Acta Universitatis Agriculturae Sueciae

Doctoral Thesis No. 2025:10
Faculty of Landscape Architecture, Horticulture and Crop Production Science

Towards sustainable onion storage

Postharvest quality diagnostics using volatile organic compounds and physiological metrics

Isabella Kleman

Towards sustainable onion storage

Postharvest quality diagnostics using volatile organic compounds and physiological metrics

Isabella Kleman
Faculty of Landscape Architecture, Horticulture and Crop Production Science
Department of Biosystems and Technology
Alnarp

SLU
SWEDISH UNIVERSITY OF AGRICULTURAL SCIENCES

DOCTORAL THESIS
Alnarp 2025

Acta Universitatis Agriculturae Sueciae
2025:10

Cover: Onion bulbs exhibiting sprouting, basal plate rot and soft rot. (photo: I. Kleman)

ISSN 1652-6880
ISBN (print version) 978-91-8046-445-1
ISBN (electronic version) 978-91-8046-495-6
https://doi.org/10.54612/a.5qnn6nnov2

Swedish University of Agricultural Sciences, Department of Biosystems and Technology, Alnarp, Sweden

Print: SLU Grafisk service, Alnarp 2025

Towards sustainable onion storage: Postharvest quality diagnostics using volatile organic compounds and physiological metrics

Abstract

Onion is an important component of the diets of many cultures worldwide, and also one of the most commonly produced vegetables. The onion bulb is a well-adapted storage organ and many of the harvested bulbs are stored for several months to ensure year-round availability to consumers. However, significant amounts are lost due to sprouting, loss of water, loss of dry matter, fungal infections and bacterial infections. To optimise storage and enable sales at the correct point in time to reduce losses decision support systems are needed to identify signs of quality problems. One potential such system is the monitoring of volatile organic compounds in the air of storage facilities to detect developing quality problems, such as storage diseases. In this thesis, the volatile organic compounds emanating into the headspace of onion bulbs affected by *Fusarium* basal plate rot, *Penicillium* rot or *Pectobacterium*-caused soft rot were investigated. The aim was to identify the compounds that are relevant as indicators of developing storage diseases, and which could be the target of future monitoring systems. Several relevant indicator compounds were found. Some of them are naturally occurring onion metabolites whose relative abundance changed, including methyl propyl disulfide and dimethyl disulfide. Others, such as ethenylbenzene and 2,3-butanediol are products typical of fungal or bacterial metabolism. Other systems for storage monitoring may include physiological measurements to predict quality changes such as sprouting. Therefore, the thesis also investigated the connection between selected physiological indicators and sprouting. It was found that the changing concentrations of glucose, fructose and sucrose, and the decreasing dry matter contents of the bulbs had a significant connection to the start of sprouting.

Keywords: *Allium cepa*, basal plate rot, blue mould, e-nose, food loss, non-structural carbohydrates, SPME-GC-MS, soft rot, sugar, VOC

Mot hållbar löklagring: Kvalitetsdiagnostik efter skörd med hjälp av flyktiga organiska ämnen och fysiologiska mätvärden

Sammanfattning

Lök är en viktig del av kosten i många kulturer världen över, och är också en av de mest producerade grönsakerna. Löken är ett välanpassat lagringsorgan, och många av de skördade lökarna lagras i flera månader för att säkerställa tillgång året runt för konsumenterna. Dock går betydande mängder förlorade på grund av groning, vattenförlust, torrsubstansförlust, svampangrepp och bakterieangrepp. För att optimera lagringen och möjliggöra försäljning vid rätt tidpunkt, och därigenom minska förlusterna, behövs beslutsstödsystem som kan identifiera tecken på utveckling av kvalitetsproblem. Ett potentiellt sådant system är övervakning av flyktiga organiska ämnen i luften i lagringsutrymmen för att upptäcka problem såsom lagringssjukdomar. I denna avhandling undersöktes de flyktiga organiska ämnen som avges från lök som påverkats av *Fusarium*-orsakad basalröta, *Penicillium*-mögel eller röta orsakad av *Pectobacterium*. Syftet var att identifiera de ämnen som är relevanta som indikatorer på utveckling av lagringssjukdomar och som kan bli mål för framtida övervakningssystem. Flera relevanta indikatorämnen identifierades. Några av dem är naturligt förekommande lökmolekyler vars relativa koncentrationer förändrades, såsom metylpropyl-disulfid och dimetyl-disulfid. Andra, såsom styren och 2,3-butanediol, är typiska produkter av svamp- eller bakteriemetabolism. Andra system för lagringsövervakning kan inkludera fysiologiska mätningar för att förutsäga kvalitetsförändringar i form av groning. Därför undersökte avhandlingen också kopplingen mellan utvalda fysiologiska indikatorer och groning. Det visade sig att förändrade koncentrationer av glukos, fruktos och sukros samt minskande torrsubstansinnehåll hade en signifikant koppling till groningens start.

Keywords: *Allium cepa*, bakterieröta, basalröta, blåmögel, elektronisk näsa, icke-strukturella kolhydrater, matsvinn, socker, SPME-GC-MS

Dedication

To all the animals in my life, past and present.

Contents

List of publications 9

List of tables 11

List of figures 13

Abbreviations 15

1. Background 17
   - 1.1 Introduction 17
   - 1.2 Onion physiology 17
     - 1.2.1 Bulb formation 17
     - 1.2.2 Dormancy, non-structural carbohydrates and sprouting 18
   - 1.3 Storage of onions and postharvest quality problems 19
     - 1.3.1 Harvest and pre storage treatment 19
     - 1.3.2 Storage conditions 20
     - 1.3.3 Losses due to quality problems 21
     - 1.3.4 Sprouting 21
     - 1.3.5 Fungal diseases 22
     - 1.3.6 Bacterial diseases 24
   - 1.4 Quality indicators for stored onion bulbs 24
     - 1.4.1 Physiological indicators 24
     - 1.4.2 The potential for volatile monitoring in crop storage 25
     - 1.4.3 Headspace volatiles of onions and postharvest pathogens 26

2. Aim and objectives 29

3. Method summary 31
   - 3.1 Plant material 31
   - 3.2 Physiological measurements (Paper I) 31
     - 3.2.1 Firmness and sprouting 31
     - 3.2.2 Sugar composition 32

3.2.3 Dry matter content 33
3.3 Pathogens (Paper II & III) 33
3.3.1 Fusarium oxysporum 33
3.3.2 Penicillium polonicum 34
3.3.3 Pectobacterium carotovorum 35
3.3.4 Other pathogens 35
3.4 Analysis of headspace volatiles (Paper II & III) 35

4. Results and discussion 37
4.1 Sugar, dry matter and firmness as indicators of sprouting 37
4.2 Headspace volatiles of uninfected bulbs 40
4.3 Headspace volatiles of bulbs with fungal infections 41
4.4 Headspace volatiles of bulbs with a bacterial infection 44
4.5 Practical applicability in storage 49

5. Conclusions and future perspectives 51

References 53

Popular science summary 65

Populärvetenskaplig sammanfattning 67

Acknowledgements 69

List of publications

This thesis is based on the work contained in the following papers, referred to by Roman numerals in the text:

I. **Kleman, I**., Rosberg, A. K., & Mogren, L. (2024). Sugar content and dry matter are key factors predicting sprouting of yellow bulb onions regardless of treatment with maleic hydrazide. *Acta Agriculturae Scandinavica, Section B—Soil & Plant Science*, *74*(1), 2366171. https://doi.org/10.1080/09064710.2024.2366171

II. **Kleman, I**., Rosberg, A. K., Guzhva, O., Karlsson, M.E., Becher, P.G. & Mogren, L. Headspace volatile organic compounds as indicators of Fusarium basal plate rot and Penicillium rot in stored onion bulbs (Submitted)

III. **Kleman, I**., Rosberg, A. K., Guzhva, O., Becher, P.G. & Mogren, L. Temporal changes in volatile organic compounds in post-harvest onion bulbs with Pectobacterium carotovorum-induced soft rot (manuscript)

The contribution of Isabella Kleman to the papers included in this thesis was as follows:

I. Investigation and method development together with LM, analysis and writing the original draft.

II. Conceptualisation and method development with PGB and LM, investigation, analysis and writing the original draft.

III. Conceptualisation and method development with PGB and LM, investigation, analysis and writing the original draft.

List of tables

Table 1: The indicator headspace volatile compounds for Fusarium oxysporum and Penicillium polonicum infection in onion bulbs. Adapted from Paper II. 43

Table 2: The indicator headspace volatile compounds for Pectobacterium carotovorum infection in onion bulbs. Adapted from Paper III. 46

List of figures

Figure 1: An onion bulb showing severe symptoms of *Fusarium* basal plate rot. 23

Figure 2: Examples of bulbs deemed to be sprouting (A) or not yet sprouting (B). Previously published in Paper I. 32

Figure 3: The setup used to sample headspace VOCs of onion bulbs using SPME. 34

Figure 4: Percentage of sampled onion bulbs showing internal sprout elongation in relation to measured HPLC peak areas of the sugars fructose, glucose and sucrose. Originally published in Paper I. 37

Figure 5: The change over time in storage of the HPLC peak area (arbitrary units), divided by milligrams of lyophilized onion extruded, of the peak separate from the individual sucrose, glucose and fructose peaks. This peak is presumed to contain fructans, among other water soluble components of the onion bulbs. Points represent average values for all samples of each respective month from start of storage in October 2019 (1) to end of storage in June 2020 (9), with error bars denoting standard deviation. 39

Figure 6: Heatmap of VOCs found in the headspace of control onion bulbs (C) and bulbs infected with *Pectobacterium carotovorum* (P) from 0 to 14 weeks post infection (WPI). Samples were taken and analysed using SPME-GC-MS equipped with a nonpolar HP5 GC column. Each sampling point represent average values for 3 bulbs. Onion furanone is synonymous with 2-

Hexyl-5-methyl-3(2H)-furanone and HA-2-EME is Hexanoic acid, 2-ethyl-, methyl ester. 44

Figure 7: Heatmap of VOCs found in the headspace of control onion bulbs (C) and bulbs infected with *Pectobacterium carotovorum* (P) from 0 to 14 weeks post infection (WPI). Samples were taken and analysed using SPME-GC-MS equipped with a polar DBWax GC column. Each sampling point represent average values for 3 bulbs. 45

Figure 8: The temporal changes in relative abundance of 2,3-butanediol and 3-hydroxy-2-butanone in headspace samples of onion bulbs infected with *Pectobacterium carotovorum*. Samples were taken from 0 to 14 weeks post infection and analysed using headspace SPME-GC-MS equipped with a polar DBWax GC column (n=3). Error bars represent standard deviations. 47

Figure 9: The temporal changes in relative abundance of 2,3-butanediol and 3-hydroxy-2-butanone in headspace samples of onion bulbs infected with *Pectobacterium carotovorum*. Samples were taken from 0 to 14 weeks post infection and analysed using headspace SPME-GC-MS equipped with a nonpolar HP5 GC column (A, n=3). Error bars represent standard deviations. 48

Abbreviations

AUC	Area under the curve
CVP	Crystal violet pectate medium
DNA	Deoxyribonucleic acid
FOS	Fructooligosaccharides
PCR	Polymerase chain reaction
GC-MS	Gas chromatography-mass spectrometry
HPLC	High performance liquid chromatography
NSC	Non-structural carbohydrates
PDA	Potato dextrose agar
PDB	Potato dextrose broth
RNA	Ribonucleic acid
SPME	Solid phase microextraction
VOC	Volatile organic compounds

1. Background

1.1 Introduction

The *Allium* genus contains several important horticultural crops, including garlic (*Allium sativum*), leek (*Allium ampeloprasum* var. *porrum*) spring onion (*Allium fistulosum*), chives (*Allium schoenoprasum*) and onion (*Allium cepa*). Of these crops, the onion is the most important economically, with over 100 million tons of onion bulbs harvested on a yearly basis worldwide (FAOSTAT 2025). Onions are used, primarily for their flavour, in cuisines all over the world. The onion follows a biennial lifecycle, growing from seed to bulb in its first season, and reproducing sexually the following year. To survive harsh conditions, the bulbs enter a dormant period, where growth is paused until conditions are more suitable. This trait is useful for human consumers, as it means bulbs may be stored for extended periods of time, allowing for the consumption of onions year round even in climates where the growing season is short. However, a number of different problems, including sprouting, rot and weight loss, often arise during storage, leading to significant losses (Anbukkarasi et al. 2013; Franke et al. 2013; Olanipekun 2018). Quality monitoring systems may help by improving storage planning and thereby reducing losses.

1.2 Onion physiology

1.2.1 Bulb formation

Bulb initiation in an onion plant requires certain threshold values of degree days and day length (Lancaster et al. 1996). The exact values required vary depending on cultivar, with cultivars requiring long days commonly used in

higher latitude regions, where days are longer during the growing season (Mashayekhi et al. 2022). In addition to these requirements, bulb initiation is affected several other factors, and occurs earlier in the presence of high plant densities and low red:far-red light ratios (Mondal et al. 1986a; b). Accelerated bulbing in highly competitive environments can be a way for the plant to enhance its survival by entering dormancy to await better conditions (Brewster 2008b). Bulb initiation can be described as having occurred at the first occurrence of leaf initial with a ratio between blade length and sheath length of less than 1 (Heath & Hollies 1965). This measurement often coincides with the more common definition of bulb initiation occurring at a "bulbing ratio" (maximum bulb diameter:minimum sheath diameter) greater than 2 (Brewster 2008b). As bulbing progresses, all new dry matter produced by the plant is stored in the storage scales and leaf growth is halted (Tei et al. 1996). Consequently, the size and number of leaves of the plant when bulb initiation occurs affects the size and number of scales of the finished bulb. The continued swelling of and dry matter transport to the storage scales eventually results in a mature bulb, at which point the leaf blades of the plant wilt.

1.2.2 Dormancy, non-structural carbohydrates and sprouting

As the bulb matures, a period of dormancy with little to no cell division activity and low respiration rates begins (Yasin & Bufler 2007). Dormancy can be defined as the period between cessation of scale formation and the initiation of sprout growth (Pak et al. 1995). Depending on the conditions the bulb is exposed to dormancy may last for a long period of time or, if the bulb is exposed to spring-like temperatures, break in just a couple of weeks (ibid.). A combination of sprouting inhibition treatment and temperature control can be utilised to enable long term storage of dormant bulbs. The dormancy can be artificially prolonged using treatments, such as maleic hydrazide application or irradiation that inhibit cell division in the bulb apical meristem and decrease respiration rate (Benkeblia et al. 2002). Application of maleic hydrazide is typically carried out before harvest, and the compound is transported from the leaves into the bulb where it can remain, inhibiting cell division, for a long period of time, thus prolonging dormancy (Ilic et al. 2011). Postharvest application has also been tested, and found to function satisfactorily (Benkeblia 2004). Through radiation damage, gamma

irradiation treatment similarly inhibits cell division activity and respiration rate in treated bulbs (Benkeblia et al. 2000).
One of the main purposes of the onion bulb is to store energy that can later be made available for growth and reproduction. Therefore, the majority, around 80 %, of a bulbs dry matter content consists of energy storing non-structural carbohydrates (NSCs) (Darbyshire & Henry 1979). In particular energy is stored as sucrose, glucose, fructose and for long-term storage, the polymeric fructooligosaccharides (FOS) or fructans (Brewster 2008a). Fructans are well-suited as long-term storage molecules as their presence, even at high concentrations, in the vacuole does not impair photosynthesis, (Vijn & Smeekens 1999). The amount of fructans contained in a bulb, and the rate at which they are hydrolysed are some of the main factors determining how long a bulb can remain dormant (Jaime et al. 2001).
As dormancy breaks, sprouting is initiated. Ahead of sprouting a redistribution of NSCs, especially fructans, takes place from the bulbs storage scales to the basal plate (Ohanenye et al. 2019). This facilitates the hydrolysis of stored fructans that enables respiration and subsequently growth. The rate of sprout elongation is dependent largely on temperature, with temperatures around 15 °C leading to faster growth than both lower and higher temperatures (Abdalla & Mann 1963), though the optimum temperature may vary depending on cultivar (Miedema 1994).

1.3 Storage of onions and postharvest quality problems

1.3.1 Harvest and pre storage treatment

While the wilting of leaf blades signifies full maturity of the bulbs, harvest often takes place before all bulbs in the field have reached this stage. Rather, the point at which the leaves have folded over on a certain percentage of the plants, a so called “tops down” percentage signifies the appropriate time for harvest. The percentage of tops down aimed for depends on the qualities desired in the crop. A moderately early lifting, around 50-90% tops down, results in bulbs that have a high number of intact dry scales, which is beneficial to storability and prevention of rots (Wright 1997; Wright et al. 2001). Lifting either too late or too early may cause an increased risk of storage rots, due to fewer protective dry scales and higher moisture retention in leaves, which promotes infection. As previously mentioned, maleic

hydrazide is normally applied to the leaves before bulbs are lifted, to allow absorption into the bulb, while gamma irradiation is carried out after field drying (Benkeblia & Varoquaux 2003). However, the use of maleic hydrazide has been questioned due to the potential risk of traces remaining in the bulbs that reach consumers and the effects of runoff into nature, and is disallowed in e.g. Estonia (Põldma et al. 2011). Gamma irradiation, while common in several parts of the world, is restricted in other parts (Stefanova et al. 2010). The use of other methods to inhibit sprouting, including ethylene or 1-methylcyclopropene treatment and controlled atmosphere has been tested with promising results (Downes et al. 2010; Põldma et al. 2011).

After lifting, bulbs must be dried, or "cured" in order to achieve the degree of dryness and closed necks necessary for good storability. The methods used to cure the bulbs vary. In some climates, leaving the bulbs to dry in the field may be sufficient, while other, moister climates may require active curing in a heated space (Downes et al. 2009). However, excessive curing may hasten the development of storage rots in cases where a pathogen is already present (Schroeder et al. 2012).

1.3.2 Storage conditions

Long term storage of onion requires appropriate conditions to minimise sprouting, transpiration and pathogen growth. As sprouting and catabolism is promoted by temperatures between 5 and 20 °C (Abdalla & Mann 1963), storage should be carried out either above or below these temperatures. Storage at higher temperatures, around 25-30 °C, will inhibit dormancy break and is often be the method of choice in warm climates where the cost of cooling storage facilities is prohibitive (Brewster 2008c). However, warm storage temperatures tend to promote pathogen growth (Schwartz & Mohan 2007) and are less effective at long term prevention of sprouting than temperatures near 0 °C (Tanaka et al., 1985, referenced in J. L. Brewster, 2008c). The relative humidity of the air in storage must also be controlled, as low humidity can accelerate weight loss due to transpiration. On the other hand, excessively high humidity and temperature differences between air and onions can cause condensation (Brewster 2008c), which promotes growth of certain pathogens (Schwartz & Mohan 2007). A high RH may also promote rooting and sprouting (Islam et al. 2015). Ideally, storage should be carried out at near, but not below freezing temperatures, and a RH around 70% (ibid.). Additionally, controlled atmosphere may be applied to further

improve storability of onion bulbs, with an atmosphere containing 3% CO_2 and 0.5-2% O_2 having been shown to reduce sprouting and rooting (Adamicki 2005).

1.3.3 Losses due to quality problems

While well suited to storage due to their biennial lifecycle and their ability to go dormant during the harsh conditions of winter, as metabolically active plant parts onion bulbs cannot be stored indefinitely. Losses occur due to a range of different quality problems arising during storage, including sprouting, pathogen infection and weight loss due to transpiration. While modern storage facilities where temperature, humidity and surrounding gas composition can be controlled can help reduce losses, they cannot fully prevent them. The amount of losses arising due to quality problems differ from case to case depending on cultivar, methods for cultivation and storage, injuries and disease pressure. An average of 13% lost during storage has been stated for Swedish conditions (Franke et al. 2013). For India, the world's biggest producer of onion, postharvest losses may be up to 35-40% (Anbukkarasi et al. 2013). Postharvest onion losses as high as 50% have been mentioned for Nigeria, which is another relatively large producer of onions (Olanipekun 2018; FAOSTAT 2025).

1.3.4 Sprouting

Sprouting is a major cause of storage losses (Adamicki 2005) as even the early stages, including root growth, are considered quality problems, lowering sales value. Visible shoot growth typically renders the bulb entirely unfit for sale (UNECE 2019), except for processing purposes. As sprouting is initiated the NSCs and water stored in the bulb are gradually depleted, furthering the storage losses by reducing bulb weight. As up to 90% of an onion bulb's dry matter content is made up of NSCs (Darbyshire & Henry 1979), the consumption of carbohydrates and transpiration over time will lead to a loss of bulb fresh weight as well as total dry mass. The loss of NSCs tends to be greater than the loss of water, resulting in the percentage of dry matter in a bulb decreasing over time in storage (Mogren et al. 2007).

1.3.5 Fungal diseases

A wide variety of fungi occur as pathogens in cultivation and postharvest storage of onions. Typically, infection that becomes obvious during storage will start toward the end of the growing season or be enabled by mechanical damage caused by harvest, topping and handling (Schwartz & Mohan 2007). It is also possible for fungal diseases to spread throughout a storage unit, as sporulation occurs in infected bulbs. Some common fungal causers of postharvest losses in onions grown in cooler temperate climates, such as northern Europe, include *Botrytis aclada* and *Botrytis allii* (neck rot), *Fusarium oxysporum* f.sp. *cepae* (*Fusarium* basal plate rot), *Penicillium* spp. (blue mould), *Peronospora destructor* (downy mildew, with associated bulb rot) and *Sclerotinia sclerotiorum* (white mould) (Schwartz & Mohan 2007). For the purposes of this thesis, the focus will be on *Fusarium oxysporum* f.sp. *cepae* and *Penicillium polonicum*.

Fusarium oxysporum is an ascomycete in the *Nectriaceae* family, with more than 100 formae speciales, many being specialized in infecting different genera and species of plants, while others are non-pathogenic (Edel-Hermann & Lecomte 2019). The species is a common soil inhabitant, with some strains also being capable of a saprophytic lifestyle, breaking down lignin and cellulose (Rodriguez et al. 1996). The main mode of reproduction and dispersal is through asexually produced soilborne macro- and microconidia as well as hardy chlamydospores that can survive adverse conditions (Schwartz & Mohan 2007). *Fusarium oxysporum* inocula may be present on seeds or in the soil (Southwood et al. 2015). Infection and symptoms can occur at any point from sprouting to postharvest storage, with fungal invasion either of undamaged tissue or facilitated by pre-existing wounds (Schwartz & Mohan 2007). *Fusarium* basal plate rot is a widespread and severe problem in stored onion bulbs, leading to significant losses (ibid.). The infection is characterized by a white mould on the basal plate surface and may eventually lead to the complete breakdown of the bulb, starting from the basal plate area (ibid., Figure 1).

Figure 1: An onion bulb showing severe symptoms of *Fusarium* basal plate rot.

Another common cause of storage losses is *Penicillium* infection. A number of different *Penicillium* spp. are known to infect onion bulbs, including *albocoremium*, *allii*, *citrinum*, *digitatum*, *expansum*, *funiculosum*, *glabrum, hirsutum*, *tulipae*, *radicicola*, *oxalicum*, *polonicum* and *venetum* (Overy et al. 2005; Vikram et al. 2005; Schwartz & Mohan 2007; Dugan et al. 2014; Çakır & Maden 2015; Duduk et al. 2017). The mode of infection and symptoms caused by the various species and strains are similar, though severity may vary, and the type of disease caused is typically referred to as blue mould. As the name implies, blue mould is characterized by bluish green conidia on the bulb surface or between the outer dry scales, with lesions penetrating deeper into the bulb. The pathogens are often present in soils or may persist in plant parts left in the field.

In general, while some fungi can grow at low temperatures, fungal infection and growth in onion bulbs are promoted by moderate to high temperatures, moisture and pre-existing damage.

1.3.6 Bacterial diseases

Another cause of many of the losses occurring during storage of onions is bacterial infections. Like in the case of fungi, there are a number of different bacterial species that commonly cause storage rots, including *Burkholderia cepacia* (sour skin), *Burkholderia gladioli* subsp. *alliicola* (slippery skin) and *Pectobacterium carotovorum* (soft rot, syn. *Erwinia carotovora*). Of the bacterial storage rots, soft rots caused by *P. carotovorum* and other closely related species are some of the more common and severe causes of postharvest losses in onion (Schwartz & Mohan 2007) as well as other stored crops, such as potato (Waterer & Pritchard 1984; Kushalappa et al. 2002). Therefore, *P. carotovorum* will be the main bacterial pathogen in the focus of this thesis.

Pectobacterium carotovorum is a gram-negative, rod-shaped and pectolytic bacteria in the *Pectobacteriaceae* family. It is capable of invading plant cells, weakening the cell wall using pectate lyases, and eventually killing the cell (Hall & Wood 1973; Hugouvieux-Cotte-Pattat et al. 2014). Due to this generalist mode of invasion *P. carotovorum* poses a threat to a wide variety of crop species, including bulbs, tubers, roots and aboveground plant parts (Schwartz & Mohan 2007; Dadaşoğlu & Kotan 2017; Meng et al. 2017; van der Wolf et al. 2021). In onions, symptoms of soft rot caused by *P. carotovorum* typically start near the neck or at a wound, gradually spreading through the bulb beginning with the central scales. Infection is promoted by warm, moist conditions, especially if these conditions are present at harvest (Schwartz & Mohan 2007).

1.4 Quality indicators for stored onion bulbs

1.4.1 Physiological indicators

As previously mentioned, there is an intimate connection between NSC or dry matter contents and onion bulb dormancy and sprouting. Dry matter contents also affect the usability of the bulbs for purposes such as frying (Hansen 1999b). Because of this, many studies have used NSC or dry matter contents or total soluble solids as indicators of onion bulb quality or storability (Rutherford & Whittle 1982; Sinclair et al. 1995; Hansen 1999a; Lancaster et al. 2001; Benkeblia et al. 2002, 2004; Benkeblia & Varoquaux 2003; Chope et al. 2006, 2012; Yasin & Bufler 2007; Downes et al. 2010;

Põldma et al. 2011; Nabi et al. 2013; Sharma et al. 2015; Petropoulos et al. 2016; Ohanenye et al. 2019; Golubkina et al. 2022). The activity of FOS-metabolising enzymes is another related factor that's been studied and found to be connected to dormancy break and the loss of FOS over time (Benkeblia et al. 2005). The rate of fructan hydrolysis and initial dry matter content in the bulbs has also been shown to be connected to storability, with dry matter contents above 16% being indicative of good storability (Jaime et al. 2001). Yet another NSC-related measurement that's been suggested as a predictor of sprouting is the ratio of monosaccharides:disaccharides, with bulbs that are actively sprouting containing a higher ratio (Chope et al. 2012). The connection between sprouting and concentrations of the phytohormones abscisic acid, zeatin riboside and isopentenyladenosine has also been investigated. It was found that high concentrations of abscisic acid at harvest may indicate better storability (Chope et al. 2006), while high concentrations of zeatin riboside indicates imminent sprouting (Chope et al. 2012). Other measurable quality indicators that are of interest to consumer acceptance and/or health, but are not necessarily connected to storability, include contents of flavonoids, phenolics and pyruvate (Wall & Corgan 1992; Mogren et al. 2006; Sharma et al. 2015). Firmness tends to decline with time in storage and bulbs with a high dry matter content tend to be firmer (Coolong et al. 2008), meaning firmness may be an indicator of current quality, but possibly not of storability.

1.4.2 The potential for volatile monitoring in crop storage

In the past two decades, systems for monitoring of VOCs using combinations of different types of gas sensors, arranged in so called gas sensor arrays or e-noses, have been developed and tested in various aspects of crop production and postharvest management (Seesaard et al. 2022). By their ability to continuously and non-invasively monitor the changes occurring in a stored crop, these systems offer advantages over more labour intensive methods for physiological measurements. But as headspace volatile compounds disperse through the air of a storage unit, the issue of pinpointing the source of the infection-related odours needs consideration. E-noses have been tested in lab conditions and found to be able to distinguish between healthy onion bulbs and bulbs infected with various storage diseases with relatively high accuracies. Examples of such studies include Li et al. (2011) and Konduru et al. (2015), who used e-noses to distinguish onion bulbs

without infection from ones exhibiting symptoms of sour skin or neck rot. Labanska et al. (2022) similarly tested an e-nose and found it could identify the extent of basal plate rot in groups of onions. E-noses can not only identify microbial infections, but also track quality aspects of non-infected crops, e.g. tomatoes (Messina et al. 2012). In order to select the most suitable and inexpensive combination sensors for detection of onion storage diseases, knowledge on the VOCs released into the headspace of infected onion bulbs is needed.

1.4.3 Headspace volatiles of onions and postharvest pathogens

S-Alkyl cysteine sulfoxides (ACSOs), when hydrolysed by alliinase, are the basis for the many sulfuric compounds responsible for the characteristic onion smell and taste (Randle & Lancaster 2002). The dominating ASCO in onion (*Allium cepa*) is trans-(+)-s-(1-propenyl)-cysteine sulfoxide or 1-PESCO (Brewster 2008a). Upon injury resulting in cell lysis the hydrolysis of ASCOs occur, resulting in a number of reactive sulfur-containing compounds, including the tear-inducing lachrymatory factor as well as thiosulfinates and sulfides (Randle & Lancaster 2002). These organosulfur compounds make up much of the headspace volatile profile of onion bulbs. Various sulfides, including dipropyl disulfide, methyl propyl disulfide and dimethyl disulfide tend to be among the most abundant compound released into the headspace of healthy onion bulbs, as has been repeatedly reported (Vikram et al. 2005; Li et al. 2011; Wang et al. 2016, 2019). Cut or otherwise injured bulbs release many of the same compounds, though the relative ratios of release may be altered (Machová et al. 2019).

Wang et al. (2019) investigated the release of volatiles by onions infected by *F. oxysporum* and found some notable compounds, of which release increased markedly in infected bulbs, including isoprene and 1-propanethiol. They also found that certain compounds were present only in samples from infected bulbs, including styrene and 3-methyl-1-butanol, both typical markers of fungal activity (Beck et al. 2008; Speckbacher et al. 2021). The volatiles released by *Penicillium* infected bulbs specifically have not been the subject of dedicated studies, but was included in a wider panel of pathogens by Vikram et al. (2005). However, the study did not identify any volatiles exclusive to *Penicillium* infection, and the relative mass ion abundance of detected compounds showed no great deviance from those of control bulbs, but the overall volatile profile enabled differentiation between

the pathogens. As *Pectobacterium carotovorum* (syn. *Erwinia carotovora*) is a threat to a number of different crop species, several studies have been carried out to identify headspace volatiles relevant to its detection in onion, potato and cabbage (Waterer & Pritchard 1984, 1985; Prithiviraj et al. 2004; Vikram et al. 2005; Yang et al. 2021; Tiwari et al. 2022). These studies found several VOCs to be indicators of soft rot, including dimethyl disulfide and 3-bromofuran in onion, and 2,3-butanediol, 3-hydroxy-2-butanone, ethanol and ethyl acetate in potato and cabbage.

2. Aim and objectives

The aim of this thesis is to find tools that can be used in order to monitor the quality of onion bulbs in storage and through this monitoring prevent losses.

In order to reach this aim the following objectives were set:

- Investigate the connection between the onset of sprouting and a selection of established physiological quality parameters, including dry matter content, firmness, and sucrose, fructose and glucose contents over two full storage seasons (Paper I).

- Compare the presence and relative abundance of individual volatile organic compounds in the headspace of onion bulbs infected with *Fusarium oxysporum* or *Penicillium polonicum* to those of uninfected control bulbs, to identify indicator compounds for fungal infection (Paper II).

- Study the temporal changes in volatile organic compounds in the headspace of onion bulbs infected with *Pectobacterium carotovorum*, in order to identify indicator compounds for early and later stages of soft rot (Paper III).

3. Method summary

This section is a summary of the methods used in the experiments that were part of the thesis work. For in depth descriptions, refer to the respective paper.

3.1 Plant material

All studies performed as part of this thesis utilized long day yellow onion bulbs intended for long term storage, commercially grown using conventional production methods in the Scania region of Sweden. Onions were harvested at full maturity and at the start of the experiments free from visible damage. For Paper I, the bulbs used were of the cultivars Hystore, Hytech and Saskia, and obtained directly from commercial storage once per month over a period of 9 months, from harvest until end of storage during two consecutive storage seasons (2019-2020 and 2020-2021). Five bulbs each a four replicates were used for each sample and month. For Paper II and III, onion bulbs were obtained from local grocery stores, but originated from the same commercial storage facility as the bulbs used for Paper I. Bulbs that were of an average size of 125 g (or 8 bulbs/kg) and labelled with the same sales batch numbers were chosen for these experiments.

3.2 Physiological measurements (Paper I)

3.2.1 Firmness and sprouting

After removal of all fully or partially dry scales, the top and basal plate of each bulb, a handheld penetrometer (6 mm pin, Digital Fruit Hardness Tester 41050, Step Systems GmbH, Germany) was used to measure the firmness at

three points along the equator of each bulb, letting the penetrometer rest on a table and pushing the onion bulb onto the pin protruding over the edge. After firmness measurement, all bulbs were halved along the vertical plane, and their sprouting status was judged as either sprouting (obvious internal sprouts) or not sprouting (no clear sprout differentiation), as is exemplified in Figure 1. The fraction of sprouting bulbs was calculated based on the number of sprouted bulbs out of the 5 bulbs in each replicate and sampling occasion.

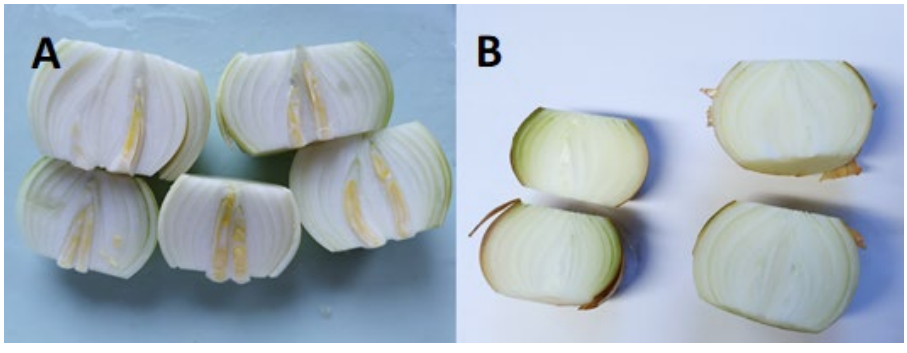

Figure 2: Examples of bulbs deemed to be sprouting (A) or not yet sprouting (B). Previously published in Paper I.

3.2.2 Sugar composition

Samples for HPLC analysis of glucose, fructose and sucrose contents were prepared by blending cut onion bulbs into a homogenous mash using a Waring blender. The resulting mash was frozen at -80°C, after which they were lyophilized for five days at -90°C using a Hetosicc CD 13-3 (Heto InterMed, Denmark). Dry samples were then ground into fine powders and sugars were extracted by adding 50 mg onion powder to 5 mL MilliQ filtered water after which the suspension was shaken overnight at 4°C. The suspensions were then centrifuged and the supernatant was collected to analyse sugar contents using high-performance liquid chromatography (HPLC) to separate the constituent sugars into separate chromatogram peaks. To identify the occurring peaks by retention time comparisons, standards of glucose, fructose and sucrose were also run.

3.2.3 Dry matter content

In order to determine the dry matter content of the bulbs, the onion mash left over from each sample after fractions were removed for sugar analysis was placed in an aluminium tray, weighed, and dried in a convection oven at 60°C for 7 days. After this the resulting dry mass was weighed again and the dry matter percentage was calculated by dividing dry weight by wet weight.

3.3 Pathogens (Paper II & III)

3.3.1 Fusarium oxysporum

Initially, attempts were made to inoculate bulbs with *Fusarium oxysporum* f.sp. *cepae* CBS 148.25 (obtained from Westerdijk FungalBio Diversity Institute, Netherlands). However, this strain failed to reliably produce symptoms of basal plate rot in bulbs kept at 20°C for 8 weeks after inoculation. Therefore, we isolated our own strain of *Fusarium oxysporum*, by scraping spores from bulbs with naturally occurring symptoms of *Fusarium* basal plate rot onto acidified potato dextrose agar (acidified PDA, or APDA) plates, and repeatedly culturing out the different colony phenotypes onto new plates, until pure cultures were obtained. DNA from these pure cultures was extracted and amplified using PCR with ribosomal RNA gene primers NL1, NL4, ITS1 and ITS4 for the fungi and ENV1 and ENV4 primers for bacteria. The amplified product was sent off for sequencing, and the results showed that sequences from one of the isolated strains were up to 99.46% similar to those found in previously sequenced strains of *Fusarium oxysporum*. DNA from other isolates could with comparable degrees of similarity be identified as *Clonostachys rosea*, *Fusarium redolens*, *Penicillium glabrum* and *Rahnella aquatilis*. The newly isolated strain of *Fusarium oxysporum* was chosen to infect bulbs to be used for sampling of headspace volatiles.

To prepare *F. oxysporum* infected bulbs for sampling of headspace volatiles, a spore suspension was created by scraping spores from a 14 day old culture of *F. oxysporum*, grown on PDA at 25°C, into sterile deionized water and straining the resulting suspension through several layers of sterile butter muslin until a spore suspension free from other particles was obtained. The spore density in the suspension was counted in a counting chamber, and adjusted to a concentration of 10^5 spores mL^{-1} using sterile deionized water.

A wound was made to a depth of 20 mm in the basal plate area of each bulb to be infected, using a sterile 4 mm injection needle, and 100 µL of spore suspension was applied to each wound and allowed to soak in for 1 hour. After this, bulbs were individually bagged in polyamide oven bags, sealed with a plastic bag clip, and incubated at 25°C for the course of the experiment. Control bulbs were left uninjured and not inoculated.

Figure 3: The setup used to sample headspace VOCs of onion bulbs using SPME.

3.3.2 Penicillium polonicum

Penicillium polonicum CBS 222.90 was purchased from Westerdijk FungalBio Diversity Institute and used to inoculate bulbs to be used for sampling of headspace volatiles, using the same methods as for the *Fusarium* inoculation, with the following modifications: Bulbs were not inoculated through the basal plate, but through wounds perpendicular to the equator, and control bulbs were similarly wounded, but inoculated only with sterile deionized water.

3.3.3 Pectobacterium carotovorum

Modified crystal violet pectate media (CVP) was prepared according to the recipe by Hyman et al. (2001) in order to enable detection of bacteria with pectolytic activity on reisolation. First attempts at infection with *Pectobacterium carotovorum* NCCB 37025 failed to produce symptoms in inoculated onion bulbs. This strain also grew very weakly, if at all, on CVP and did not produce the divots indicative of pectolytic activity on these plates. It was concluded that this strain lacked the desired pathogenicity. A new strain, *P. carotovorum* DSM 30168, was purchased from Leibniz Institute DSMZ (Braunschweig, Germany). This strain grew well on CVP and produced the characteristic divots around its colonies. It also showed a strong ability to cause soft rot in inoculated onion bulbs when preliminary test were carried out. Therefore, this strain was chosen for use to study the effect of soft rot on the headspace volatiles of onion bulbs.

Bulbs to be used for headspace volatile sampling were inoculated with 10^5 colony forming units (CFU) suspended in sterile 0.086% NaCl solution to a depth of 2 cm through the center of the neck of the bulb, and individually bagged before being incubated at 25°C for the duration of the experiment. Bulbs to be used as negative controls were left unwounded and uninoculated, but otherwise treated the same.

3.3.4 Other pathogens

In addition to the previously mentioned three pathogens used, infection attempts and volatile sampling was carried out using *Burkholderia cepacia*, the causal agent of sour skin in onions, as well as lung infections in vulnerable humans. However, despite the fact that the strain (DSM 7288) used was originally isolated from onions, it failed to reliably produce symptoms, and the data from these samplings were not processed further.

3.4 Analysis of headspace volatiles (Paper II & III)

Headspace samples were taken using solid phase microextraction (SPME) fibers coated in a blend of three adsorbents (Divinylbenzene/Carboxen/Poly-dimethylsiloxane or DVB/CAR/PDMS (gray-coded hub); Supelco, Bellefonte, PA). Fibers were exposed to the air inside each onion bag (Figure 1) for 45 minutes, after which volatiles were desorbed in a gas chromatograph inlet to be separated and tentatively identified using gas

chromatography-mass spectrometry (GC-MS). A polar DBWax column and a nonpolar HP5 column were used in all experiments, to identify compounds with a higher degree of certainty than would be possible using only a single type of column. To enable calculation of Kovats retention indices for the detected peaks, an alkane blend (C7-C30) was injected using the same methods as for the onion headspace samples, but adding a 6 minute solvent delay. Obtained peak areas were recalculated as a percentage of the total area under the chromatogram (% AUC), to enable comparisons between samples. Compound identification was carried out through comparison off mass spectra using library searches in NIST MSSearch 2.4, comparisons of obtained polar and nonpolar retention indices with those previously published, and for some compounds of interest by injection of synthetic standards. Peaks identified as compounds present in only one sample or in samples of empty oven bags or clean SPME fibers were included as basis for the % AUC calculations, but were removed from further analysis.

4. Results and discussion

4.1 Sugar, dry matter and firmness as indicators of sprouting

In the study of physiological indicators of sprouting status (Paper I), it was found that the contents of the sugars glucose, fructose and sucrose changed over time, and notably the contents of all measured sugars peaked between the 5th and 7th month in storage. This was also the point at which an average of 60-80% of the bulbs showed signs of internal sprout elongation. All three sugars showed statistically significant connections between measured peak area and degree of sprouting ($p<0.05$), and the trend was especially clear for glucose and fructose (Figure 1).

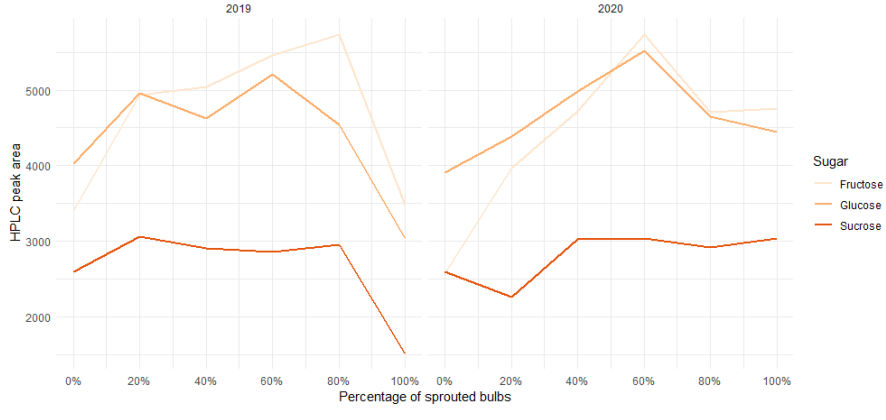

Figure 4: Percentage of sampled onion bulbs showing internal sprout elongation in relation to measured HPLC peak areas of the sugars fructose, glucose and sucrose. Originally published in Paper I.

.

The changing patterns of sugar concentration in the bulbs over time in storage and in connection to onset of sprouting matches observed in this study matches established knowledge on onion carbohydrate metabolism.
The storage and metabolism of NSCs as an energy source in the onion bulb is intimately connected to the bulbs function as a survival organ, as FOS are converted into simpler sugars such as sucrose, fructose and glucose, which can then be metabolised to enable the sprouting process. In addition to the three separate HPLC chromatogram peaks for sucrose, fructose and glucose, a fourth peak, present in all samples, could be seen decreasing in size as the storage season and sprouting progressed. For the 2019/2020 season the peak started at an average peak area of 2533±921 one month into storage, and decreased to an average peak area of 919±91 at the last sampling occasion, 8 months later (Figure 2). Though the method used does not allow us to ascertain the composition of compounds in this peak, it is reasonable to assume it at least partially consists of other water soluble NSCs, in particular fructans. As such, the steady decrease in peak area over time seems to show the decrease in content of fructans in the bulbs, as they are gradually hydrolysed to release the simpler sugars needed for respiration.

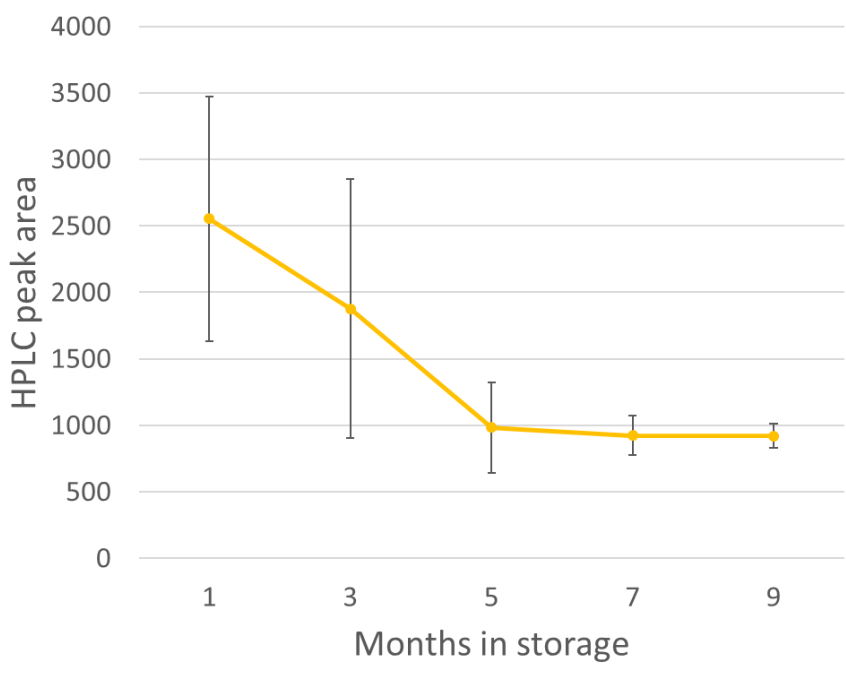

Figure 5: The change over time in storage of the HPLC peak area (arbitrary units), divided by milligrams of lyophilized onion extruded, of the peak separate from the individual sucrose, glucose and fructose peaks. This peak is presumed to contain fructans, among other water soluble components of the onion bulbs. Points represent average values for all samples of each respective month from start of storage in October 2019 (1) to end of storage in June 2020 (9), with error bars denoting standard deviation.

The results of the dry matter percentage measurements showed a similar decreasing trend over time, inversely correlating to the extent of sprouting (correlation of -0.390, $p<0.001$, 95% confidence interval -0.497; -0.270). The loss of dry matter was relatively similar between the different cultivars and seasons, with values ranging between 4.9 and 11.1% observed, with an average loss of 7.65%. The average initial dry matter contents in 2019 and 2020 were 10.6% and 11.4%, respectively. As rates of dry matter loss were similar, the samples that started with a high dry matter content also retained the highest contents until the end of the storage period. This is consistent with previous studies, where higher dry matter contents have been found to be indicators of good storability, as certain levels of NSCs are typically

required by processers and end users and the NSCs are depleted over time due to respiration (Hansen 1999a).
While firmness significantly declined over time when all samples were considered as a collective ($p<0.001$), no connection to degree of sprouting could be found. This was possibly caused by the somewhat uneven measurements obtained from the handheld penetrometer, as the method is quite sensitive to variations in handling by the person carrying out the measurements (English et al. 2022). Tukey pairwise comparisons between the total firmness changes for each sample type showed some grouping, but as a whole, the change in firmness for most samples overlapped.

Several of the measured mineral nutrient contents in the bulbs, measured as weight per kg dry matter, were found to significantly correlate with sprouting. However, this seems to merely be an artefact of the decreasing dry matter content correlating with sprouting while mineral contents remained relatively unchanged. Similar effects have been noticed in previous studies (Grevsen & Sorensen 2004).

4.2 Headspace volatiles of uninfected bulbs

The VOCs detected in the headspace of onion bulbs free from infection were consistent with what has been reported in previously published work, with dipropyl disulfide and methyl propyl disulfide being some of the main components of the volatile profile in solvent extracts (Kuo & Ho 1992). In the samples run on the polar GC column of control bulbs injected with sterile water and unwounded control bulbs (Paper II and III) dipropyl disulfide was the main compound in terms of percent of the total area under the curve (% AUC), with an average of 5.59% and 2.89% in samples of water-treated and unwounded bulbs, respectively. Similarly, methyl propyl disulfide had average % AUCs of 1.66% and 0.55%. In the unwounded control samples run on the nonpolar column, dipropyl disulfide and methyl propyl disulfide made up an average of 5.05% and 1.17% of the AUC. A total of 24 compounds were detected in the water-treated control samples. In the unwounded control, 16 compounds were detected using the GC-MS equipped with the polar column, and 22 were detected using the nonpolar column. The headspace samples of water-treated control in the *P. polonicum* experiment also contained minor amounts, with averages between 0.1% and

0.7% AUC, of phenol, 2-ethyl-1-hexanol, decanol, hexanal, octanal and decanal, 1-methylthio-1-propene, dimethyl disulfide, 1-allyl-2-isopropyldisulfane, 6-methyl-5-hepten-2-one, methyl 1-propenyl disulfide, 1-pentanol and 2,4-dimethyl-thiophene and 2-hexyl-5-methyl-3(2H)-furanone. Unwounded control bulbs released similar VOCs, but lacked phenol, 1-allyl-2-isopropyldisulfane, 1-pentanol and 2-hexyl-5-methyl-3(2H)-furanone. Propyl propenyl disulfide, 1-ethoxy-2-propanol and acetic acid were detected in unwounded control bulbs but not in the water-treated control. Many of the compounds occurred in only a minority of control samples. Some of the compounds occurring in a majority of unwounded control bulb samples include dipropyl disulfide, methyl propyl disulfide, propyl propenyl disulfide, 6-methyl-5-hepten-2-one and the previously mentioned aldehydes. The limited detection of sulfurous VOCs in unwounded bulbs in this study is likely because the release of various ASCO-derived volatiles is triggered by cell lysis (Randle & Lancaster 2002). Other studies, where a wider variety and higher relative abundances of sulfurous compounds were detected used sliced or mashed bulbs (Kuo & Ho 1992; Machová et al. 2019).

4.3 Headspace volatiles of bulbs with fungal infections

Infections with *P. polonicum* and *F. oxysporum* both resulted in changes to the headspace volatile profiles of the infected bulbs as compared to their respective uninfected control bulbs. A total of 23 compounds were detected in samples of *Penicillium* infected bulbs. As was the case with uninfected bulbs, dipropyl disulfide and methyl propyl disulfide were the two main compounds, but with relatively higher amounts than in the control, with 14.89% and 2.82% AUC, respectively. Multilevel pattern analysis comparing the relative abundances of compounds in control and *Penicillium* infected bulbs revealed five indicator compounds for *P. polonicum* infection, with 2,4-dimethyl-thiophene and 2-hexyl-5-methyl-3(2H)-furanone being the most consistent, both occurring in 19 out of 23 infected samples (Table 1). Both of these compounds were also present in certain of the water-treated control bulb samples, but at lower frequencies and relative abundances.

Infection with *F. oxysporum* created more distinct changes to the headspace volatile profile of the bulbs. Nine indicator compounds were found (Table 1), five of which were found in at least 23 out of 31 samples

and not at all in control samples; 1-(Methylthio)-propane, dimethyl disulfide, 2,2-bis(methylthio)-propane, ethenylbenzene and 1-(methylsulfinyl)-propane. Methyl propyl disulfide was indicated as an indicator compound of *Fusarium* infection, being detected in every infected sample, but only 2 out of 13 control samples. However, as previously stated, methyl propyl disulfide is known as a regularly occurring headspace volatile of healthy onions. It is possible that *Fusarium* infection raised the normal amounts of methyl propyl disulfide released above the detection threshold in this particular case.

Table 1: The indicator headspace volatile compounds for Fusarium oxysporum and Penicillium polonicum infection in onion bulbs. Adapted from Paper II.

Indicators for *Fusarium*	**CAS**	**Polar RI**	**Nonpolar RI**	**Occurs in F**	**Occurs in C**	**stat**	**p.value**
Methyl propyl disulfide	2179-60-4	1214.9	934.6	31/31	2/13	0.992	0.005
1-(Methylthio)-propane	3877-15-4	n/a	n/a	30/31	0/13	0.984	0.005
Dimethyl disulfide	624-92-0	1033	n/a	29/31	0/13	0.967	0.005
2,2-bis(methylthio)-propane	6156-18-9	1357	1036.8	29/31	0/13	0.967	0.005
Ethenylbenzene	100-42-5	1235.4	890.7	23/31	0/13	0.95	0.005
1-(methylsulfinyl)-propane	14094-08-7	1696	n/a	24/31	0/13	0.861	0.005
1-Octen-3-ol	3391-86-4	1427.4	978.6	19/31	3/13	0.715	0.02
2-(Methylsulfonyl)propane	4853-74-1	1948.1	n/a	11/31	0/13	0.568	0.045
Methyl 1-propenyl disulfide	5905-47-5	1272.4	n/a	7/31	0/13	0.539	0.04
Indicators for *Penicillium*							
2,4-Dimethyl-thiophene	638-00-6	1241.2	910.0	19/23	2/19	0.86	0.005
2-Hexyl-5-methyl-3(2H)-furanone	33922-66-6	2044.0	1445.9	19/23	9/19	0.849	0.005
Methyl 1-propenyl disulfide	5905-47-5	1279.1	942.4	13/23	5/19	0.663	0.02
1-Ethoxy-2-propanol	1569-02-4	1147.2	n/a	9/23	0/19	0.626	0.005
2-Methyl-5-pentanolide	n/a	1569.3	n/a	6/23	0/19	0.511	0.02

4.4 Headspace volatiles of bulbs with a bacterial infection

Pectobacterium carotovorum infection caused the increased relative abundance of several typical bacterial metabolites in the headspace of infected bulbs. A total of 29 recurring compounds were found in samples of *P. carotovorum*-infected bulbs analysed using GC-MS equipped with the nonpolar HP5 column (Figure 6), and 20 in samples analysed with GC-MS equipped with the polar DBWax GC column (Figure 7).

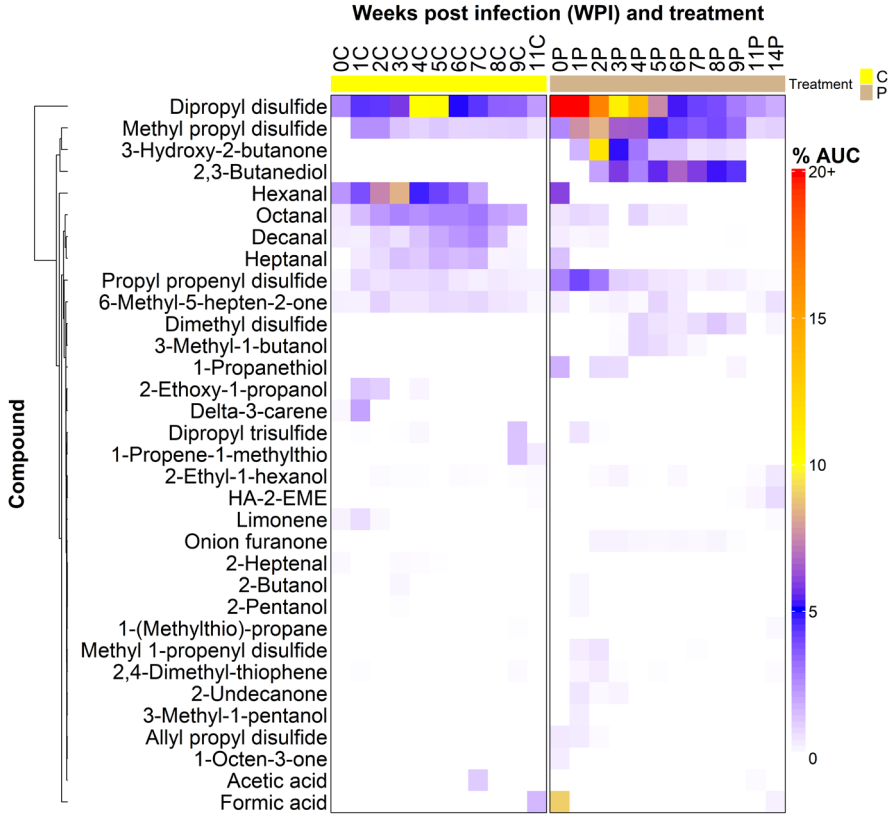

Figure 6: Heatmap of VOCs found in the headspace of control onion bulbs (C) and bulbs infected with *Pectobacterium carotovorum* (P) from 0 to 14 weeks post infection (WPI). Samples were taken and analysed using SPME-GC-MS equipped with a nonpolar HP5 GC column. Each sampling point represent average values for 3 bulbs. Onion furanone is synonymous with 2-Hexyl-5-methyl-3(2H)-furanone and HA-2-EME is Hexanoic acid, 2-ethyl-, methyl ester.

Among these were several indicator compounds found only in samples of infected bulbs, including 3-hydroxy-2-butanone, 2,3-butanediol, 3-methyl-1-butanol, dimethyl disulfide and ethanol (Table 2).

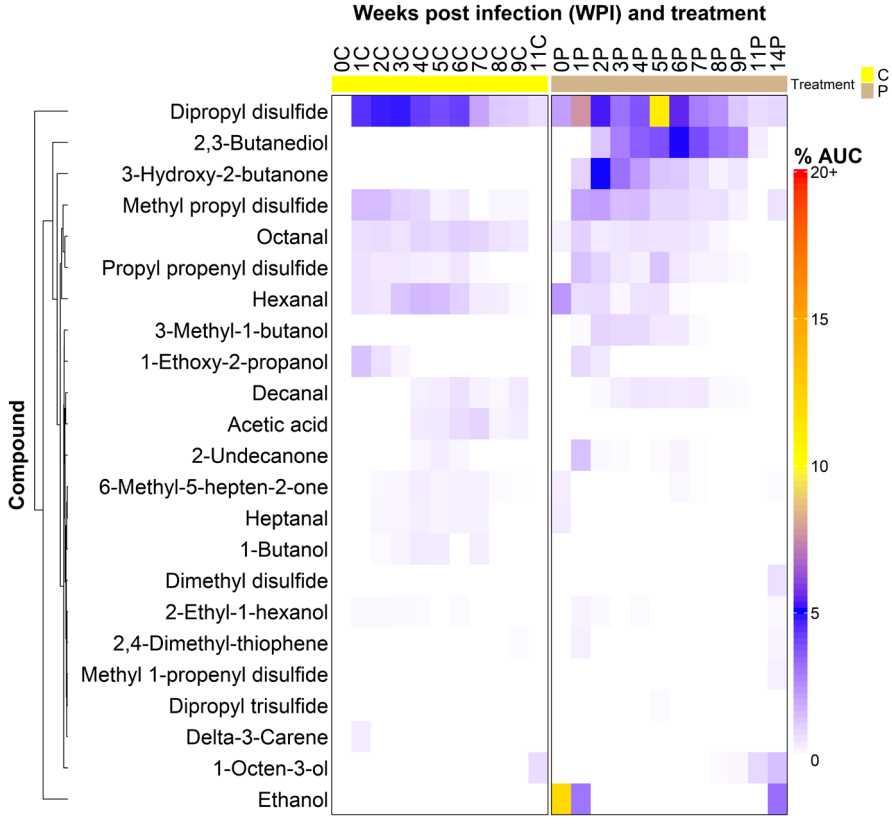

Figure 7: Heatmap of VOCs found in the headspace of control onion bulbs (C) and bulbs infected with *Pectobacterium carotovorum* (P) from 0 to 14 weeks post infection (WPI). Samples were taken and analysed using SPME-GC-MS equipped with a polar DBWax GC column. Each sampling point represent average values for 3 bulbs.

As 3-hydroxy-2-butanone and 2,3-butanediol are part of the butanediol fermentation found in Enterobacteriaceae (Blomqvist et al. 1993), including *Pectobacterium* spp., their presence in the headspace of infected bulbs is a clear indication of bacterial fermentation activity occurring in the bulbsThe headspace samples of infected bulbs over time showed high relative abundances of 3-hydroxy-2-butanone up to and including the 3 weeks post

infection point, after which it decreased, in favour of an increase in 2,3-butanediol (Figure 8 & 9).

Table 2: The indicator headspace volatile compounds for Pectobacterium carotovorum infection in onion bulbs. Adapted from Paper III.

Nonpolar column indicators	**CAS**	**HP5 RI**	**Occurs in P**	**Mean in P**	**Occurs in C**	**Mean in C**	**stat**	**p.value**
3-Hydroxy-2-butanone	513-86-0	n/a (RT 5.308 min)	26/35	2.97%	0/33		0.862	0.005
2,3-Butanediol	513-85-9	n/a (RT 5.455 min)	21/35	5.20%	0/33		0.775	0.005
2-Hexyl-5-methyl-3(2H)-furanone	33922-66-6	1448.1	20/35	0.22%	0/33		0.756	0.005
Dimethyl disulfide	624-92-0	n/a (RT 5.864 min)	12/35	1.14%	0/33		0.586	0.005
1-Propanethiol	107-03-9	n/a (RT 4.311)	10/35	1.03%	0/33		0.535	0.005
3-Methyl-1-butanol	123-51-3	n/a (RT 5.733 min)	12/35	0.61%	0/33		0.561	0.005
2-Undecanone	112-12-9	16.464	5/35	0.55%	1/33	0.25%	0.398	0.050

Polar column indicators	**CAS**	**DBWax RI**	**Occurs in P**	**Mean in P**	**Occurs in C**	**Mean in C**	**stat**	**p.value**
3-Hydroxy-2-butanone	513-86-0	1259.3	26/36	1.76%	0/32		0.819	0.005
2,3-Butanediol	513-85-9	1524.7	26/36	3.04%	0/32		0.794	0.005
3-Methyl-1-butanol	123-51-3	1182.5	17/36	0.67%	0/32		0.697	0.005
Ethanol	64-17-5	n/a (RT 5.675 min)	9/36	6.08%	0/32		0.507	0.005
1-Octen-3-ol	3391-86-4	1426.5	9/36	0.85%	2/32	0.74%	0.453	0.025

By week 11-14 post infection, toward the end of the sampling period, the relative abundance of the bacterial indicator compounds was reduced in

favour of an increased relative abundance of 1-octen-3-ol (Figure 7). This suggests an increased fungal activity in the infected bulbs, as the compound is a common fungal metabolite (Assaf et al. 1997), and has been indicated as a sign of fungal infection in onion bulbs (Paper II).

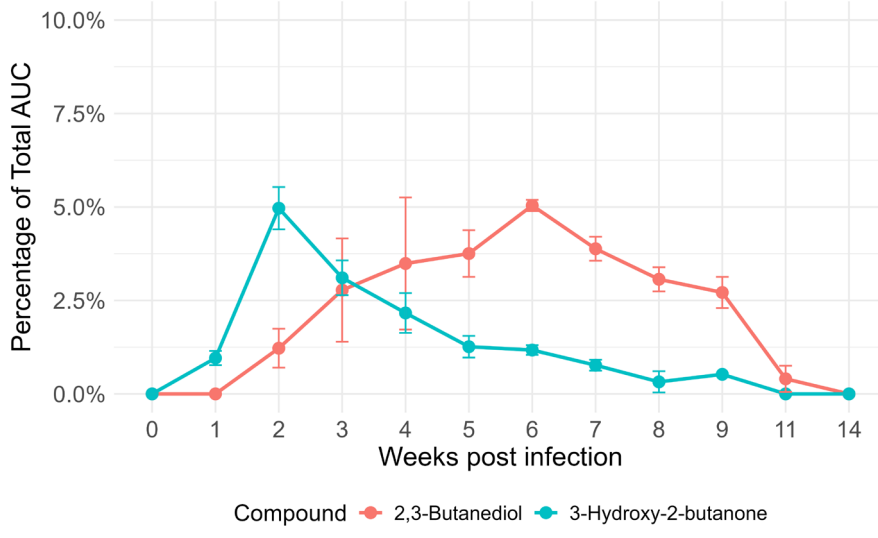

Figure 8: The temporal changes in relative abundance of 2,3-butanediol and 3-hydroxy-2-butanone in headspace samples of onion bulbs infected with *Pectobacterium carotovorum*. Samples were taken from 0 to 14 weeks post infection and analysed using headspace SPME-GC-MS equipped with a polar DBWax GC column (n=3). Error bars represent standard deviations.

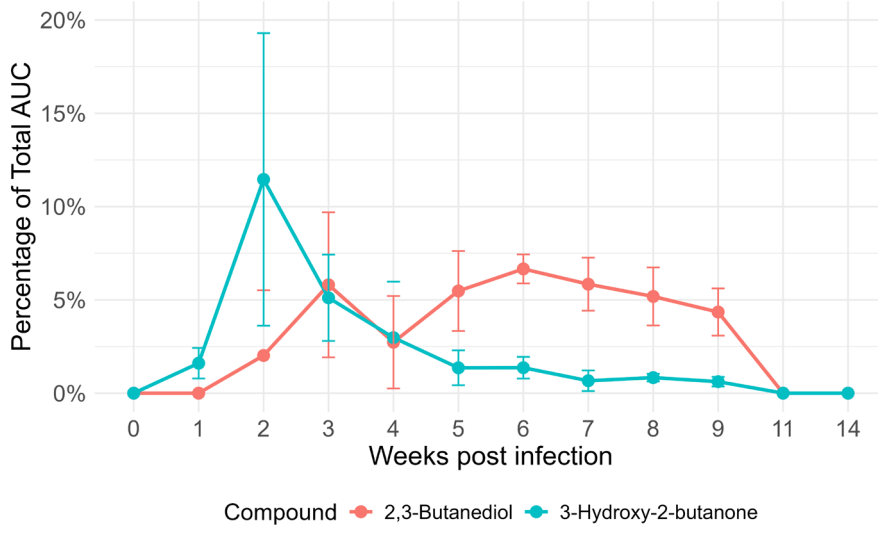

Figure 9: The temporal changes in relative abundance of 2,3-butanediol and 3-hydroxy-2-butanone in headspace samples of onion bulbs infected with *Pectobacterium carotovorum*. Samples were taken from 0 to 14 weeks post infection and analysed using headspace SPME-GC-MS equipped with a nonpolar HP5 GC column (A, n=3). Error bars represent standard deviations.

4.5 Practical applicability in storage

Various physiological measurements have been studied previously and show promise as indicators that predict sprouting. One such indicator is the changing ratio of mono- and disaccharides as sprouting is initiated and progresses. The connection between increased monosaccharide contents in the bulb and visible internal sprouting has been shown both in the study carried out as part of this thesis (Paper I) and in previously published studies (Chope et al. 2012). The contents of sucrose and glucose can be measured using a simple reflectometer and test strip system (Kleman et al. 2023). Dry matter content is also easy to measure and showed a connection to sprouting as dry matter was consumed at a higher rate than water in the sprouting process.

With regards to headspace VOC monitoring of *F. oxysporum*, *P. polonicum* and *P. carotovorum*-infection in storage, several indicator compounds were identified in this thesis. However, certain of the sulfuric compounds found to be indicators of fungal infection (Paper II) are naturally occurring also in healthy onions. These compounds include methyl propyl disulfide, dimethyl disulfide, 2,4-dimethyl-thiophene and methyl 1-propenyl disulfide (Bernhard 1969; Kuo & Ho 1992; Machová et al. 2019), though dimethyl disulfide can also be produced independently by bacteria (Wang et al. 2021). Despite being native to healthy onions, the changing abundance of sulfuric VOCs has been shown to be an important factor in identifying *F. oxysporum*-infected onion bulbs using an e-nose (Labanska et al. 2022). 2-Hexyl-5-methyl-3(2H)-furanone, while not necessarily unique to infected bulbs, is typically released in small amounts by healthy bulbs and in greater amounts by injured or otherwise stressed onion bulbs (Li et al. 2011) and may be an appropriate target for monitoring quality decline in stored bulbs, though the availability of sensors targeting this compound is unclear. Ethenylbenzene is a metabolite commonly found in fungi, including *F. oxysporum* (Beck et al. 2008), and is also one of the indicator compounds found in bulbs affected by *Fusarium* basal plate rot (Paper II). Ethenylbenzene is also of interest as it is hazardous to human health if inhaled, and consequently research into sensors capable of detecting its presence has been carried out (Liu et al. 2018). For *P. carotovorum*, several distinct compounds were found to be indicative of infection (Paper III). Among them, 3-hydroxy-2-butanone and ethanol were

among the more consistent indicators at the earliest stages of infection. Gas sensors have been tested and found to be capable of detect these two compounds (Park et al. 2017; Tian et al. 2020), though the specific sensors have yet to be tested in crop storage situations. Li et al. (2011) used a 32 sensor array to sample the headspace volatiles in a concentration chamber containing healthy onion bulbs or bulbs infected with either *Botrytis* neck rot or sour skin and found that it could successfully and with high accuracy determine which group of bulbs the sample came from. In a previous study, a gas sensor array with just 6 sensors coupled with a support vector machine, was successfully trained to detect sour skin with relatively high accuracy under similar conditions (Li et al. 2009). Konduru et al. (2015) achieved a similar classification accuracy (85%) of sour skin infected bulbs, sampled in small plastic containers with a custom gas sensor array. Another study found that the e-nose PEN 3 could identify and differentiate between different rates of *Fusarium* basal plate rot in batches of onion bulbs with high accuracy, and that while all sensors in the array contributed to differentiation the ones targeting organic sulfur compounds and nitrogen oxides were particularly important (Labanska et al. 2022). Further studies in real storage conditions in combination with improved knowledge on which VOCs to target, such as those found in Paper II and III, and thus which sensors to use, could further refine the system to produce effective and cost-efficient storage monitoring.

5. Conclusions and future perspectives

- Recurring measurements of mono- and disaccharide contents in stored onion bulbs can help predict sprouting and guide decisions on removal from storage and sale before losses are incurred.

- The exact sugar ratios or dry matter contents that indicate sprouting initiation will differ between cultivars and conditions, and further studies are needed to establish applicable guidelines for specific cases.

- VOC monitoring has promise as a method to detect quality changes, including development of storage diseases, but there is still room for fine-tuning in the selection of target compounds and thus sensors.

- Compounds relevant as targets for automated monitoring systems using gas sensor arrays include sulfur-containing compounds native to onions, ethenylbenzene and 1-octen-3-ol for fungal infections, and 3-hydroxy-2-butanone, 2,3-butanediol, 3-methyl-1-butanol and ethanol for *Pectobacterium* soft rot. 2-Hexyl-5-methyl-3(2H)-furanone may be a more general indicator of injury or infection, but the usability of this compound for monitoring requires further studies.

References

Abdalla, A.A. & Mann, L.K. (1963). Bulb development in the onion (Allium cepa L.) and the effect of storage temperature on bulb rest. *Hilgardia*, 35 (5), 85–112. https://doi.org/10.3733/hilg.v35n05p085

Adamicki, F. (2005). EFFECTS OF PRE-HARVEST TREATMENTS AND STORAGE CONDITIONS ON QUALITY AND SHELF-LIFE OF ONIONS. *Acta Horticulturae*, (688), 229–238. https://doi.org/10.17660/ActaHortic.2005.688.31

Anbukkarasi, V., Paramaguru, P., Pugalendhi, L., Ragupathi, N. & Jeyakumar, P. (2013). Studies on pre and post-harvest treatments for extending shelf life in onion – A review. *Agricultural Reviews*, 34 (4), 256. https://doi.org/10.5958/j.0976-0741.34.4.011

Assaf, S., Hadar, Y. & Dosoretz, C.G. (1997). 1-Octen-3-ol and 13-hydroperoxylinoleate are products of distinct pathways in the oxidative breakdown of linoleic acid by Pleurotus pulmonarius. *Enzyme and Microbial Technology*, 21 (7), 484–490. https://doi.org/10.1016/S0141-0229(97)00019-7

Beck, J.J., Merrill, G.B., Palumbo, J.D. & O'Keeffe, T.L. (2008). Strain of Fusarium oxysporum Isolated from Almond Hulls Produces Styrene and 7-Methyl-1,3,5-cyclooctatriene as the Principal Volatile Components. *Journal of Agricultural and Food Chemistry*, 56 (23), 11392–11398. https://doi.org/10.1021/jf802570w

Benkeblia, N. (2004). Effect of maleic hydrazide on respiratory parameters of stored onion bulbs (Allium cepa L.). *Brazilian Journal of Plant Physiology*, 16 (1), 47–52. https://doi.org/10.1590/S1677-04202004000100007

Benkeblia, N., Onodera, S. & Shiomi, N. (2004). Effect of gamma irradiation and temperature on fructans (fructo-oligosaccharides) of stored onion bulbs Allium cepa L. *Food Chemistry*, 87 (3), 377–382. https://doi.org/10.1016/j.foodchem.2003.12.010

Benkeblia, N., Ueno, K., Onodera, S. & Shiomi, N. (2005). Variation of Fructooligosaccharides and their Metabolizing Enzymes in Onion Bulb (Allium cepa L. cv. Tenshin) During Long-term Storage. *Journal of Food Science*, 70 (3), S208–S214. https://doi.org/10.1111/j.1365-2621.2005.tb07159.x

Benkeblia, N. & Varoquaux, P. (2003). Effects of gamma-irradiation, temperature and storage time on the status of the glucose, fructose

and sucrose in onion bulbs Allium cepa L. *International agrophysics*, 17 (1)

Benkeblia, N., Varoquaux, P., Gouble, B. & Selselet-Attou, G. (2000). Respiratory parameters of onion bulbs (Allium cepa) during storage. Effects of ionising radiation and temperature. *Journal of the Science of Food and Agriculture*, 80 (12), 1772–1778. https://doi.org/10.1002/1097-0010(20000915)80:12<1772::AID-JSFA700>3.0.CO;2-5

Benkeblia, N., Varoquaux, P., Shiomi, N. & Sakai, H. (2002). Storage technology of onion bulbs c.v. Rouge Amposta: effects of irradiation, maleic hydrazide and carbamate isopropyl, N-phenyl (CIP) on respiration rate and carbohydrates. *International Journal of Food Science & Technology*, 37 (2), 169–175. https://doi.org/10.1046/j.1365-2621.2002.00554.x

Bernhard, R.A. (1969). The sulfur components of Allium species as flavoring matter. *Qualitas Plantarum et Materiae Vegetabiles*, 18 (1), 72–84. https://doi.org/10.1007/BF01100215

Blomqvist, K., Nikkola, M., Lehtovaara, P., Suihko, M.-L., Airaksinen, U., Stråby, K.B., Knowles, J. & Penttilä, M.E. (1993). Characterization of the genes of the 2, 3-butanediol operons from Klebsiella terrigena and Enterobacter aerogenes. *Journal of Bacteriology*, 175 (5), 1392–1404

Brewster, J. (2008a). Biochemistry, health benefits and food science of alliums. In: J. L. Brewster (ed.) *Onions and other vegetable alliums*. CABI. 347–372. https://doi.org/10.1079/9781845933999.0347

Brewster, J. (2008b). Physiology of crop growth, development and yield. In: J. L. Brewster (ed.) *Onions and other vegetable alliums*. CABI. 85–170. https://doi.org/10.1079/9781845933999.0085

Brewster, J.L. (2008c). Crop storage and dormancy. In: *Onions and other vegetable alliums*. (Crop production science in horticulture; 15). 2. ed. CABI. 309–341.

Çakır, E. & Maden, S. (2015). First report of Penicillium polonicum causing storage rots of onion bulbs in Ankara province, Turkey. *New Disease Reports*, 32 (1), 24–24. https://doi.org/10.5197/j.2044-0588.2015.032.024

Chope, G.A., Cools, K., Hammond, J.P., Thompson, A.J. & Terry, L.A. (2012). Physiological, biochemical and transcriptional analysis of onion bulbs during storage. *Annals of Botany*, 109 (4), 819–831. https://doi.org/10.1093/aob/mcr318

Chope, G.A., Terry, L.A. & White, P.J. (2006). Effect of controlled atmosphere storage on abscisic acid concentration and other

biochemical attributes of onion bulbs. *Postharvest Biology and Technology*, 39 (3), 233–242. https://doi.org/10.1016/j.postharvbio.2005.10.010

Coolong, T.W., Randle, W.M. & Wicker, L. (2008). Structural and chemical differences in the cell wall regions in relation to scale firmness of three onion (Allium cepa L.) selections at harvest and during storage. *Journal of the Science of Food and Agriculture*, 88 (7), 1277–1286. https://doi.org/10.1002/jsfa.3219

Dadaşoğlu, F. & Kotan, R. (2017). Identification and characterization of Pectobacterium carotovorum. *J Anim Plant Sci*, 27 (2), 647–654

Darbyshire, B. & Henry, R.J. (1979). The association of fructans with high percentage dry weight in onion cultivars suitable for dehydrating. *Journal of the Science of Food and Agriculture*, 30 (11), 1035–1038. https://doi.org/10.1002/jsfa.2740301103

Downes, K., Chope, G.A. & Terry, L.A. (2009). Effect of curing at different temperatures on biochemical composition of onion (Allium cepa L.) skin from three freshly cured and cold stored UK-grown onion cultivars. *Postharvest Biology and Technology*, 54 (2), 80–86. https://doi.org/10.1016/j.postharvbio.2009.05.005

Downes, K., Chope, G.A. & Terry, L.A. (2010). Postharvest application of ethylene and 1-methylcyclopropene either before or after curing affects onion (Allium cepa L.) bulb quality during long term cold storage. *Postharvest Biology and Technology*, 55 (1), 36–44. https://doi.org/10.1016/j.postharvbio.2009.08.003

Duduk, N., Lazarević, M., Žebeljan, A., Vasić, M. & Vico, I. (2017). Blue mould decay of stored onion bulbs caused by Penicillium polonicum, P. glabrum and P. expansum. *Journal of Phytopathology*, 165 (10), 662–669. https://doi.org/10.1111/jph.12605

Dugan, F.M., Lupien, S.L., Vahling-Armstrong, C.M., Chastagner, G.A. & Schroeder, B.K. (2014). Host ranges of North American isolates of Penicillium causing blue mold of bulb crops. *Crop Protection*, 64, 129–136. https://doi.org/10.1016/j.cropro.2014.06.013

Edel-Hermann, V. & Lecomte, C. (2019). Current Status of Fusarium oxysporum Formae Speciales and Races. *Phytopathology*®, 109 (4), 512–530. https://doi.org/10.1094/PHYTO-08-18-0320-RVW

English, W., Ekelöf, J., Vancutsem, F., Leijdekkers, M., Kleuker, G. & Hoffmann, C.M. (2022). Method for in-field texture analysis of sugar beet roots using a handheld penetrometer. *Acta Agriculturae Scandinavica, Section B — Soil & Plant Science*, 72 (1), 623–634. https://doi.org/10.1080/09064710.2022.2042589

FAOSTAT (2025). *FAOSTAT*. *FAOSTAT Statistical Database*. http://www.fao.org/faostat/en/#data/QCL [2023-03-02]

Franke, U., Einarson, E., Andrésen, N., Svanes, E., Hartikainen, H. & Mogensen, L. (2013). *Kartläggning av matsvinnet i primärproduktionen*. Nordisk Ministerråd. (TemaNord; TemaNord 2013:581)

Golubkina, N., Amalfitano, C., Sekara, A., Tallarita, A., Pokluda, R., Stoleru, V., Cuciniello, A., Agafonov, A.F., Kalisz, A., Hamburdă, S.B. & Caruso, G. (2022). Yield and bulb quality of storage onion cultivars as affected by farming system and nitrogen dose. *Scientia Horticulturae*, 293, 110751. https://doi.org/10.1016/j.scienta.2021.110751

Grevsen, K. & Sorensen, J.N. (2004). Sprouting and yield in bulb onions (Allium cepa L.) as influenced by cultivar, plant establishment methods, maturity at harvest and storage conditions. *The Journal of Horticultural Science and Biotechnology*, 79 (6), 877–884

Hall, J.A. & Wood, R. (1973). The killing of plant cells by pectolytic enzymes. *Fungal pathogenicity and the plant's response*, 19–31

Hansen, S.L. (1999a). Content and Composition of Dry Matter in Onion (Allium cepa L.) as Influenced by Developmental Stage at Time of Harvest and Long-term Storage. *Acta Agriculturae Scandinavica, Section B — Soil & Plant Science*, 49 (2), 103–109. https://doi.org/10.1080/09064719950135614

Hansen, S.L. (1999b). Do changes in chemical composition during storage of raw onions (Allium cepa) influence the quality of deep fat fried onions? *SPECIAL PUBLICATION-ROYAL SOCIETY OF CHEMISTRY*, 229, 138–141

Heath, O.V.S. & Hollies, M.A. (1965). Studies in the Physiology of the Onion Plant: VI. A Sensitive Morphological Test For Bulbing And Its Use In Detecting Bulb Development In Sterile Culture. *Journal of Experimental Botany*, 16 (1), 128–144. https://doi.org/10.1093/jxb/16.1.128

Hugouvieux-Cotte-Pattat, N., Condemine, G. & Shevchik, V.E. (2014). Bacterial pectate lyases, structural and functional diversity. *Environmental Microbiology Reports*, 6 (5), 427–440. https://doi.org/10.1111/1758-2229.12166

Hyman, L.J., Sullivan, L., Toth, I.K. & Perombelon, M.C.M. (2001). Modified crystal violet pectate medium (CVP) based on a new polypectate source (Slendid) for the detection and isolation of soft rot erwinias. *Potato Research*, 44 (3), 265–270. https://doi.org/10.1007/BF02357904

Ilic, Z., Filipovic-Trajkovic, R., Lazić, S., Bursic, V. & Šunjka, D. (2011). Maleic hydrazide residues in the onion bulbs induce dormancy and hamper sprouting for long periods. *Journal of Food Agriculture and Environment*, 9, 113–118

Islam, M.N., Wang, A., Skov Pedersen, J. & Edelenbos, M. (2015). Microclimate tools to monitor quality changes in stored onions., 2015. 229–234

Jaime, L., Martín-Cabrejas, M.A., Mollá, E., López-Andréu, F.J. & Esteban, R.M. (2001). Effect of Storage on Fructan and Fructooligosaccharide of Onion *(Allium cepa* L.). *Journal of Agricultural and Food Chemistry*, 49 (2), 982–988. https://doi.org/10.1021/jf000921t

Kleman, I., HellstrÃ¶m, M., Rosberg, A.K., Becher, P.G. & Mogren, L. (2023). Simple quality prediction measurements for stored onions., April 30 2023. 183–186. International Society for Horticultural Science (ISHS), Leuven, Belgium. https://doi.org/10.17660/ActaHortic.2023.1364.24

Konduru, T., Rains, G.C. & Li, C. (2015). Detecting sour skin infected onions using a customized gas sensor array. *Journal of Food Engineering*, 160, 19–27. https://doi.org/10.1016/j.jfoodeng.2015.03.025

Kuo, M.C. & Ho, C.T. (1992). Volatile constituents of the solvent extracts of Welsh onions (Allium fistulosum L. variety maichuon) and scallions (A. fistulosum L. variety caespitosum). *Journal of agricultural and food chemistry*, 40 (10), 1906–1910

Kushalappa, A.C., Lui, L.H., Chen, C.R. & Lee, B. (2002). Volatile Fingerprinting (SPME-GC-FID) to Detect and Discriminate Diseases of Potato Tubers. *Plant Disease*, 86 (2), 131–137. https://doi.org/10.1094/PDIS.2002.86.2.131

Labanska, M., van Amsterdam, S., Jenkins, S., Clarkson, J.P. & Covington, J.A. (2022). Preliminary Studies on Detection of Fusarium Basal Rot Infection in Onions and Shallots Using Electronic Nose. *Sensors*, 22 (14). https://doi.org/10.3390/s22145453

Lancaster, J.E., Farrant, J. & Shaw, M.L. (2001). Sulfur Nutrition Affects Cellular Sulfur, Dry Weight Distribution, and Bulb Quality in Onion. *Journal of the American Society for Horticultural Science jashs*, 126 (2), 164–168. https://doi.org/10.21273/JASHS.126.2.164

Lancaster, J.E., Triggs, C.M., De Ruiter, J.M. & Gandar, P.W. (1996). Bulbing in Onions: Photoperiod and Temperature Requirements and Prediction of Bulb Size and Maturity. *Annals of Botany*, 78 (4), 423–430. https://doi.org/10.1006/anbo.1996.0138

Li, C., Gitaitis, R., Tollner, B., Sumner, P. & MacLean, D. (2009). Onion sour skin detection using a gas sensor array and support vector machine. *Sensing and Instrumentation for Food Quality and Safety*, 3 (4), 193–202. https://doi.org/10.1007/s11694-009-9085-1

Li, C., Schmidt, N.E. & Gitaitis, R. (2011). Detection of onion postharvest diseases by analyses of headspace volatiles using a gas sensor array and GC-MS. *LWT - Food Science and Technology*, 44 (4), 1019–1025. https://doi.org/10.1016/j.lwt.2010.11.036

Liu, B., Li, Y., Gao, L., Zhou, F. & Duan, G. (2018). Ultrafine Pt NPs-Decorated SnO2/α-Fe2O3 Hollow Nanospheres with Highly Enhanced Sensing Performances for Styrene. *Journal of Hazardous Materials*, 358, 355–365. https://doi.org/10.1016/j.jhazmat.2018.07.021

Machová, M., Bajer, T., Šilha, D., Ventura, K. & Bajerová, P. (2019). Release of volatile compounds from sliced onion analysed by gas chromatography coupled to mass spectrometry and its antimicrobial activity. *Journal of Food and Nutrition Research, volume 58, issue: 4*,

Mashayekhi, K., Kiani, Z., Mousavizadeh, S.J. & Zeinalinezhad, K. (2022). Optimizing sowing date for short- and long-day onion. *International Journal of Vegetable Science*, 28 (2), 132–143. https://doi.org/10.1080/19315260.2021.1909686

Meng, X., Chai, A., Shi, Y., Xie, X., Ma, Z. & Li, B. (2017). Emergence of Bacterial Soft Rot in Cucumber Caused by Pectobacterium carotovorum subsp. brasiliense in China. *Plant Disease*, 101 (2), 279–287. https://doi.org/10.1094/PDIS-05-16-0763-RE

Messina, V., Domínguez, P.G., Sancho, A.M., Walsöe de Reca, N., Carrari, F. & Grigioni, G. (2012). Tomato Quality during Short-Term Storage Assessed by Colour and Electronic Nose. *International Journal of Electrochemistry*, 2012 (1), 687429. https://doi.org/10.1155/2012/687429

Miedema, P. (1994). Bulb dormancy in onion. I. The effects of temperature and cultivar on sprouting and rooting. *Journal of Horticultural Science*, 69 (1), 29–39. https://doi.org/10.1080/14620316.1994.11515245

Mogren, L.M., Olsson, M.E. & Gertsson, U.E. (2006). Quercetin content in field-cured onions (Allium cepa L.): effects of cultivar, lifting time, and nitrogen fertilizer level. *Journal of agricultural and food chemistry*, 54 (17), 6185–6191

Mogren, L.M., Olsson, M.E. & Gertsson, U.E. (2007). Quercetin content in stored onions (Allium cepa L.): effects of storage conditions,

cultivar, lifting time and nitrogen fertiliser level. *Journal of the Science of Food and Agriculture*, 87 (8), 1595–1602

Mondal, M.F., Brewster, J.L., Morris, G.E.L. & Butler, H.A. (1986a). Bulb Development in Onion (Allium cepa L.) I. Effects of Plant Density and Sowing Date in Field Conditions. *Annals of Botany*, 58 (2), 187–195. https://doi.org/10.1093/oxfordjournals.aob.a087197

Mondal, M.F., Brewster, J.L., Morris, G.E.L. & Butler, H.A. (1986b). Bulb Development in Onion (Allium cepa L.) II. The Influence of Red: Far-red Spectral Ratio and of Photon Flux Density. *Annals of Botany*, 58 (2), 197–206. https://doi.org/10.1093/oxfordjournals.aob.a087198

Nabi, G., Rab, A., Bhutta, M., Farhatullah, D., Abbas, S. & Ali, A. (2013). Influence of curing methods and storage conditions on the post-harvest quality of onion bulbs. *Pakistan Journal of Botany*, 45, 455–460

Ohanenye, I.C., Alamar, M.C., Thompson, A.J. & Terry, L.A. (2019). Fructans redistribution prior to sprouting in stored onion bulbs is a potential marker for dormancy break. *Postharvest Biology and Technology*, 149, 221–234. https://doi.org/10.1016/j.postharvbio.2018.12.002

Olanipekun, C.I. (2018). Increasing production and reducing postharvest losses of onion in Nigeria. *World Vegetable Center*. https://avrdc.org/increasing-production-and-reducing-postharvest-losses-of-onion-in-nigeria/ [2023-07-03]

Overy, D.P., Frisvad, J.C., Steinmeier, U. & Thrane, U. (2005). Clarification of the agents causing blue mold storage rot upon various flower and vegetable bulbs: implications for mycotoxin contamination. *Postharvest Biology and Technology*, 35 (2), 217–221. https://doi.org/10.1016/j.postharvbio.2004.08.001

Pak, C., van der Plas, L.H.W. & de Boer, A.D. (1995). Importance of dormancy and sink strength in sprouting of onions (Allium cepa) during storage. *Physiologia Plantarum*, 94 (2), 277–283. https://doi.org/10.1111/j.1399-3054.1995.tb05312.x

Park, S.W., Lee, S.J., Sim, Y.S., Choi, J.Y., Park, E.Y. & Noh, B.S. (2017). Analysis of ethanol in soy sauce using electronic nose for halal food certification. *Food Science and Biotechnology*, 26 (2), 311–317. https://doi.org/10.1007/s10068-017-0042-1

Petropoulos, S.A., Fernandes, Ã., Barros, L., Barreira, J.C.M., Ferreira, I.C.F.R., Ntatsi, G. & Antoniadis, V. (2016). Effect of storage on quality features of local onion landrace 'Vatikiotiko'., October 28 2016. 125–132. International Society for Horticultural Science

(ISHS), Leuven, Belgium. https://doi.org/10.17660/ActaHortic.2016.1143.18
Põldma, P., Moor, U., Merivee, A. & Tõnutare, T. (2011). Effect of controlled atmosphere storage on storage life of onion and garlic cultivars., 2011. 63–69
Prithiviraj, B., Vikram, A., Kushalappa, A.C. & Yaylayan, V. (2004). Volatile Metabolite Profiling for the Discrimination of Onion Bulbs Infected by Erwinia carotovora ssp. carotovora, Fusarium oxysporum and Botrytis allii. *European Journal of Plant Pathology*, 110 (4), 371–377. https://doi.org/10.1023/B:EJPP.0000021058.81491.f8
Randle, W. & Lancaster, J. (2002). Sulphur compounds in alliums in relation to flavour quality. In: H. D. Rabinowitch, L.C. (ed.) *Allium crop science: recent advances*. CABI Publishing. 329–356. https://doi.org/10.1079/9780851995106.0329
Rodriguez, A., Perestelo, F., Carnicero, A., Regalado, V., Perez, R., de la Fuente, G. & Falcon, M.A. (1996). Degradation of natural lignins and lignocellulosic substrates by soil-inhabiting fungi imperfecti. *FEMS Microbiology Ecology*, 21 (3), 213–219. https://doi.org/10.1111/j.1574-6941.1996.tb00348.x
Rutherford, P.P. & Whittle, R. (1982). The Carbohydrate Composition of Onions During Long Term Cold Storage. *Journal of Horticultural Science*, 57 (3), 349–356. https://doi.org/10.1080/00221589.1982.11515063
Schroeder, B.K., Humann, J.L. & du Toit, L.J. (2012). Effects of Postharvest Onion Curing Parameters on the Development of Sour Skin and Slippery Skin in Storage. *Plant Disease*, 96 (10), 1548–1555. https://doi.org/10.1094/PDIS-02-12-0117-RE
Schwartz, H.F. & Mohan, S.K. (2007). *Compendium of Onion and Garlic Diseases and Pests, Second Edition*. The American Phytopathological Society. https://doi.org/10.1094/9780890545003
Seesaard, T., Goel, N., Kumar, M. & Wongchoosuk, C. (2022). Advances in gas sensors and electronic nose technologies for agricultural cycle applications. *Computers and Electronics in Agriculture*, 193, 106673. https://doi.org/10.1016/j.compag.2021.106673
Sharma, K., Asnin, L., Ko, E.Y., Lee, E.T. & Park, S.W. (2015). Phytochemical composition of onion during long-term storage. *Acta Agriculturae Scandinavica, Section B — Soil & Plant Science*, 65 (2), 150–160. https://doi.org/10.1080/09064710.2014.983151
Sinclair, P.J., Blakeney, A.B. & Barlow, E.W.R. (1995). Relationships between bulb dry matter content, soluble solids concentration and

non-structural carbohydrate composition in the onion (Allium cepa). *Journal of the Science of Food and Agriculture*, 69 (2), 203–209. https://doi.org/10.1002/jsfa.2740690210

Southwood, M.J., Viljoen, A. & McLeod, A. (2015). Inoculum sources of Fusarium oxysporum f.sp. cepae on onion in the Western Cape Province of South Africa. *Crop Protection*, 75, 88–95. https://doi.org/10.1016/j.cropro.2015.05.014

Speckbacher, V., Zeilinger, S., Zimmermann, S., Mayhew, C.A., Wiesenhofer, H. & Ruzsanyi, V. (2021). Monitoring the volatile language of fungi using gas chromatography-ion mobility spectrometry. *Analytical and Bioanalytical Chemistry*, 413 (11), 3055–3067. https://doi.org/10.1007/s00216-021-03242-6

Stefanova, R., Vasilev, N.V. & Spassov, S.L. (2010). Irradiation of food, current legislation framework, and detection of irradiated foods. *Food Analytical Methods*, 3, 225–252

Tanaka, M., Chee, K. & Komochi, S. (1985). Studies on the storage of autumn-harvested onion bulbs. I. Influence of storage temperature and humidity on sprouting during storage. *Research Bulletin of the Hokkaido National Agricultural Experiment Station*,

Tei, F., Scaife, A. & Aikman, D. (1996). Growth of lettuce, onion, and red beet. 1. Growth analysis, light interception, and radiation use efficiency. *Annals of Botany*, 78 (5), 633–643

Tian, H., Yu, B., Yu, H. & Chen, C. (2020). Evaluation of the synergistic olfactory effects of diacetyl, acetaldehyde, and acetoin in a yogurt matrix using odor threshold, aroma intensity, and electronic nose analyses. *Journal of Dairy Science*, 103 (9), 7957–7967. https://doi.org/10.3168/jds.2019-17495

Tiwari, S., Goswami, U., Kate, A., Modhera, B., Tripathi, M.K. & Mohapatra, D. (2022). Biological relevance of VOCs emanating from red onions infected with Erwinia (Pectobacterium) carotovora under different storage conditions. *Postharvest Biology and Technology*, 184, 111761. https://doi.org/10.1016/j.postharvbio.2021.111761

UNECE (2019). UNECE Standard FFV-25 concerning the marketing and commercial quality control of onions. UNITED NATIONS New York and Geneva. https://unece.org/fileadmin/DAM/trade/agr/standard/fresh/FFV-Std/English/25_Onions.pdf [2021-09-12]

Vijn, I. & Smeekens, S. (1999). Fructan: More Than a Reserve Carbohydrate?1. *Plant Physiology*, 120 (2), 351–360. https://doi.org/10.1104/pp.120.2.351

Vikram, A., Hamzehzarghani, H. & Kushalappa, A.C. (2005). Volatile metabolites from the headspace of onion bulbs inoculated with postharvest pathogens as a tool for disease discrimination. *Canadian Journal of Plant Pathology*, 27 (2), 194–203. https://doi.org/10.1080/07060660509507216

Wall, M.M. & Corgan, J.N. (1992). Relationship between Pyruvate Analysis and Flavor Perception for Onion Pungency Determination. *HortScience HortSci*, 27 (9), 1029–1030. https://doi.org/10.21273/HORTSCI.27.9.1029

Wang, A., Casadei, F., Johansen, A., Bukman, H. & Edelenbos, M. (2016). Emission of volatile organic compounds from healthy and diseased onions. *Acta Horticulturae*, (1144), 333–340. https://doi.org/10.17660/ActaHortic.2016.1144.49

Wang, A., Islam, Md.N., Johansen, A., Haapalainen, M., Latvala, S. & Edelenbos, M. (2019). Pathogenic Fusarium oxysporum f. sp. cepae growing inside onion bulbs emits volatile organic compounds that correlate with the extent of infection. *Postharvest Biology and Technology*, 152, 19–28. https://doi.org/10.1016/j.postharvbio.2019.02.010

Wang, W., Kang, S. & Vikesland, P.J. (2021). Surface-Enhanced Raman Spectroscopy of Bacterial Metabolites for Bacterial Growth Monitoring and Diagnosis of Viral Infection. *Environmental Science & Technology*, 55 (13), 9119–9128. https://doi.org/10.1021/acs.est.1c02552

Waterer, D.R. & Pritchard, M.K. (1984). Monitoring of volatiles: A technique for detection of soft rot (Erwinia carotovora) in potato tubers. *Canadian Journal of Plant Pathology*, 6 (2), 165–171. https://doi.org/10.1080/07060668409501578

Waterer, D.R. & Pritchard, M.K. (1985). Production of volatile metabolites in potatoes infected by Erwinia carotovora var. carotovora and E. carotovora var. atroseptica. *Canadian Journal of Plant Pathology*, 7 (1), 47–51. https://doi.org/10.1080/07060668509501513

van der Wolf, J.M., Acuña, I., De Boer, S.H., Brurberg, M.B., Cahill, G., Charkowski, A.O., Coutinho, T., Davey, T., Dees, M.W., Degefu, Y., Dupuis, B., Elphinstone, J.G., Fan, J., Fazelisanagri, E., Fleming, T., Gerayeli, N., Gorshkov, V., Helias, V., le Hingrat, Y., Johnson, S.B., Keiser, A., Kellenberger, I., Li, X. (Sean), Lojkowska, E., Martin, R., Perminow, J.I., Petrova, O., Motyka-Pomagruk, A., Rossmann, S., Schaerer, S., Sledz, W., Toth, I.K., Tsror, L., van der Waals, J.E., de Werra, P. & Yedidia, I. (2021). Diseases Caused by Pectobacterium and Dickeya Species Around the World. In: Van

Gijsegem, F., van der Wolf, J.M., & Toth, I.K. (eds) *Plant Diseases Caused by Dickeya and Pectobacterium Species*. Springer International Publishing. 215–261. https://doi.org/10.1007/978-3-030-61459-1_7

Wright, P.J. (1997). Effects of cultural practices at harvest on onion bulb quality and incidence of rots in storage. *New Zealand Journal of Crop and Horticultural Science*, 25 (4), 353–358. https://doi.org/10.1080/01140671.1997.9514026

Wright, P.J., Grant, D.G. & Triggs, C.M. (2001). Effects of onion (*Allium cepa*) plant maturity at harvest and method of topping on bulb quality and incidence of rots in storage. *New Zealand Journal of Crop and Horticultural Science*, 29 (2), 85–91. https://doi.org/10.1080/01140671.2001.9514166

Yang, J.-S., Lee, H.-W., Song, H. & Ha, J.-H. (2021). Volatile Metabolic Markers for Monitoring *Pectobacterium carotovorum* subsp. *carotovorum* Using Headspace Solid-Phase Microextraction Coupled with Gas Chromatography-Mass Spectrometry. *Journal of Microbiology and Biotechnology*, 31 (1), 70–78. https://doi.org/10.4014/jmb.2009.09028

Yasin, H.J. & Bufler, G. (2007). Dormancy and sprouting in onion (*Allium cepa* L.) bulbs. I. Changes in carbohydrate metabolism. *The Journal of Horticultural Science and Biotechnology*, 82 (1), 89–96. https://doi.org/10.1080/14620316.2007.11512203

Popular science summary

Onion is a vegetable consumed in large quantities in many parts of the world. To ensure year-round availability, onions often need to be stored, sometimes for many months. Since the onion bulb is the plant's way of storing energy for later use, it is well-adapted for storage. However, losses still occur during storage due to onion bulbs being affected by various rots or beginning to sprout. To reduce these losses, different monitoring systems need to be developed. Such systems could measure various changes occurring within the bulbs and enable better planning for sales and storage. This could be achieved either by removing a small number of bulbs to measure different properties or by using technology to detect, for instance, the odours emitted by the bulbs.

This thesis investigated how the measurement of various sugars and dry matter content in onions can be used to predict when sprouting will begin. It was found that the levels of glucose, fructose, and sucrose, as well as the decrease in dry matter, are linked to the onset of sprouting. The thesis also examined which odours are released when onions are infected by various types of rot. The odours emitted by onions affected by one of two different types of mould or a bacterial rot were analysed chemically. It became clear that the different types of mould and rot lead to the emission of distinct odour molecules.

The information from the first part of the study can be used in future development of sugar content sampling to predict sprouting. The results from the odour analyses can be applied in the development of electronic sensor systems capable of detecting early stages of rot during storage. This would make it possible to remove onions from storage before the rot spreads to more of the stored bulbs. Overall, the development of such systems can help to reduce losses during storage.

Populärvetenskaplig sammanfattning

Lök är en grönsak som konsumeras i stora mängder i många delar av världen. För att löken ska kunna finnas tillgänglig året om måste den i de flesta fall lagras, ibland i många månader. Eftersom löken är lökplantans sätt att spara energi till senare är den väl anpassad för att kunna lagras. Men ändå uppstår svinn under lagringen, på grund av att de lagrade lökarna drabbas av olika rötor eller börjar bilda blad. För att minska svinnet behöver olika övervakningssystem utvecklas. Sådana övervakningssystem skulle kunna mäta olika förändringar som sker inom lökarna och göra att man kan planera försäljning och lagring. Det kan fungera antingen genom att man tar ut provlökar och mäter olika egenskaper hos dem, eller genom teknologi som mäter exempelvis dofter som lökarna avger.

I denna avhandling undersöktes hur mätning av bland annat olika sockerarter och torrsubstans i lökar kan användas för att förutsäga när löken kommer att börja bilda blad. Det visade sig att innehållet av glukos, fruktos och sukros och minskningen av torrsubstans kan kopplas till början av bladbildning. I avhandlingen undersöktes också vilka dofter som avges när lökar smittas av olika rötor. Doften från lökar angripna av en av två olika typer av mögel eller en bakterieröta undersöktes genom kemisk analys. Det blev tydligt att de olika typerna av mögel och röta ger upphov till att olika sorters doftmolekyler avges. Informationen från det första delförsöket kan komma till användning i framtida utveckling provtagning av sockerhalt för att förutse bladbildning. Resultaten från doftanalyserna kan användas vid utvecklingen av elektroniska sensorsystem, som kan upptäcka tidiga stadier av röta under lagringen. På så vis kan man ta hand om löken innan rötan spridit sig till fler av de lagrade lökarna. Sammantaget kan utvecklingen av sådana system hjälpa till att minska svinn under lagring.

Acknowledgements

To my supervisors, **Lars**, **Paul**, **Anna Karin** and **Oleksiy**, thank you for all your guidance, support and feedback throughout this project.

Maria, thanks for all the support, friendship, help in the lab, study sessions, cat catching, adventures throughout Skåne and abroad, and so much more.

Helene, thank you for the conversations, guidance on all things PhD, the many walks – with or without bird watching, and the afternoon table tennis sessions.

Thanks to **Ida** and **Jojo** for the friendship, board game nights and horsey escapades, and also to the rest of hortonommaffian, **Camilla** and **Erika**.

The people from Berzan and elsewhere (including, but not limited to, **Claudia, Linnea, Isak, Joakim, Fredrik, Alice** and **Erik)**, who I get to see at New Years, Lincon, and spontaneous board game nights, getting together with you is always a highlight of coming home.

Thanks to **Anna** for always coming to the rescue when anything GC-related went south.

To **Guillermo** and **Charles**, thanks for the good times in the SWD group and for making me realise I wanted to do a PhD.

Thanks to the people of the **PhD council**, **DN** and **FUN**, it's been a fun learning experience to take part in council and board work.

I would also like to thank everyone who's been a member of the **MicroHort** group through the years, everyone else at **BT**, especially the **BT PhD students**, it's been nice to meet and talk at our various fikas, writing days and lunches.

Thanks also to my dogs, Ester and Leia, and cats, Boris, Sickan and Soja.

And, of course, I would like to thank my **family**, for always being there, welcoming me home anytime, and taking care of my pets whenever I attended a conference abroad.

Finally, to whomever else it may concern, tack och förlåt :)

I

Acta Agriculturae Scandinavica, Section B — Soil & Plant Science

ISSN: (Print) (Online) Journal homepage: www.tandfonline.com/journals/sagb20

Sugar content and dry matter are key factors predicting sprouting of yellow bulb onions regardless of treatment with maleic hydrazide

Isabella Kleman, Anna Karin Rosberg & Lars Mogren

To cite this article: Isabella Kleman, Anna Karin Rosberg & Lars Mogren (2024) Sugar content and dry matter are key factors predicting sprouting of yellow bulb onions regardless of treatment with maleic hydrazide, Acta Agriculturae Scandinavica, Section B — Soil & Plant Science, 74:1, 2366171, DOI: 10.1080/09064710.2024.2366171

To link to this article: https://doi.org/10.1080/09064710.2024.2366171

Published online: 27 Jun 2024.

ACTA AGRICULTURAE SCANDINAVICA, SECTION B — SOIL & PLANT SCIENCE
2024, VOL. 74, NO. 1, 2366171
https://doi.org/10.1080/09064710.2024.2366171

Sugar content and dry matter are key factors predicting sprouting of yellow bulb onions regardless of treatment with maleic hydrazide

Isabella Kleman, Anna Karin Rosberg and Lars Mogren

Department of Biosystems and Technology, Swedish University of Agricultural Sciences, Alnarp, Sweden

ABSTRACT

Onions are produced and consumed in large amounts all over the world. Even though the dry onion bulb is well suited to storage, significant losses occur due to sprouting and diseases during the storage period. The objective of this study was to find methods to support decision-making in storage and prevent some of these losses. Three onion cultivars were tested during two storage seasons, and quality indicators such as firmness, sugar content, dry matter content and mineral content were measured. All but one of the samples were treated with maleic hydrazide for sprouting inhibition. Contents of fructose and glucose were found to be connected to the extent of sprouting, with the highest contents coinciding with the onset of sprouting in spring. Firmness and dry matter losses differed between samples from different growing conditions with firmness losses up to 35.2% and dry matter losses between 4.9% and 11.1% found. Dry matter content was significantly connected to the fraction of sprouted bulbs in a sample. While firmness had a decreasing trend for all but one sample, the firmness measurements carried out with a handheld penetrometer were not consistent enough to be a reliable indicator of sprouting.

ARTICLE HISTORY
Received 20 March 2024
Accepted 5 June 2024

KEYWORDS
Dormancy; postharvest quality; sprouting; sugar; waste reduction

Introduction

As stated both by the Sustainable Development Goals of the United Nations, specifically goal 12, target 12.3 (UN General Assembly 2015) and the Recommendations for Action in Food Waste Prevention produced by the European Union (EU Platform on Food Losses and Food Waste 2019), reducing losses and waste of food is an important step towards sustainability. The bulb onion, *Allium cepa* L., is often stored for several months after harvest and during storage it suffers losses. These losses are caused by a variety of factors including storage diseases, caused by a number of different fungal and bacterial pathogens (Schwartz and Mohan 2007), sprouting and related weight loss due to transpiration and respiration consuming stored carbohydrates (Jaime et al. 2001). As stated in the UNECE standard for onions, later stages of sprouting, where visible leaf growth has occurred, render the bulbs unsellable (UNECE 2019). The amount of postharvest losses on a national level vary depending on cultivation and storage circumstances, and examples of losses as low as 13% (Franke et al. 2013) and as high as 50% (Olanipekun 2018) have been reported. With a worldwide production of over 100 million tons each year, onions are one of the most produced and consumed vegetable crops, second only to tomatoes (FAOSTAT 2023). Sprouting can be inhibited using chemical control; one common method is application of maleic hydrazide before harvest, which inhibits cell division in the bulb meristem (Greulach and Atchison 1950). Exposing the bulbs to ethylene or 1-methylcyclopropene is another method that has been shown to inhibit sprouting (Downes et al. 2010). Ionising irradiation is a third method that is used in many places (Sharma et al. 2020). The methods available to a grower depend on regional laws and regulations, as not all countries allow the use of maleic hydrazide (Põldma et al. 2011) or irradiation (Stefanova et al. 2010). Preventing development of storage diseases is more challenging. Ozone application has been found to reduce growth of fungal and bacterial causers of storage diseases (Shelake et al. 2022). The effect of air temperature and humidity during storage has been studied by many; it has been found that temperatures of 0–5°C inhibit both sprouting and disease development, while temperatures of 25–30°C inhibit sprouting but may promote

CONTACT Isabella Kleman isabella.kleman@slu.se Department of Biosystems and Technology, Swedish University of Agricultural Sciences, Alnarp, Sweden

disease development (Abdalla and Mann 1963; Islam et al. 2015; Petropoulos et al. 2017) A RH of 55–70% is beneficial as it helps limit disease and root development while also preventing excessive fresh weight loss (Gubb and MacTavish 2002). However, no method can guarantee complete long-term prevention of sprouting or disease. Therefore, there is a need for tools that help predict quality decline and support decision making regarding which batches are suitable for long-term storage.

Factors influencing the storage potential of a bulb are affected to varying degrees both by the genetics of the particular cultivar and by cultivation and handling. Approximately 80–90% of the onion bulb's dry matter content consists of non-structural carbohydrates such as fructans, sucrose, fructose and glucose (Darbyshire and Henry 1979). Contents of nonstructural carbohydrates change during the storage season and these changes are connected to the sprouting process (Downes et al. 2010). The content of fructans and the rate of fructan hydrolysis has been shown to be connected to onion bulb storability (Jaime et al. 2001). Onions containing high amounts of fructans and with low rates of fructan hydrolysis can be stored for longer periods of time before sprouting. The changing ratio between monosaccharides and disaccharides over the course of a storage period has also been pointed out as a potential predictor of sprouting (Chope et al. 2012). Additionally, bulbs with high firmness typically also have higher dry matter content, making firmness another indicator of storability (Coolong et al. 2008). A third factor influencing onion quality and storability is the content of mineral nutrients in the bulb, which is affected by fertiliser application. It has been shown that reduced doses of nitrogen fertiliser application can result in better storability (Golubkina et al. 2022). Increased doses of sulphur fertilisation can improve bulb growth, firmness and sulfur content, but no effect on total soluble solids or storability has been found (Lancaster et al. 2001; Forney et al. 2010).

Numerous studies have been performed over the years in an attempt to elucidate the effect of cultivar choice, cultivation methods and storage technology on onion quality and losses, and many have delved into the finer details of the bulbs' carbohydrate metabolism and enzymatic activity, and how this connects to the sprouting process (e.g. Pak et al. 1995; Carter et al. 1999; Ko et al. 2002; Benkeblia et al. 2005; Yasin and Bufler 2007; Chope et al. 2012; Sharma et al. 2015; Golubkina et al. 2022). In this grower initiated study we aim to look into how some of these well studied aspects of onion physiology could be of use for storage planning in commercial onion production under Nordic conditions. In order to do so, we worked in collaboration with a large scale commercial onion producer and took monthly samples of onions of three cultivars in their storage over the course of two whole storage seasons. The cultivars chosen were 'Hystore', 'Hytech' and 'Saskia', three commonly grown very long day cultivars with similar traits, all well adapted to Nordic conditions and suited to long term storage.

The objective of this study was to compare measurements of onion quality indicators with sprouting, with the goal of identifying methods that could be easily applied in storage facilities. In this study, we used a combination of easily applicable methods and well-developed scientific methods. The quality indicators measured were firmness, sugar composition, dry matter and mineral content. We hypothesise that these indicators are connected to sprouting status and can be used to predict storability of onions.

Materials and methods

Plant material

The onions were produced by commercial growers in the region of Skåne, Sweden, in 2019 and 2020. Harvested onion bulbs were kept in a storage facility operated by Almhaga Grönsaker AB (Höllviken, Sweden). The three cultivars, Hystore (HYS), Hytech (HYT) and Saskia (SAS), were harvested in September and sampled once a month during storage, from October until June. To obtain four replicates, bulbs were collected from squares placed at least 40 m apart in each field. For each sampling occasion, samples consisting of five random average sized (diameter 65–80 mm) onion bulbs without visible defects were taken from each replicate batch. In 2019, each cultivar was grown in two different fields for a total of six sample IDs, and in 2020, HYS and SAS were grown in two fields each while HYT was grown in three fields, for a total of seven sample IDs (Table 1). Before harvest, all batches except HYT20k were treated with full dose maleic hydrazide (Fazor, Arysta LifeScience Great Britain Ltd, United

Table 1. A list of the onion cultivars used and the ID each batch was given.

Cultivar	ID	
	2019	2020
Hystore	HYS19a	HYS20g
	HYS19b	HYS20h
Hytech	HYT19c	HYT20i
	HYT19d	HYT20j
		HYT20k
Saskia	SAS19e	SAS20l
	SAS19f	SAS20m

Each ID represents a batch of onions from a certain year (2019 or 2020) and a certain field (a-m). All samples except HYT20k were treated with full dose maleic hydrazide (Fazor, Arysta LifeScience Great Britain Ltd, United Kingdom) at 80% tops down.

Table 2. The sowing dates for the sampled onions and the pH, content of clay, sand and organic matter in soil samples from the fields where the onions were grown.

ID	Sowing date	Lifting date	pH	*P*-AL mg/100 g DW	K-AL mg/100 g DW	Mg-AL mg/100 g DW	K/Mg	Ca-AL mg/100 g DW	Clay	Sand	Organic matter	Classification
2019												
HYS19a	17-Apr	02-Sep	7.6	14	7.2	12	0.6	310	17%	53%	2.5%	Sandy loam
HYS19b	17-Apr	08-Sep	6.6	17	6.7	5	1.3	100	15%	65%	1.9%	Sandy loam
HYT19c	08-Apr	28-Aug	7.6	11	11	9.5	1.2	291	12%	66%	2.5%	Sandy loam
HYT19d	21-Apr	15-Sep	7.0	9.1	10	11	0.9	350	12%	67%	3.1%	Sandy loam
SAS19e	12-Apr	06-Sep	8.4	17	17	11	1.5	390	20%	60%	2.3%	Sandy clay loam
SAS19f	09-Apr	07-Sep	6.6	13	4.6	7.3	0.6	410	11%	70%	0.2%	Sandy loam
2020												
HYS20g	08-Apr	07-Sep	8.1	26	9.7	21	0.5	540	13%	66%	3.9%	Sandy loam
HYS20h	20-Apr	15-Sep	6.3	6.8	8.6	7.8	1.1	250	11%	79%	2.1%	Sandy loam
HYT20i	06-Apr	02-Sep	6.5	11	9.8	11	0.9	270	13%	60%	3.5%	Sandy loam
HYT20j	08-Apr	03-Sep	7.5	8.2	10	8.2	1.2	320	17%	54%	2.4%	Sandy loam
HYT20k	06-Apr	05-Sep	8.0	7.5	7.1	15	0.5	1000	17%	47%	4.2%	Loam
SAS20l	10-Apr	16-Sep	7.5	5.3	9	6.1	1.6	250	15%	62%	2.3%	Sandy loam
SAS20m	28-Apr	20-Sep	6.1	3.8	8	6.2	1.3	170	17%	60%	2.2%	Sandy loam

Values are given for soil dry weight.

Kingdom) at 80% tops down. Bulbs were lifted in August or September (Table 2) and allowed to dry in field until September 25th in 2019 or until October 5th in 2020. After field drying, the bulbs were dried for an additional four weeks in trays, protected from rain by a roof but otherwise exposed to ambient conditions, before being placed in refrigerated storage. The refrigerated storage was kept between 0 and 1°C with a RH of 80 to 85%. Onions intended for the experiment were kept separate from the bulbs meant for commercial sale, but in identical storage conditions. In 2019, grower reported losses, defined as the fraction of stored bulbs not suitable for retail sale, were 19% (HYS19a), 17% (HYS19b), 7% (HYT19c), 22% (HYT19d), 6.5% (SAS19e) and 7% (SAS19f). In 2020 the losses were 7% (HYS20 g), 9% (HYS20 h), 7% (HYT20i), 100% (HYT20j), 8% (HYT20k), 9% (SAS20 l) and 100% (SAS20 m). The 100% losses recorded for HYT20j and SAS20 m were due to the whole batches being sold to industrial processing at the end of storage as a large percentage of the bulbs had sprouted.

Field data

Soil samples were taken from the fields during the respective growing seasons (18 September 2019 and 15 August 2020), fractions of clay, sand and organic matter were measured and ammonium lactate (AL) analyses for mineral contents were performed (LMI, Helsingborg, Sweden). Results are shown in Table 2.

Sample preparations and firmness measurements

The bulbs were rinsed with tap water, and the top and basal plate and all fully or partially dry scales were removed. Bulb firmness was measured at three points along each bulb's equator using a handheld penetrometer (6 mm pin, Digital Fruit Hardness Tester 41050, Step Systems GmbH, Germany). For better uniformity in measurements, the penetrometer was laid flat on a table with the measuring pin sticking out and held still while the bulb was pushed onto the pin. After this, the bulbs were cut into quarters and sprouting status was noted. Bulbs where internal sprout elongation had started and the sprouts had visibly changed colour were deemed to be sprouting (Figure 1). The quartered bulbs were turned into a fine mash using a Waring blender (Model 91-358, USA). For sugar content analysis, onion mash was placed in a 50 mL polypropylene tube (Sarstedt, Germany), weighed and frozen at −80 °C. The samples were then lyophilised at −90°C using a Hetosicc CD 13-3 (Heto InterMed, Denmark) for five days, and weighed again.

Sugar analysis

To analyse the contents of sucrose, glucose and fructose using HPLC, sugars were extracted from the samples by adding 5 mL MilliQ filtered water to 50 mg of finely ground lyophilised onion sample and shaking overnight

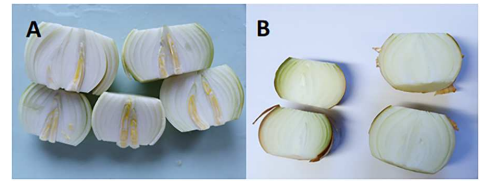

Figure 1. Examples of bulbs deemed to be sprouting (A) and not yet sprouting (B).

at 120 rpm and 4°C. Samples were then centrifuged for five minutes at 18000 G and 4 °C, and the supernatant was collected for analysis. For the samples collected in October 2020 and June 2021, a Phenomenex LUNA omega column and a mobile phase consisting of 75% acetonitrile was used. For all other samples, a Sarasep Car-H column was used with a mobile phase consisting of water and 3 mM HCl. Standards containing D-(+)-saccharose (VWR, USA), D-(+)-glucose (Merck, Germany) and D-(−)-fructose (Sigma-Aldrich, USA) at known concentrations were run using the same methods to ensure consistent measurements.

Mineral and dry matter content

After samples for flavonoid and sugar analysis had been collected, samples to be sent for mineral content analysis were prepared from the remaining onion mash. Each replicate was poured into an aluminium tray and weighed before and after being fully dried in an oven at 60 °C for seven days. The dry matter content was recorded. In order to study the mineral content dynamics, the dried samples from the storage season starting in 2019, months October, February and June, were analysed regarding a plant nutrient status panel of 17 minerals (Eurofins Agro Testing Sweden AB, Kristianstad, Sweden).

Statistics

Statistical analysis was carried out using Minitab version 19.2020.1 (Minitab, LLC) and R version 4.1.2. The correlation between mineral and dry matter content was calculated using Pearson pairwise correlation in Minitab. The connections between sprouting, mineral content and sugar content were calculated using the Anova function in R. A general mineral model was applied to the sprouting, sugar and mineral data in Minitab. Tukey pairwise comparisons were made for average sprouting and mineral content, and a main effects plot of the sugar content and sprouting was created using Minitab.

Ethics statement

This study did not involve human or animal subjects and therefore ethical approval was not necessary.

Results

Firmness

Firmness, as measured with the penetrometer, decreased significantly over time ($p<0.001$). There was a large variability between individual measurements, and no significant correlation with sprouting could be found. The total firmness decline varied somewhat, but it did not significantly differ between most sample IDs (Table 3).

Sprouting

The first signs of sprouting were observed in January of both years, with an average of 26% of the bulbs showing signs of sprouting from January to February. By March, 72% of the bulbs showed signs of sprouting, increasing to 92-99% in the period April-June. Slight differences between sample IDs could be seen, but the only significant difference found when IDs were compared using the Tukey method was that the group HYT20k, HYT20j and HYS19a had a higher percentage of sprouted bulbs than HYT19d (Table 4). The fact that HYT20k was not treated with maleic hydrazide did not have a significant effect on the sprouting status of the tested bulbs.

Sugars and dry matter content

The HPLC analysis of sugar concentration throughout the storage seasons showed that concentrations of the sugars fructose, glucose and sucrose were higher around mid-storage season, coinciding with the start of the internally visible sprouting process (Figure 2).

Table 3. The average firmness of onion bulbs of the cultivars 'Hystore' (HYS), 'Hytech' (HYT) and 'Saskia' (SAS) harvested in 2019 (19) or 2020 (20) from 13 fields (a-m) in Skåne, Sweden.

Months after harvest				
Sample ID	1 (N)	9 (N)	Mean change	Grouping
2019				
HYS19a	21.2±3.3	14.7±2.1	−30.9%	CD
HYS19b	20.6±2.2	16.8±2.4	−18.6%	ABCD
HYT19c	27.0±3.7	17.5±4.2	−35.2%	D
HYT19d	20.9±1.8	15.6±3.6	−24.9%	BCD
SAS19e	21.7±4.2	17.4±4.2	−19.1%	ABCD
SAS19f	23.7±2.5	18.6±3.6	−21.7%	ABCD
2020				
HYS20g	23.3±4.6	18.6±2.5	−19.5%	ABCD
HYS20h	21.0±3.3	18.0±3.2	−13.3%	ABCD
HYT20i	19.9±4.2	18.6±2.5	−5.9%	AB
HYT20j	21.7±3.4	16.9±4.8	−21.5%	ABCD
HYT20k	19.2±3.2	17.0±2.7	−10.8%	ABC
SAS20l	21.8±3.1	19.8±5.0	−8.0%	ABC
SAS20m	18.1±6.0	18.1±3.8	1.6%	A

All samples except HYT20k were treated with full dose maleic hydrazide (Fazor, Arysta LifeScience Great Britain Ltd, United Kingdom) at 80% tops down. Four replicates consisting of five random bulbs of each ID were sampled from storage every month from October (one month after harvest) until June (nine months after harvest) with the exception of November 2020, when no samples were taken. Firmness is given in Newton (N) ± SD, as measured with a handheld penetrometer with a pin diameter of 6 mm. Measurements shown were taken at the start (October) and end (June) of storage, with three measurements taken per bulb. Grouping information was acquired using Tukey pairwise comparisons and 95% confidence.

Table 4. Sprouting in onion bulbs of the cultivars 'Hystore' (HYS), 'Hytech' (HYT) and 'Saskia' (SAS) harvested in 2019 (19) or 2020 (20) from 13 fields (a-m) in Skåne, Sweden.

ID	N	Sprouted	Grouping
HYT20k	32	52.5%	A
HYT20j	32	50.0%	A
HYS19a	36	50.0%	A
HYT20i	32	48.2%	AB
SAS19f	36	47.8%	AB
HYS20h	32	46.9%	AB
SAS19e	36	46.7%	AB
SAS20m	32	45.0%	AB
HYS19b	36	44.4%	AB
HYT19c	36	43.9%	AB
SAS20l	32	43.8%	AB
HYS20g	32	40.7%	AB
HYT19d	36	35.6%	B

All samples except HYT20k were treated with full dose maleic hydrazide (Fazor, Arysta LifeScience Great Britain Ltd, United Kingdom) at 80% tops down. Four replicates consisting of five random bulbs of each ID were sampled from storage every month from October until June, with the exception of November 2020, when no samples were taken, and the resulting 32–36 values were pooled to calculate the average fraction of sprouting bulbs. Sprouting was defined as visible elongation and colour change of internal sprouts, as could be seen when bulbs were halved (Figure 1). Grouping information was acquired using Tukey pairwise comparisons and 95% confidence.

This trend was particularly clear with fructose and glucose (Figure 3(A, B)), which showed a marked increase coinciding with many of the bulbs starting their internal sprout growth. The monosaccharide content tended to peak at the time when 60–80% of the bulbs had sprouted, after which content decreased. The sucrose content did not peak in the same way. As sprouting progressed further (Figure 2), the fructose levels dropped off to near the levels found at the start of each storage season. Fructose was found to be significantly connected with sprouting status, both as a standalone factor ($p<0.05$) and when taking sampling month and sample ID into account ($p<0.01$). A significant connection was found between glucose content (Figure 3(B)), month, and sprouting ($p<0.01$). Sucrose content (Figure 3(C)) was significantly connected with sprouting on its own ($p<0.01$) and in connection to month ($p<0.05$), but not when sample ID was taken into account. No differences in average peak area could be found between samples run with the two different HPLC columns and solvents, and data have therefore been treated as part of the same set. Dry matter content varied between sample IDs at the beginning of the season and decreased at similar rates after that point (Table 5). Dry matter had a significant correlation of −0.390 with sprouting ($p<0.001$, 95% confidence interval −0.497; −0.270). On average, dry matter contents were higher in 2020 compared to 2019.

Mineral content

The concentration of certain minerals in the bulb dry weight was found to be significantly connected to the sprouting status. Concentrations of nitrogen ($p < 0.001$), sulphur ($p < 0.01$), calcium ($p < 0.001$) and potassium ($p < 0.01$) were significantly connected to the extent of sprouting when taking the sampling month into account. Additionally, sulphur content was significantly connected with sprouting when taking both the sampling month and sample ID into account ($p < 0.05$). Contents of calcium ($p < 0.05$) and chlorine ($p < 0.01$)

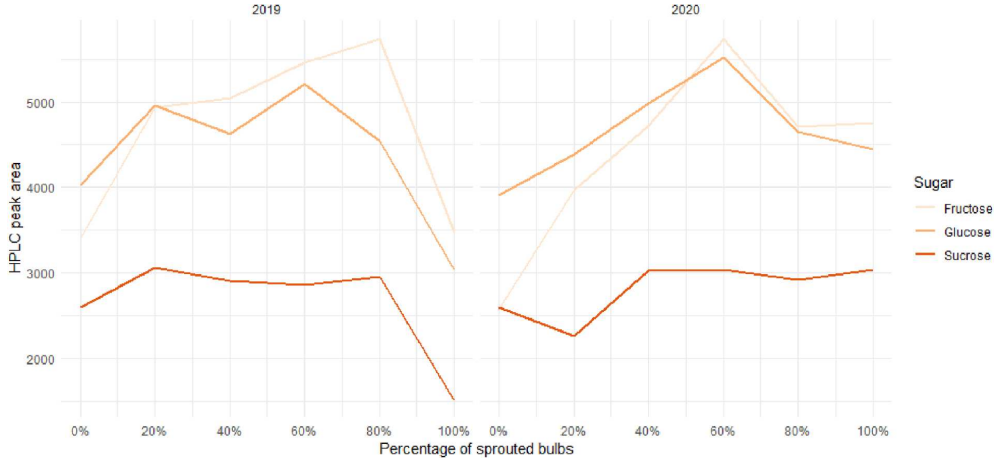

Figure 2. The relation between the percentage of sprouted onion (*Allium cepa* L.) bulbs and the contents of fructose, glucose and sucrose (expressed as HPLC peak area). Bulbs were harvested in 2019 and 2020, and sugar content was measured once a month from October until June using HPLC.

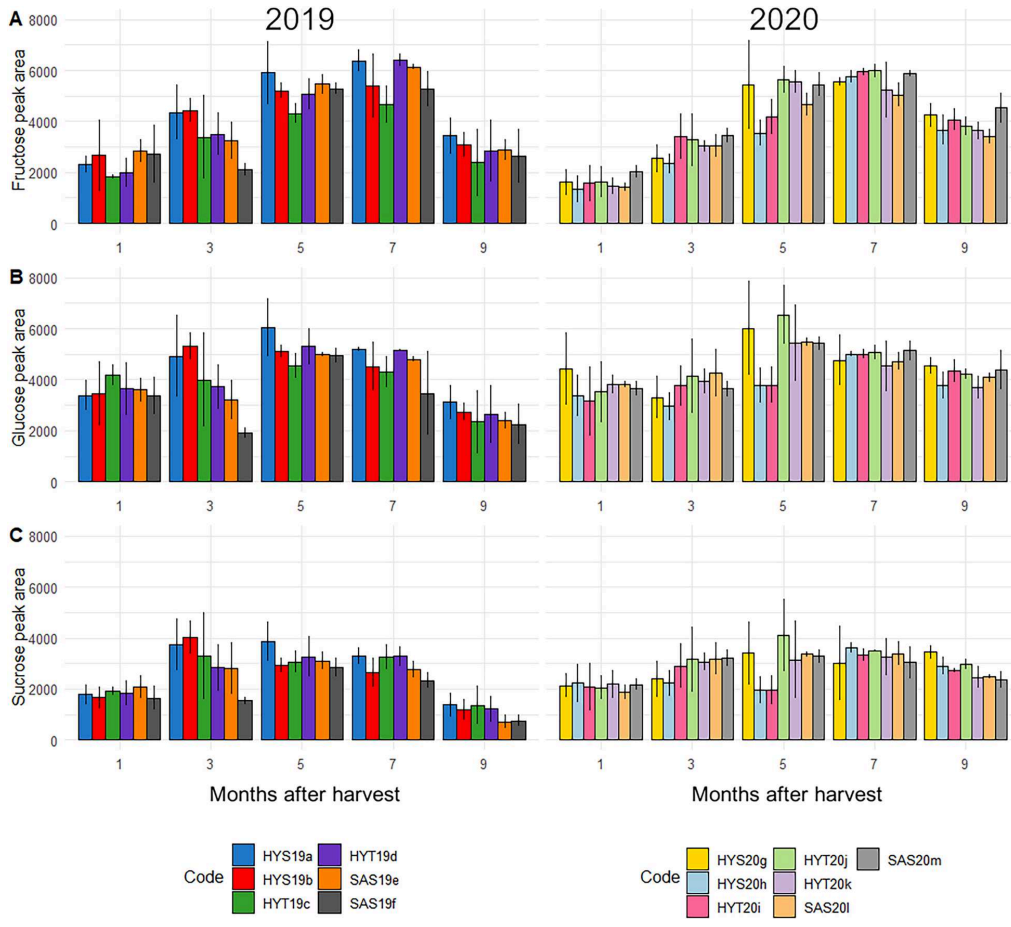

Figure 3. The amounts of fructose (A), glucose (B) and sucrose (C) found when sugars were extracted from homogenised and lyophilised onion bulbs collected from storage facilities from October (1) until June (9). Data shown as HPLC chromatogram peak area per mg extracted dry onion. Error bars represent the average value ($n = 4$) ± the standard deviation for the sample.

also showed a significant connection with sprouting without consideration of sampling month or sample ID. No significance was found for the other tested minerals: magnesium, copper, zinc, iron, sodium, manganese, boron and selenium. Nitrate and cobalt levels were below the detection threshold (0.3 g kg^{-1} and 43 µg kg^{-1}, respectively) and molybdenum content was either exactly at or below the detection threshold (0.2 mg kg^{-1}) in all samples. The overall dry weight percentages of the major minerals N, P, K, Ca and Mg were 1.28%, 0.34%, 1.76%, 0.26% and 0.09%, respectively. Appendix 1 lists all measured mineral contents. All minerals except magnesium, sodium and iron showed negative correlations with the dry matter content of the bulbs (Table 6).

Discussion

The purpose of this project was to measure a variety of onion quality indicators and identify those that showed promise as storability indicators, in order to help reduce storage losses. The hypothesis was that the measured quality indicators could be used to predict the storability of onions. We chose to focus on how the measured quality indicators changed over time and in relation to internal sprout development, as sprouting is a common cause of storage losses (Anbukkarasi et al. 2013; Sharma et al. 2016). We found that contents of sugars, dry matter and certain minerals were connected to the fraction of sprouting bulbs over time, while firmness showed no significant connection, but also tended to decrease over time. It can be assumed that

Table 5. The measured dry matter content of onion bulbs of the cultivars 'Hystore' (HYS), 'Hytech' (HYT) and 'Saskia' (SAS) harvested in 2019 (19) or 2020 (20) from 13 fields (a-m) in Skåne, Sweden.

	Months after harvest									
ID	1	2	3	4	5	6	7	8	9	Change (total)
2019										
HYS19a	10.2±0.2	10.4±0.3	10.3±0.3	9.6±0.3	9.6±0.4	9.2±0.3	9.3±0.1	9.3±0.4	9.5±0.2	−6.4%
HYS19b	11.0±0.2	10.6±0.9	10.8±0.3	10.2±0.4	10.3±0.4	10.2±0.3	10.0±0.1	9.9±0.3	10.2±0.6	−7.3%
HYT19c	12.2±0.1	11.8±0.1	11.9±0.4	10.7±0.1	11.3±0.3	11.2±0.1	10.9±0.3	11.1±0.2	10.8±0.2	−11.1%
HYT19d	10.8±1.0	10.6±0.4	10.6±0.5	10.3±0.3	10.0±0.2	10.0±0.3	9.7±0.4	9.8±0.4	9.9±0.3	−8.4%
SAS19e	9.6±0.4	9.8±0.4	9.5±0.4	9.3±0.3	9.0±0.8	9.0±0.4	9.0±0.4	8.8±0.3	8.9±0.2	−7.8%
SAS19f	9.8±0.9	9.7±0.4	9.6±0.4	9.3±0.5	9.3±0.7	9.0±0.3	9.3±0.3	8.8±0.7	9.3±0.7	−4.9%
Average	10.6	10.5	10.4	9.9	9.9	9.8	9.7	9.6	9.8	−7.8%
2020										
HYS20g	11.8±0.2		11.6±0.3	11.6±0.3	11.2±0.6	11.2±0.5	10.9±0.7	11.1±0.3	11.0±0.5	−6.6%
HYS20h	11.2±0.7		11.6±0.5	11.0±0.2	11.0±0.2	10.7±0.6	11.0±0.6	10.6±0.2	10.5±0.6	−6.2%
HYT20i	11.0±0.4		11.4±0.4	10.5±0.5	10.5±0.3	10.1±0.4	10.4±0.2	10.5±0.4	10.2±0.3	−7.0%
HYT20j	11.7±0.6		11.5±0.5	11.5±0.4	10.7±0.3	11.0±0.2	10.8±0.3	10.7±0.3	10.9±0.3	−7.1%
HYT20k	11.4±0.3		11.3±0.1	10.7±0.3	10.7±0.3	10.7±0.5	10.5±0.4	10.4±0.4	10.4±0.2	−8.7%
SAS20l	12.1±0.4		11.6±0.8	11.2±0.4	10.7±1.3	11.1±0.6	11.4±0.4	11.0±1.0	11.1±0.7	−7.8%
SAS20m	10.4±0.3		9.9±0.3	9.9±0.2	10.0±0.3	9.8±0.4	9.7±0.3	9.6±0.5	9.5±0.8	−8.9%
Average	11.4		11.3	10.9	10.7	10.7	10.7	10.6	10.5	−7.5%

All samples except HYT20k were treated with full dose maleic hydrazide (Fazor, Arysta LifeScience Great Britain Ltd, United Kingdom) at 80% tops down. Four replicates consisting of five random bulbs of each ID were sampled from storage every month from October until June, with the exception of November 2020, when no samples were taken. Average values for each month of the storage season are given in percentage dry matter with standard deviation, along with the overall change in dry matter content.

the increased content of minerals per gram of dry matter with time in storage (Table 6) are merely artefacts of the decreasing dry matter content, and as such are only indirect predictors of sprouting. This is in line with findings by Grevsen and Sorensen (2004), who found that the content of ash per fresh weight increased towards the end of the storage season, as the bulbs lost both carbohydrates and water. The contents of the minerals N, P, K, Ca and Mg found in the samples in this study were similar to values reported in the literature (Furlan and Bernier-Cardou 1989). In conclusion, the content of minerals at harvest had no effect on other quality indicators in this study.

As dry matter content seemed to decrease at similar rates for all samples (Table 5), it seems onion bulbs with a high initial dry matter content will retain a relatively high content later in the season, when compared to bulbs with a lower initial content. Jaime et al. (2001) found that onions with an initial dry matter content of at least 16% were better suited for storage, as fewer had sprouted after a six-month storage period. In our samples, we had no measured dry matter content above 13%, and most samples started the storage seasons with values below 11% in 2019 and below 12% in 2020. These values seem typical of onions produced in Scandinavia, as other studies have found that dry matter content of 11–13% is common at the start of storage and that this decreases to 9–11% towards the end of storage (Hansen 1999; Mogren et al. 2007). It has been shown that a relatively early harvest, at 20–50% so called 'tops down' rather than 80%, can result in a higher dry weight percentage in the harvested bulb and somewhat slower sprouting (Grevsen and Sorensen 2004). However, the earlier harvest also

Table 6. Pairwise Pearson correlations for the dry matter and mineral content in onion bulbs during storage.

Sample 1	Sample 2	N	Correlation	95% CI for ρ	*P*-Value	Significance
Sulphur (S)	Dry matter	72	−0.650	(−0.766; −0.493)	0.000	***
Nitrogen (N)	Dry matter	72	−0.347	(−0.536; −0.125)	0.003	**
Phosphorus (*P*)	Dry matter	72	−0.704	(−0.804; −0.564)	0.000	***
Potassium (K)	Dry matter	72	−0.298	(−0.495; −0.071)	0.011	*
Calcium (Ca)	Dry matter	72	−0.249	(−0.454; −0.018)	0.035	*
Magnesium (Mg)	Dry matter	72	−0.075	(−0.301; 0.159)	0.531	
Sodium (Na)	Dry matter	72	−0.086	(−0.311; 0.149)	0.474	
Chlorine (Cl)	Dry matter	72	−0.400	(−0.578; −0.185)	0.001	***
Zinc (Zn)	Dry matter	72	−0.304	(−0.501; −0.078)	0.009	**
Iron (Fe)	Dry matter	67	0.018	(−0.223; 0.257)	0.886	
Manganese (Mn)	Dry matter	72	−0.277	(−0.478; −0.049)	0.018	*
Copper (Cu)	Dry matter	72	−0.575	(−0.711; −0.395)	0.000	***
Boron (B)	Dry matter	72	−0.656	(−0.770; −0.500)	0.000	***
Selenium (Se)	Dry matter	72	−0.644	(−0.762; −0.484)	0.000	***

Onion bulbs harvested in 2019 were sampled at the beginning, middle and end (October, February and June) of the storage season. All samples were pooled for pairwise comparison.

resulted in a lower total weight of marketable bulbs, and the long-term sprouting reduction was not as effective as for bulbs treated with maleic hydrazide (ibid.). We found that SAS20 m, which was sown later than any other ID in 2020, had the lowest dry matter content, but did not differ from the other IDs in terms of sprouting (Table 4). We also found a significant connection between the onset of sprouting and the increase in content of the monosaccharides glucose and fructose (Figure 2). The changing ratio between mono- and disaccharides has previously been reported to be a good indicator of sprouting status by Chope et al. (2012). The subsequent decrease in fructose, glucose and sucrose in our study is also as expected, as mono- and disaccharides are consumed in the sprouting process (Sheikh et al. 2022). The peak of mono- and disaccharide content coinciding with the start of sprouting can be attributed to an increased activity of fructan-degrading enzymes, a process that has been pointed out as a possible strong indicator of the start of the sprouting stage (Benkeblia et al. 2005). In our study, we did not examine enzyme activity, but it seems the increase in monosaccharides can serve as an easily measurable, if a bit later, indicator of the same process. HPLC analysis of sugar content showed changes over time (Figure 3). Similar measurements could also be made with simple and relatively cheap methods, e.g. Merck reflectoquant system (Kleman et al. 2023). Another process that can be indicative of sprouting initiation is the redistribution of fructans in the bulb (Ohanenye et al. 2019). However, we homogenised all non-dry scales of the bulbs and could therefore not detect movement of sugars. Further studies regarding the redistribution of sugars in bulbs and whether this could be useful in storage decision making may be relevant.

The varying rates of decrease in firmness is another interesting quality indicator. We found significant differences in loss of firmness for different samples of the cultivar 'Hytech' (Table 3). It could be assumed that this was influenced by the cultivation and handling of the onions, and could be predictive of storability. Firmness is connected to dry matter content and, therefore, storability (Coolong et al. 2008), and is a relatively easy parameter to measure. Regular penetrometer measurements could easily be applied in storage, and batches of onions with faster rates of decline in firmness could be sold sooner than batches that retain their firmness well. A mounted penetrometer should be used, as we found that the values obtained from the handheld penetrometer were rather variable, even if the overall trend remained clear. The high variability in the measurements obtained from the handheld penetrometer may be part of the reason we did not see any significant connection between sprouting and firmness. However, it has been shown that a handheld penetrometer can give relatively reliable and consistent measurements in the case of sugar beets, but it requires a defined operating procedure and training of the person performing the measurements (English et al. 2022). Dry matter content is another quality indicator that is easy, albeit slower, to measure and only requires access to an oven or lyophiliser and scales.

Conclusion

In summary, measurements of dry weight, firmness and sugar content are useful ways of following the quality of a batch of onion bulbs over the course of a storage season. The content of monosaccharides is a good indication of sprouting status and could aid in deciding which stored batches to sell and thus help reduce losses in storage.

Acknowledgements

The authors wish to thank Oskar Hansson and Almhaga Grönsaker for providing us with the onions and Karl-Erik Gustavsson for the help with running the HPLC sugar samples.

Disclosure statement

No potential conflict of interest was reported by the authors.

Funding

This work was supported by Stiftelsen Svensk Lantbruksforskning under Grant R-18-25-146. Funding was also received from the Swedish University for Agricultural Sciences.

Notes on contributors

Isabella Kleman is a PhD student at the Department of Biosystems and Technology, at the Swedish University of Agricultural Sciences in Alnarp, Sweden. Her thesis focuses on the prediction and detection of quality problems during onion storage.

Anna Karin Rosberg is a Senior Lecturer in horticulture with emphasis on systems biology in horticultural value networks at the Department of Biosystems and Technology at the Swedish University of Agricultural Sciences, Alnarp, Sweden.

Lars Mogren is an Associate Professor in Horticulture with emphasis on produce quality in vegetable production at the Department of Biosystems and Technology at the Swedish University of Agricultural Sciences, Alnarp, Sweden.

CRediT authorship contribution statement

Isabella Kleman: PhD student, performed majority of experiments, data curation, prepared the manuscript.

Anna Karin Rosberg: Supervision, edited and reviewed the manuscript. **Lars Mogren**: Performed early experiments, method supervision, edited and reviewed the manuscript. All authors have reviewed the final version of the manuscript and agree to the submission and to be accountable for all aspects of the work.

Data availability statement

The data that support the findings of this study are available from the corresponding author, IK, upon reasonable request.

References

Abdalla AA, Mann LK. 1963. Bulb development in the onion (*Allium cepa* L.) and the effect of storage temperature on bulb rest. Hilgardia. *35*(5):85–112. doi:10.3733/hilg.v35n05p085.

Anbukkarasi V, Paramaguru P, Pugalendhi L, Ragupathi N, Jeyakumar P. 2013. Studies on pre and post-harvest treatments for extending shelf life in onion – a review. Agricultural Reviews. *34*(4):256. doi:10.5958/j.0976-0741.34.4.011.

Benkeblia N, Ueno K, Onodera S, Shiomi N. 2005. Variation of fructooligosaccharides and their metabolizing enzymes in onion bulb (*Allium cepa* L. cv. Tenshin) during long-term storage. Journal of Food Science. *70*(3):S208–S214. doi:10.1111/j.1365-2621.2005.tb07159.x.

Carter CE, Partis MD, Thomas B. 1999. The expression of histone 2A in onion (*Allium cepa*) during the onset of dormancy, storage and emergence from dormancy. New Phytologist. *143*(3):461–470. doi:10.1046/j.1469-8137.1999.00484.x.

Chope GA, Cools K, Hammond JP, Thompson AJ, Terry LA. 2012. Physiological, biochemical and transcriptional analysis of onion bulbs during storage. Annals of Botany. *109*(4):819–831. doi:10.1093/aob/mcr318.

Coolong TW, Randle WM, Wicker L. 2008. Structural and chemical differences in the cell wall regions in relation to scale firmness of three onion (*Allium cepa* L.) selections at harvest and during storage. Journal of the Science of Food and Agriculture. *88*(7):1277–1286. doi:10.1002/jsfa.3219.

Darbyshire B, Henry RJ. 1979. The association of fructans with high percentage dry weight in onion cultivars suitable for dehydrating. Journal of the Science of Food and Agriculture. *30*(11):1035–1038. doi:10.1002/jsfa.2740301103.

Downes K, Chope GA, Terry LA. 2010. Postharvest application of ethylene and 1-methylcyclopropene either before or after curing affects onion (*Allium cepa* L.) bulb quality during long term cold storage. Postharvest Biology and Technology. *55*(1):36–44. doi:10.1016/j.postharvbio.2009.08.003.

English W, Ekelöf J, Vancutsem F, Leijdekkers M, Kleuker G, Hoffmann CM. 2022. Method for in-field texture analysis of sugar beet roots using a handheld penetrometer. Acta Agriculturae Scandinavica, Section B — Soil & Plant Science. *72*(1):623–634. doi:10.1080/09064710.2022.2042589.

EU Platform on Food Losses and Food Waste. 2019. Recommendations for action in food waste prevention. EU Platform on Food Losses and Food Waste Brussels.

FAOSTAT. 2023. *FAOSTAT*. FAOSTAT Statistical Database. http://www.fao.org/faostat/en/#data/QCL.

Forney CF, Jordan MA, Campbell-Palmer L, Fillmore S, McRae K, Best K. 2010. Sulfur fertilization affects onion quality and flavor chemistry during storage. Acta Horticulturae. *877*:163–168. doi:10.17660/ActaHortic.2010.877.14.

Franke U, Ministerråd N, Råd N. 2013. *Kartläggning av matsvinnet i primärproduktionen*. Nordisk Ministerråd : Nordisk Råd : [Eksp.] www.norden.org/order.

Furlan V, Bernier-Cardou M. 1989. Effects of N, P, and K on formation of vesicular-arbuscular mycorrhizae, growth and mineral content of onion. Plant and Soil. *113*(2):167–174. doi:10.1007/BF02280177.

Golubkina N, Amalfitano C, Sekara A, Tallarita A, Pokluda R, Stoleru V, Cuciniello A, Agafonov AF, Kalisz A, Hamburdă SB, Caruso G. 2022. Yield and bulb quality of storage onion cultivars as affected by farming system and nitrogen dose. Scientia Horticulturae. *293*:110751. doi:10.1016/j.scienta.2021.110751.

Greulach VA, Atchison E. 1950. Inhibition of growth and cell division in onion roots by maleic hydrazide. Bulletin of the Torrey Botanical Club. *77*(4):262–267. doi:10.2307/2481897.

Grevsen K, Sorensen J. 2004. Sprouting and yield in bulb onions (*Allium cepa* L.) as influenced by cultivar, plant establishment methods, maturity at harvest and storage conditions. Journal of Horticultural Science and Biotechnology. *79*:877–884. doi:10.1080/14620316.2004.11511860.

Gubb IR, MacTavish HS. 2002. 10 onion pre-and postharvest considerations. Allium Crop Science: Recent Advances. 233.

Hansen SL. 1999. Content and composition of dry matter in onion (*Allium cepa* L.) as influenced by developmental stage at time of harvest and long-term storage. Acta Agriculturae Scandinavica, Section B-Plant Soil Science. *49*(2):103–109.

Islam MN, Wang A, Skov Pedersen J, Edelenbos M. 2015. Microclimate tools to monitor quality changes in stored onions. In: V International symposium on applications of modelling as an innovative technology in the horticultural supply chain-model-IT 1154. p. 229–234.

Jaime L, Martín-Cabrejas MA, Mollá E, López-Andréu FJ, Esteban RM. 2001. Effect of storage on fructan and fructooligosaccharide of onion *(Allium cepa* L.). Journal of Agricultural and Food Chemistry. *49*(2):982–988. doi:10.1021/jf000921t.

Kleman I, Hellström M, Rosberg AK, Becher PG, Mogren L. 2023. Simple quality prediction measurements for stored onions. Acta Horticulturae. *1364*:183–186. doi:10.17660/ActaHortic.2023.1364.24.

Ko S-S, Chang W-N, Wang J-F, Cherng S-J, Shanmugasundaram S. 2002. Storage variability among short-day onion cultivars under high temperature and high relative humidity, and its relationship with disease incidence and bulb characteristics. Journal of the American Society for Horticultural Science Jashs. *127*(5):848–854. doi:10.21273/JASHS.127.5.848.

Lancaster JE, Farrant J, Shaw ML. 2001. Sulfur nutrition affects cellular sulfur, dry weight distribution, and bulb quality in onion. Journal of the American Society for Horticultural Science Jashs. *126*(2):164–168. doi:10.21273/JASHS.126.2.164.

Mogren LM, Olsson ME, Gertsson UE. 2007. Quercetin content in stored onions (*Allium cepa* L.): effects of storage

conditions, cultivar, lifting time and nitrogen fertiliser level. Journal of the Science of Food and Agriculture. *87*(8):1595–1602.

Ohanenye IC, Alamar MC, Thompson AJ, Terry LA. 2019. Fructans redistribution prior to sprouting in stored onion bulbs is a potential marker for dormancy break. Postharvest Biology and Technology. *149*:221–234. doi:10.1016/j.postharvbio.2018.12.002.

Olanipekun CI. 2018, June 15. Increasing production and reducing postharvest losses of onion in Nigeria. *World Vegetable Center*. https://avrdc.org/increasing-production-and-reducing-postharvest-losses-of-onion-in-nigeria/.

Pak C, van der Plas LHW, de Boer AD. 1995. Importance of dormancy and sink strength in sprouting of onions (*Allium cepa*) during storage. Physiologia Plantarum. *94*(2):277–283. doi:10.1111/j.1399-3054.1995.tb05312.x.

Petropoulos SA, Ntatsi G, Ferreira ICFR. 2017. Long-term storage of onion and the factors that affect its quality: a critical review. Food Reviews International. *33*(1):62–83. doi:10.1080/87559129.2015.1137312.

Põldma P, Moor U, Merivee A, Tõnutare T. 2011. Effect of controlled atmosphere storage on storage life of onion and garlic cultivars. In: IV International conference postharvest unlimited 2011 945. p. 63–69.

Schwartz HF, Mohan SK. 2007. *Compendium of onion and garlic diseases and pests*, Second Edition. City: The American Phytopathological Society.

Sharma K, Asnin L, Ko EY, Lee ET, Park SW. 2015. Phytochemical composition of onion during long-term storage. Acta Agriculturae Scandinavica, Section B — Soil & Plant Science. *65*(2):150–160. doi:10.1080/09064710.2014.983151.

Sharma K, Rok Lee Y, Park SW, Nile SH. 2016. Importance of growth hormones and temperature for physiological regulation of dormancy and sprouting in onions. Food Reviews International. *32*(3):233–255. doi:10.1080/87559129.2015.1058820.

Sharma P, Sharma SR, Dhall RK, Mittal TC. 2020. Effect of γ-radiation on post-harvest storage life and quality of onion bulb under ambient condition. Journal of Food Science and Technology. *57*(7):2534–2544. doi:10.1007/s13197-020-04290-z.

Sheikh FR, Jose-Santhi J, Kalia D, Singh K, Singh RK. 2022. Sugars as the regulators of dormancy and sprouting in geophytes. Industrial Crops and Products. *189*:115817. doi:10.1016/j.indcrop.2022.115817.

Shelake PS, Mohapatra D, Tripathi MK, Giri SK, Kate A, Kumar M. 2022. Inactivation of Aspergillus niger and Erwinia carotovora in onion (*Allium cepa* L.) bulbs subjected to pulsed ozone treatment. Postharvest Biology and Technology. *192*:111998. doi:10.1016/j.postharvbio.2022.111998.

Stefanova R, Vasilev NV, Spassov SL. 2010. Irradiation of food, current legislation framework, and detection of irradiated foods. Food Analytical Methods. *3*:225–252.

UN General Assembly. 2015. *Transforming our world: The 2030 Agenda for Sustainable Development* (A/RES/70/1). https://www.refworld.org/docid/57b6e3e44.html.

UNECE. 2019. *UNECE Standard FFV-25 concerning the marketing and commercial quality control of onions*. UNITED NATIONS New York and Geneva. https://unece.org/fileadmin/DAM/trade/agr/standard/fresh/FFV-Std/English/25_Onions.pdf.

Yasin HJ, Bufler G. 2007. Dormancy and sprouting in onion (*Allium cepa* L.) bulbs. I. changes in carbohydrate metabolism. The Journal of Horticultural Science and Biotechnology. *82*(1):89–96. doi:10.1080/14620316.2007.11512203.

Appendix

The contents of 14 minerals in onion bulbs of the cultivars 'Hystore' (HYS), 'Hytech' (HYT) and 'Saskia' (SAS) harvested in 2019 from six fields (a-f) in Skåne, Sweden. Four replicates consisting of five random bulbs of each ID were sampled from storage in October, February and June, and the resulting 12 values were pooled to calculate the average contents of each mineral. Mineral contents are given per kg dry matter. [1] n = 7, as five of the samples had iron content below the detection threshold of 22 mg kg^{-1}. Capital letters indicate grouping, generated using Tukey pairwise comparisons and 95% confidence.

ID	N g kg^{-1}		S g kg^{-1}		*P* g kg^{-1}		K g kg^{-1}		Ca g kg^{-1}		Mg g kg^{-1}		Na g kg^{-1}		Cl g kg^{-1}		Fe mg kg^{-1}		Zn mg kg^{-1}		Mn mg kg^{-1}		Cu mg kg^{-1}		B mg kg^{-1}		Se µg kg^{-1}	
HYS19a	12.15	BC	4.53	B	3.17	B	13.58	C	2.70	B	0.83	BC	1.23	A	2.73	B	27.58	BC	15.25	D	13.42	BC	4.45	BC	15.26	BC	21.08	A
HYS19b	12.12	BC	4.59	B	3.30	B	18.86	A	2.16	C	0.83	BC	0.34	D	2.24	D	32.83	A	19.92	B	15.25	B	4.48	BC	14.64	C	13.75	B
HYT19c	14.08	A	4.64	B	3.20	B	16.90	B	2.94	AB	0.87	AB	0.58	C	2.63	B	28.17	AB	17.42	C	13.25	BC	4.33	BC	15.76	BC	9.92	C
HYT19d	10.76	C	4.08	B	3.20	B	18.24	A	1.78	C	0.92	A	0.76	B	2.50	C	27.92	BC	17.08	CD	12.08	C	4.13	C	16.13	B	14.75	B
SAS19e	13.57	AB	5.90	A	3.86	A	18.81	A	3.17	A	0.80	C	0.33	D	2.51	C	24.04[1]	C	16.33	CD	12.08	C	5.41	A	17.93	A	24.67	A
SAS19f	14.01	A	5.64	A	3.85	A	19.20	A	2.69	B	0.89	A	0.53	C	3.08	A	31.92	AB	23.33	A	19.92	A	4.89	AB	17.53	A	14.75	B

Acta Universitatis agriculturae Sueciae
Doctoral Thesis No. 2025:10

Onions are typically stored for weeks or months before consumption, and during storage losses occur due to sprouting and storage diseases. To prevent these losses, storage monitoring systems can be developed and improved. In this thesis, the influence of physiological parameters on sprouting, and the volatile compounds released by diseased bulbs were investigated, with the aim to identify metrics useful for storage monitoring.

Isabella Kleman received her doctoral education at the Department of Biosystems and Technology, SLU, Alnarp. Her BSc and MSc in Biology were received at SLU, Alnarp.

Acta Universitatis agriculturae Sueciae presents doctoral theses from the Swedish University of Agricultural Sciences (SLU).

SLU generates knowledge for the sustainable use of biological natural resources. Research, education, extension, as well as environmental monitoring and assessment are used to achieve this goal.

ISSN 1652-6880
ISBN (print version) 978-91-8046-445-1
ISBN (electronic version) 978-91-8046-495-6