7 b-e	9.2	± 2.4 bc	1.7	± 0.4 a-c	108.2	± 16.1 ab	10.5	± 3.2 b-e
Leningrad	6.5	± 0.7 b	28.8	± 6.2 ab	9.7	± 2.6 ab	1.6	± 0.4 a-d	114.6	± 15.7 ab	10.9	± 2.5 b-d
Northen Quebec	6.3	± 0.6 bc	30.0	± 4.0 a	10.0	± 2.3 ab	1.3	± 0.3 ef	116.1	± 7.9 a	9.9	± 2.0 c-e
Ukrainian Mavniv	6.1	± 0.7 b-d	26.1	± 5.5 b-d	9.9	± 2.3 ab	1.6	± 0.4 a-d	110.1	± 21.4 ab	14.1	± 2.3 a
Yugoslavian Porcelain	6.1	± 0.3 b-f	31.0	± 3.6 a	9.8	± 1.6 ab	1.5	± 0.4 b-e	103.9	± 18.4 bc	13.8	± 4.1 a
Chesnok Red	6.3	± 0.5 bc	24.7	± 5.4 c-e	7.8	± 1.7 c-f	1.4	± 0.5 c-e	84.6	± 9.4 e-g	9.5	± 2.0 d-f
Persian star	5.6	± 0.7 f-i	21.6	± 3.3 e-g	7.7	± 1.4 d-f	1.2	± 0.3 e-g	83.6	± 7.7 e-g	9.3	± 1.8 d-f
Rocambole												
Baba Franchuk's	5.9	± 0.7 c-g	20.7	± 4.1 e-g	8.6	± 1.3 b-e	1.7	± 0.4 ab	102.2	± 8.5 b-d	8.2	± 2.5 d-f
French Rocambole	5.9	± 0.8 c-g	20.1	± 4.0 fg	7.5	± 2.0 ef	1.6	± 0.6 a-d	83.4	± 9.0 e-g	8.0	± 1.7 ef
Italian Purple	5.5	± 0.9 g-i	17.1	± 5.1 g	6.4	± 1.9 fg	1.4	± 0.7 c-e	83.0	± 16.9 e-g	8.2	± 0.4 d-f
Killarney red	5.4	± 0.5 i	20.0	± 3.5 fg	6.1	± 1.2 g	1.9	± 0.5 a	91.8	± 11.6 c-e	7.2	± 1.6 f
Marino	5.2	± 1.0 i	22.1	± 3.7 e-g	8.2	± 1.5 b-f	2.0	± 12.0 a	90.7	± 4.0 d-f	10.5	± 2.3 b-e
Mountain top	5.8	± 0.5 c-i	22.4	± 3.2 ef	8.3	± 1.5 b-e	1.7	± 0.5 a-c	89.0	± 12.7 d-f	10.1	± 2.4 b-e
Penn wonder	5.8	± 1.0 c-i	17.4	± 5.3 g	6.4	± 2.0 fg	1.4	± 0.6 d-f	85.5	± 7.2 e-g	7.8	± 1.3 ef
Silverskin												
Silver White	7.4	± 1.3 a	18.4	± 6.2 fg	7.2	± 2.4 e-g	-	- -	-	- -	-	- -
Turban												
Maiskij	5.5	± 0.7 hi	23.4	± 3.9 e	8.5	± 1.9 b-e	1.1	± 0.3 f-h	73.8	± 3.5 fg	10.8	± 0.3 b-e
Portugese	6.0	± 0.5 c-f	24.2	± 4.3 de	8.8	± 1.3 b-d	1.3	± 0.4 e-g	86.0	± 12.9 ef	10.5	± 3.0 b-e
Unclassified												
Sweet Haven	6.1	± 0.6 b-f	20.6	± 3.4 e-g	6.5	± 1.1 fg	1.4	± 0.8 c-f	78.2	± 8.7 fg	8.5	± 0.1 d-f
Max	8.3		31.0		10.5		2.0		116.1		14.1	
Average	6.1		23.9		8.6		1.5		90.8		9.9	
Min	5.2		17.1		6.1		0.9		73.4		7.2	

Suppl. Table 5: Relative frequency (%) of plant qualitative traits of 36 garlic cultivars grown at Sainte-Anne-de-Bellevue, Québec, Canada in 2019 (S19).

Cultivar	Pseudostem Coloration		Leaves Rigidity		Leaves attitude		
	Absence	Presence	High	Low	Erect	Intermdiate	Semi-erect
Artichoke							
Italian	11.1	88.9	0.0	100.0	100.0	0.0	0.0
Asiatic							
Tibetan red	58.1	41.9	0.0	100.0	64.5	35.5	0.0
Tibétain red	92.3	7.7	5.1	94.9	38.5	61.5	0.0
Creole							
Creole Red	100.0	0.0	100.0	0.0	0.0	100.0	0.0
Rose de Lautrec	80.6	19.4	58.1	41.9	19.4	38.7	41.9
Marble Purple Stripe							
Duganskij	0.0	100.0	100.0	0.0	93.3	6.7	0.0
Eureka Myrtis	0.0	100.0	66.7	33.3	0.0	66.7	33.3
Kostyn's Red Russian	0.0	100.0	100.0	0.0	0.0	34.3	65.7
Metechi	0.0	100.0	83.3	16.7	0.0	100.0	0.0
Metechi-2	77.8	22.2	94.4	5.6	0.0	100.0	0.0
Northern Siberian	2.3	97.7	65.1	34.9	0.0	34.9	65.1
Siberian red	25.0	75.0	100.0	0.0	0.0	78.6	21.4
Wenger's Russian	2.9	97.1	100.0	0.0	0.0	57.1	42.9
Porcelain							
Georgian Crystal	100.0	0.0	100.0	0.0	0.0	100.0	0.0
German White	100.0	0.0	100.0	0.0	6.5	93.5	0.0
Leningrad	89.5	10.5	100.0	0.0	42.1	57.9	0.0
Music	88.6	11.4	100.0	0.0	0.0	100.0	0.0
Music-2	50.0	50.0	100.0	0.0	0.0	100.0	0.0
Northen Quebec	94.4	5.6	100.0	0.0	5.6	94.4	0.0
Susan Delafield	84.2	15.8	100.0	0.0	0.0	100.0	0.0
Ukrainian Mavniv	64.3	35.7	100.0	0.0	0.0	100.0	0.0
Yugoslavian Porcelain	90.0	10.0	100.0	0.0	0.0	100.0	0.0
Purple Stripe							
Chesnok Red	48.4	51.6	100.0	0.0	0.0	100.0	0.0
Rocambole							
Baba Franchuk's	52.6	47.4	0.0	100.0	78.9	21.1	0.0
French Rocambole	60.0	40.0	0.0	100.0	42.9	57.1	0.0
German Red	100.0	0.0	40.0	60.0	0.0	100.0	0.0
Italian Purple	40.0	60.0	0.0	100.0	26.7	73.3	0.0
Killarney red	37.5	62.5	3.1	96.9	56.2	0.0	43.8
Marino	57.1	42.9	7.1	92.9	92.9	7.1	0.0
Mountain top	79.5	20.5	0.0	100.0	65.9	34.1	0.0
Penn wonder	77.5	22.5	0.0	100.0	100.0	0.0	0.0
Silverskin							
Silver Rose	85.7	14.3	100.0	0.0	39.3	60.7	0.0
Silver White	84.8	15.2	0.0	100.0	42.4	57.6	0.0
Turban							
Maiskij	0.0	100.0	100.0	0.0	0.0	9.4	90.6
Portugese	100.0	0.0	100.0	0.0	0.0	35.0	65.0

Suppl. Table 6: Relative frequency (%) of plant qualitative traits of 36 garlic cultivars grown at Sainte-Anne-de-Bellevue, Québec, Canada in 2020 (S20).

	Pseudostem Coloration		Leaves Rigidity		Leaves attitude		
Cultivar	Absence	Presence	High	Low	Erect	Intermdiate	Semi-erect
Artichoke							
Italian	1.8	98.2	0.0	100.0	100.0	0.0	0.0
Asiatic							
Tibetan red	13.7	86.3	0.0	100.0	72.5	27.5	0.0
Tibétain red	22.0	78.0	0.0	100.0	100.0	0.0	0.0
Creole							
Ajo Rojo	40.0	60.0	26.7	73.3	33.3	66.7	0.0
Creole Red	12.5	87.5	60.0	40.0	22.5	77.5	0.0
Rose de Lautrec	65.4	34.6	34.6	65.4	36.5	61.5	1.9
Marble Purple Stripe							
Duganskij	0.0	100.0	100.0	0.0	0.0	84.6	15.4
Eureka Myrtis	19.2	80.8	100.0	0.0	34.6	32.7	32.7
Kostyn's Red Russian	12.5	87.5	100.0	0.0	0.0	64.6	35.4
Metechi	7.7	92.3	69.2	30.8	34.6	61.5	3.8
Metechi-2	85.3	14.7	61.8	38.2	35.3	64.7	0.0
Northern Siberian	8.0	92.0	100.0	0.0	0.0	34.0	66.0
Siberian red	0.0	100.0	100.0	0.0	0.0	100.0	0.0
Wenger's Russian	6.7	93.3	100.0	0.0	3.3	91.7	5.0
Georgian Crystal	100.0	0.0	69.2	30.8	34.6	65.4	0.0
Porcelain							
German White	100.0	0.0	72.7	27.3	0.0	100.0	0.0
Leningrad	100.0	0.0	100.0	0.0	0.0	100.0	0.0
Music	96.2	3.8	100.0	0.0	0.0	100.0	0.0
Music-2	60.0	40.0	100.0	0.0	0.0	100.0	0.0
Northen Quebec	100.0	0.0	100.0	0.0	0.0	100.0	0.0
Susan Delafield	100.0	0.0	100.0	0.0	0.0	65.6	34.4
Ukrainian Mavniv	80.4	19.6	100.0	0.0	0.0	100.0	0.0
Yugoslavian Porcelain	93.9	6.1	100.0	0.0	3.0	51.5	45.5
Purple Stripe							
Chesnok Red	98.3	1.7	100.0	0.0	0.0	81.7	18.3
Persian star	100.0	0.0	100.0	0.0	0.0	0.0	100.0
Rocambole							
Baba Franchuk's	34.0	66.0	0.0	100.0	100.0	0.0	0.0
French Rocambole	50.8	49.2	0.0	100.0	30.5	69.5	0.0
German Red	43.8	56.2	0.0	100.0	43.8	56.2	0.0
Italian Purple	29.4	70.6	2.0	98.0	29.4	35.3	35.3
Killarney red	38.3	61.7	1.7	98.3	81.7	18.3	0.0
Marino	20.0	80.0	0.0	100.0	100.0	0.0	0.0
Mountain top	53.4	46.6	0.0	100.0	32.8	67.2	0.0
Penn wonder	46.6	53.4	0.0	100.0	34.5	65.5	0.0
Silverskin							
Silver Rose	18.8	81.2	68.8	31.2	100.0	0.0	0.0
Silver White	53.2	46.8	37.1	62.9	91.9	8.1	0.0
Turban							
Maiskij	0.0	100.0	94.6	5.4	0.0	3.6	96.4
Portugese	98.3	1.7	100.0	0.0	11.7	56.7	31.7
Unclassified							
Sweet Haven	16.0	84.0	0.0	100.0	64.0	36.0	0.0

Suppl. Table 7: Relative frequency (%) of plant qualitative traits of 36 garlic cultivars grown at l'Ascension-de-Notre-Seigneur, Québec, Canada in 2019 (N19)**.**

	Pseudostem Coloration		Leaves Rigidity		Leaves attitude		
Cultivar	Absence	Presence	High	Low	Erect	Intermdiate	Semi-erect
Artichoke							
Italian	7.7	92.3	0.0	100.0	100.0	0.0	0.0
Creole							
Ajo Rojo	30.4	69.6	0.0	100.0	0.0	100.0	0.0
Creole Red	19.2	80.8	0.0	100.0	0.0	100.0	0.0
Rose de Lautrec	91.3	8.7	52.2	47.8	0.0	52.2	47.8
Marble Purple Stripe							
Duganskij	0.0	100.0	45.0	55.0	45.0	55.0	0.0
Eureka Myrtis	15.8	84.2	63.2	36.8	0.0	100.0	0.0
Kostyn's Red Russian	35.0	65.0	0.0	100.0	0.0	100.0	0.0
Metechi-2	83.3	16.7	0.0	100.0	0.0	100.0	0.0
Siberian red	15.8	84.2	0.0	100.0	5.3	94.7	0.0
Wenger's Russian	13.6	86.4	45.5	54.5	0.0	0.0	100.0
Porcelain							
Georgian Crystal	60.0	40.0	100.0	0.0	0.0	53.3	46.7
German White	94.1	5.9	100.0	0.0	0.0	58.8	41.2
Leningrad	64.7	35.3	47.1	52.9	0.0	52.9	47.1
Northen Quebec	93.3	6.7	0.0	100.0	0.0	53.3	46.7
Ukrainian Mavniv	87.5	12.5	100.0	0.0	0.0	100.0	0.0
Yugoslavian Porcelain	94.1	5.9	47.1	52.9	0.0	0.0	100.0
Purple Stripe							
Chesnok Red	88.9	11.1	100.0	0.0	0.0	63.0	37.0
Rocambole							
Baba Franchuk's	33.3	66.7	0.0	100.0	100.0	0.0	0.0
French Rocambole	0.0	100.0	0.0	100.0	100.0	0.0	0.0
Italian Purple	41.4	58.6	17.2	82.8	0.0	79.3	20.7
Marino	35.0	65.0	0.0	100.0	100.0	0.0	0.0
Mountain top	13.8	86.2	0.0	100.0	41.4	58.6	0.0
Penn wonder	53.8	46.2	15.4	84.6	84.6	0.0	15.4
Silverskin							
Silver Rose	93.3	3.3	43.3	56.7	56.7	43.3	0.0
Silver White	78.9	21.1	0.0	100.0	68.4	31.6	0.0
Turban							
Maiskij	0.0	100.0	50.0	50.0	0.0	78.6	21.4
Portugese	57.1	42.9	100.0	0.0	45.7	54.3	0.0
Unclassified							
Sweet Haven	21.1	78.9	0.0	100.0	100.0	0.0	0.0

Suppl. Table 8: Relative frequency (%) of plant qualitative traits of 36 garlic cultivars grown at Alma, Québec, Canada in 2020 (N20).

	Pseudostem Coloration		Leaves Rigidity		Leaves attitude		
Cultivar	Absence	Presence	High	Low	Erect	Intermdiate	Semi-erect
Artichoke							
Italian	0.0	100.0	0.0	100.0	100.0	0.0	0.0
Asiatic							
Tibetan red	0.0	100.0	0.0	100.0	80.0	20.0	0.0
Creole							
Rose de Lautrec	68.8	31.2	87.5	12.5	87.5	12.5	0.0
Marble Purple Stripe							
Duganskij	20.5	79.5	100.0	0.0	0.0	100.0	0.0
Eureka Myrtis	31.8	68.2	100.0	0.0	0.0	50.0	50.0
Kostyn's Red Russian	0.0	100.0	100.0	0.0	0.0	0.0	100.0
Metechi	42.1	57.9	100.0	0.0	0.0	57.9	42.1
Wenger's Russian	15.6	84.4	87.8	12.2	0.0	63.3	36.7
Porcelain							
Georgian Crystal	76.9	23.1	100.0	0.0	15.4	84.6	0.0
German White	83.3	16.7	25.0	75.0	29.2	70.8	0.0
Leningrad	35.7	64.3	100.0	0.0	35.7	64.3	0.0
Northen Quebec	69.0	31.0	100.0	0.0	0.0	100.0	0.0
Ukrainian Mavniv	83.9	16.1	51.6	48.4	16.1	51.6	32.3
Yugoslavian Porcelain	58.8	41.2	100.0	0.0	5.9	94.1	0.0
Purple Stripe							
Chesnok Red	100.0	0.0	33.3	66.7	0.0	91.7	8.3
Persian star	91.7	8.3	100.0	0.0	0.0	58.3	41.7
Rocambole							
Baba Franchuk's	5.3	94.7	0.0	100.0	10.5	89.5	0.0
French Rocambole	0.0	100.0	0.0	100.0	17.9	82.1	0.0
Italian Purple	17.6	82.4	11.8	88.2	88.2	11.8	0.0
Killarney red	10.5	89.5	0.0	100.0	42.1	57.9	0.0
Marino	0.0	100.0	0.0	100.0	25.0	75.0	0.0
Mountain top	0.0	100.0	0.0	100.0	0.0	100.0	0.0
Penn wonder	0.0	100.0	0.0	100.0	45.0	55.0	0.0
Silverskin							
Silver White	0.0	100.0	0.0	100.0	60.0	40.0	0.0
Turban							
Maiskij	0.0	100.0	100.0	0.0	0.0	4.3	95.7
Portugese	100.0	0.0	100.0	0.0	0.0	58.5	41.5
Unclassified							
Sweet Haven	0.0	100.0	0.0	100.0	100.0	0.0	0.0