Content of dietary fibre and phenolic compounds in broccoli side streams

Emilia Berndtsson

Faculty of Landscape Architecture, Horticulture and Crop Production Science Department of Plant Breeding Alnarp



Licentiate thesis Swedish University of Agricultural Sciences Alnarp 2020

Content of dietary fibre and phenolic compounds in broccoli side streams

Emilia Berndtsson

Faculty of Landscape Architecture, Horticulture and Crop Production Science Department of Plant Breeding Alnarp

> Licentiate thesis Swedish University of Agricultural Sciences Alnarp 2020

Cover image: Pictures of broccoli plant, freeze-dried broccoli leaves, methanol extraction of broccoli leaves, chemical structures of some phenolic compounds and dietary fibre constituents and a broccoli cake with cream cheese frosting. (photos: Emilia Berndtsson)

ISBN (print version) 978-91-576-9740-0 ISBN (electronic version) 978-91-576-9741-7 © 2020 Emilia Berndtsson, Alnarp Print: SLU Service/Repro, Alnarp 2020

Content of dietary fibre and phenolic compounds in broccoli side streams

Abstract

Shortage of food is an alarming problem today, with up to 821 million people that are undernourished world-wide. At the same time, enough edible foodstuff to feed 1.9 billion people are wasted or lost in the food supply chain that for aesthetic reasons, handling and transportation inadequacies in the food supply chain and lack of market. In 2014, this wasted or lost food (including only commonly consumed plant parts) corresponded to 3.49 GT CO₂ equivalents globally, which was more than half the total amount of emissions in the USA that year. This means that there is much biomass that could be valorised into nutritional food ingredients or used for extraction of health beneficial compounds, *e.g.* dietary fibre. Many consumers currently eat too little dietary fibre and phenolic compounds, which can lead to increased risk of developing some forms of cancer and cardiovascular diseases.

In this thesis, the content of dietary fibre and phenolic compounds in the broccoli leaves and stems was determined. The relationship between the dietary fibre and phenolic compounds in broccoli leaves was also analysed, since recent research indicates that interactions between these plant components may have an impact on the uptake in the human gastrointestinal tract. The results revealed that broccoli leaves contain similar levels of dietary fibre (26-32 % of dry weight (DW)) to cabbage, broccoli florets and kale leaves, which are regarded as beneficial to human health. The content of phenolic compounds (6.3-15.2 mg/g DW) in broccoli leaves was similar to that in kale leaves and much higher than that in broccoli florets. Some phenolic acids showed positive correlations with soluble dietary fibre in the broccoli leaves, but no correlation was found between the insoluble dietary fibre and phenolic compounds. A pilot study on field waste showed that leaves and stems of broccoli plants make up 43-87 % of total plant weight, indicating that substantial amounts of biomass are left in the field at harvest.

Overall, the analysis of this thesis showed that the broccoli leaves are interesting from a food ingredient perspective. Possible uses could be to add broccoli leaf powder to everyday food products, such as pasta or bread, or into gluten-free products to increase the nutritional and technical properties.

Keywords: broccoli, dietary fibre, field waste, food loss, food waste, health benefits, phenolic compounds

Author's address: Emilia Berndtsson, SLU, Department of Plant Breeding, P.O. Box 101 23053, Alnarp, Sweden

Innehåll av kostfibrer och fenoliska ämnen i sidoströmmar hos broccoli

Abstrakt

Brist på mat är ett alarmerande problem idag, med upp till 821 miljoner människor som är undernärda i världen. Samtidigt slängs tillräckligt mycket fullt ätliga livsmedel i livsmedelskedjan som hade kunna mätta 1,9 miljarder människor, på grund av kosmetiska orsaker, brist i hanteringen och transporter i livsmedelskedjan och brist på efterfrågan. Under 2016 motsvarande detta matsvinn och matavfall 3,49 GT CO₂-ekvivalenter, vilket var mer än hälften av USA:s totala utsläpp under det året. Detta innebär att det är mycket biomassa som skulle kunna bli förädlad till näringsrika livsmedelsingredienser eller som kan användas som råmaterial för att extrahera hälsofrämjande ämnen, exempelvis kostfibrer. Många konsumenter äter för närvarande för lite kostfibrer och fenoliska ämnen, vilket kan leda till en ökad risk för att utveckla vissa former av cancer och även hjärt-kärl-sjukdomar.

I denna avhandling mättes halterna av kostfibrer och fenoliska ämnen i broccoliblad och broccolistam. Samband mellan kostfibrer och fenoliska ämnen i broccoliblad analyserades, eftersom nya forskningsrön har indikerat att det finns samband mellan dessa växtkomponenter, vilket kan påverka upptaget i mag-tarm-kanalen hos människor. Resultaten visade att broccoliblad innehåller halter av kostfibrer (26–32 % av torrvikten (DW)) som är motsvarande halterna som finns i vitkål, broccolibuketter och grönkålsblad, som anses vara hälsofrämjande. Halterna av fenoliska ämnen (6,3–15,2 mg/g DW) i broccoliblad var liknande de som tidigare hittats i grönkål och mycket högre jämfört med innehållet i broccolibuketter. Några av de fenoliska syrorna visade även på positiva korrelationer med några lösliga kostfibrerna i broccolibladen, men ingen korrelation hittades mellan de olösliga fibrerna och de fenoliska ämnena. En förstudie visade att bladen och stammen av broccoliplantan utgör en 43–87 % av plantans vikt, vilket indikerar att det är väsentlig del av biomassan som lämnas på fältet vid skörd.

Sammanfattningsvis så har analyserna i denna avhandling visat att broccoliblad är intressanta ur ett livsmedelsperspektiv. Möjliga användningsområden kan vara att tillsätta pulver av broccoliblad i vardagliga livsmedel, exempelvis pasta och bröd, eller i glutenfria produkter för att öka näringsvärdet och de tekniska egenskaperna.

Nyckelord: broccoli, fenoliska ämnen, fältavfall, hälsofrämjande, kostfibrer, matsvinn

Författarens adress: Emilia Berndtsson, SLU, Avdelningen för Växtförädling, Box 101 230 53, Alnarp, Sweden

Dedication

To my family, both biological and social

"Too many scholars think of research as purely a cerebral pursuit. If we do nothing with the knowledge we gain, then we have wasted our study. Books can store information better than we can – what we do that books cannot is interpret. So if one is not going to draw conclusions, then one might as well just leave the information in the texts"

"Alltför många forskare anser att forskning är en ren cerebral strävan. Om vi inte gör något med den kunskap vi får så har vi slösat bort våra studier. Böcker kan lagra information bättre än vi kan - det vi gör som böcker inte kan är att tolka. Så om man inte kommer att dra slutsatser, kan man lika gärna lämna informationen i texterna"

Brandon Sanderson, The Way of Kings, p 462

Contents

List	of publications	11
List	of tables	14
List	of figures	15
Abbı	reviations	16
1	Introduction	17
1.1	We have to eat for the climate	17
1.2	We have to eat for our health	19
	1.2.1 Dietary fibre	19
	1.2.2 Phenolic compounds	20
	1.2.3 Antioxidant dietary fibre - Together we stand?	22
1.3	Broccoli - an underutilised superplant	23
1.4	Side streams, food loss and food waste - confusing terms	25
1.5	Products from broccoli side streams?	26
2	Thesis aims	29
3	Methods	31
3.1	Plant material	31
3.2	Soluble and insoluble dietary fibre	32
3.3	Quantification and identification of phenolic compounds	32
3.4	Relationships between phenolic compounds and dietary fibre	32
3.5	Field waste in commercial broccoli production (pilot study)	33
4	Results and discussion	35
4.1	Dietary fibre and phenolic content in broccoli side streams	35
	4.1.1 Dietary fibre	35
	4.1.2 Phenolic compounds	36
	4.1.3 Possible uses	40
4.2	Field waste in the broccoli fields (pilot study)	40
	4.2.1 Amount left in the field at harvest	40

4.3	Ethics	41
5	Conclusions	43
6	Future perspectives	45
Refer	ences	47
Popu	lar science summary	55
Popu	lärvetenskaplig sammanfattning	57
Ackno	owledgements	59

List of publications

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

- I E. Berndtsson*, A-L. Nynäs, W. Newson, M. Langton, R. Andersson, E. Johansson, M.E. Olsson (2019). The underutilised side streams of broccoli and kale Valorisation via proteins and phenols. In: *Sustainable Governance and Management of Food Systems* (eds. Eija Vinnari & Markus Vinnari), Wageningen Academic Publishers, Wageningen, pp. 153-159.
- II E. Berndtsson*, R. Andersson, E. Johansson, M.E. Olsson (2020). Side stream of broccoli leaves: a climate smart and healthy food ingredient. *International Journal of Environmental Research and Public Health*. 17(7), pp 2506.

Paper I is reproduced with the permission of the publishers. Paper II is Open Access.

* Corresponding author.

The contribution of Emilia Berndtsson to the papers included in this thesis was as follows:

- I Planned and gathered reference articles for the review together with Anna-Lovisa Nynäs. Wrote the manuscript together with Anna-Lovisa Nynäs and with input from other co-authors.
- II Planned the study together with supervisors, collected samples and analysed dietary fibre and phenolic compounds with the aid from laboratory technicians. Analysed the data and performed statistical analyses with the aid of the supervisors and laboratory technicians. Wrote the manuscript with input from the co-authors

List of tables

Table 1: Content of dietary fibre (insoluble dietary fibre (IDF), soluble dietary fibre (SDF) and total dietary fibre (TDF) in different broccoli parts sampled in the two study years	36
Table 2 Levels of total dietary fibre in some vegetables and vegetable parts.	36
Table 3: Total content of phenolic compounds (assessed as gallic acid equivalents (GAE) in broccoli leaves and stems collected from two different fields in southern Sweden in 2018	י 37
Table 4: Content of phenolic compounds determined by methanol extraction different broccoli plant parts and in kale leaves.	in 38
Table 5: Measured weight [kg] of separate parts of broccoli plant and of the whole broccoli plant the three sampling squares used in the pilot study on field wast	41

List of figures

Figure 1. Emissions of CO2-equivalents from food waste compared with tota emissions in different countries in 2014	l 18
Figure 2: Model of the cell wall of a plant cell	20
Figure 3: Examples of phenolic compounds commonly found in Brassica vegetables.	21
Figure 4: A broccoli plant divided into broccoli head (with florets), stem (with roots) and leaves	23
Figure 5: A harvested broccoli field, with remaining stems, leaves and discarded broccoli heads	24
Figure 6: Expressions applied at different stages in the food supply to denote non-eaten produce.	, 25
Figure 7: Principal component analysis of relationship between dietary fibre constituents and phenolic compounds in broccoli leaves	39

Abbreviations

IDF	Insoluble dietary fibre
SDF	Soluble dietary fibre
TDF	Total dietary fibre
SCFA	Short chain fatty acids
~ ~	

CO₂e Carbon dioxide equivalents, kg CO2 emitted per kg produce

1 Introduction

1.1 We have to eat for the climate

The food situation in the world today is unbalanced. For example, data for 2017 showed that up to 821 million people world-wide were undernourished that year (FAO *et al.*, 2018), an increase from 815 million people in 2016 (FAO *et al.*, 2017). In order to feed a growing global population, food production must increase. According to FAO, global food production must increase by 60 % compared to the levels in 2007 in order to feed the population by 2050 (Alexandratos & Bruinsma, 2012). This could be done by increasing the total area of cultivated land and/or by cultivating the crops more effectively, but this also means increased use of limited resources. An alternative way is a more extensive use of the biomass produced on the land cultivated today, *i.e.* development of strategies for using plant waste as an additional food resource. But is this feasible? Is the nutritional content of the wasted plant parts sufficient to support such strategies? And which wasted parts that might be used?

Approximately 30 % of all food becomes waste (Gustavsson *et al.*, 2011). This volume of waste corresponds to the nutrition needed to feed 1.9 billion people (Kummu *et al.*, 2012). If all the food produced world-wide were eaten, there would be enough food for the entire global population and more, without increasing the use of limited resources such as water, fertilisers and arable land.

Throughout the food supply chain, water, fertilisers, farmland, and energy are invested, which results in greenhouse gas emissions (Kummu *et al.*, 2012; Bryngelsson *et al.*, 2016; Röös *et al.*, 2018). Transportation, storage and handling of food also have an impact on emissions of carbon dioxide equivalents (CO₂e), per kg product in food production (Wakeland *et al.*, 2012), but their actual contribution is difficult to evaluate. The FAO has estimated that 1.7 billion tonnes of food waste are produced every year throughout the chain, which

requires 0.9 million hectares of farmland and 306 km³ of drinkable water while emitting 3.49 gigatonnes of CO₂e (FAO, 2014). Comparing the emissions generated by the total amount of food waste to the total emissions of individual countries, it would have been ranked it as the third-largest country in the world, after China and the US (Figure 1).



Emissions of CO₂e [Gt], 2014

Figure 1. Emissions of CO₂-equivalents from food waste compared with total emissions in different countries in 2014. Data from www.climatewatchdata.org and FAO (2014).

However, in most calculations of the resources needed for producing food that will become waste, only the harvested parts of plants are included, *i.e.* parts which can be eaten, but these often constitute a minor proportion of the total biomass. Thus for a wide range of fruits and vegetables, the majority of the plant biomass is wasted already in the field before or at harvest. Iceberg lettuce is an example of a vegetable with a high level of waste in the entire food supply chain. In the primary production, more than 60% of the lettuce biomass produced is left as field waste due to damages, for aesthetic reasons or because of over-production, and an additional 12% of the harvested lettuce is lost on the way to the consumer (Strid *et al.*, 2014). This represents unsustainable use of nutrients and resources in an era of increasing global hunger and a changing climate, but as long as there is insufficient data on the problem, it is difficult to know how to handle the field waste. If the total amount of waste at the beginning of the food supply chain were to be measured, it could also be managed, *e.g.* by using it for food purposes.

1.2 We have to eat for our health

1.2.1 Dietary fibre

The term dietary fibre was coined in the 1950s (Hipsley 1953) and ever since there has been active discussion about the definition of the term. One part of the definition that has remained relatively unchanged over the years is that dietary fibre is resistant to the digestive enzymes of humans and mainly consists of polysaccharides. Examples of dietary fibre are soluble pectins, gums (e.g. guar gum) and mucilage (a gelatinous substance from e.g. flax seeds), and insoluble hemicellulose and cellulose (Nawirska & Kwasniewska, 2005). Compounds other than carbohydrates may be included in dietary fibre, such as the phenolic compounds lignin, if they are associated with polysaccharides in the plant cell wall (Codex Alimentarius, 2017). At the beginning of the 21st century, healthpromoting aspects were also included in the definition of dietary fibre (Food and Nutrition Board, 2001). The current international definition from Codex Alimentarius states that dietary fibre comprises carbohydrate polymers, or associated compounds, with a degree of polymerisation not lower than 3. Moreover, they are not digested nor absorbed in the small intestine, and they decrease the intestinal transit time, increase stool bulk, are fermentable by gut microbiota and can reduce cholesterol levels in the blood (Codex Alimentarius, 2017).

There are ongoing discussions about the consequences of classification of dietary fibre into soluble and insoluble, *e.g.* that solubility is an insufficient characteristic due to the matrix in the plant material and the diverse chemical structures of dietary fibre (Williams *et al.*, 2019). An alternative suggestion is that dietary fibre should be classified according to other characteristics, such as level of fermentability by the gut microbiota (Williams *et al.*, 2019).

Numerous functions are associated with the cell wall in the living plant, with its content of dietary fibre (Figure 2). It provides strength to the plant cell, affects the transportation of larger compounds in and out of the cell, it influences the growth of the cell and can protect against herbivores (Brett & Waldron, 1996; Taiz *et al.*, 2015). For the plant as a whole, the cell wall provides structural support against gravity and environmental forces and also makes water transport possible in tall plants (Brett & Waldron, 1996; Taiz *et al.*, 2015)

In humans, high intake of dietary fibre is associated with lower mortality from cardiovascular disease, coronary heart disease, and cancer (Kim & Je, 2016). Dietary fibre also lowers the levels of cholesterol in the blood (Mandimika *et al.*, 2012), has an impact on the rate of gastric emptying (Mackie *et al.*, 2016), promotes peristaltic movement in the intestines (Wrick *et al.*, 1983)



Figure 2: Model of the cell wall of a plant cell. Pectin, cellulose, soluble proteins and hemicellulose build up a matrix.

Licence under Creative Commons from author LadyOfHats: https://commons.wikimedia.org/wiki/File:Plant_cell_wall_diagram-en.svg

and affects the gut microbiota (Yang et al., 2013). However, some of the positive health effects reported for high intake of dietary fibre might not be from the fibres themselves. A group of by-products, e.g. short-chained fatty acids (SCFA), which are excreted by the gut bacteria when fermenting the dietary fibre, are been reported to have similar positive effects as dietary fibre on the general health (den Besten et al., 2013; Sawicki et al., 2017). The SCFA have been found to lower the risk of depression (Miki et al., 2016), regulate the uptake of lipids, affect the cholesterol metabolism (den Besten et al., 2013) and improve the immune system (Corrêa-Oliveira et al., 2016; Makki et al., 2018). Bacteria strains in the gut microbiota are affected differently by the dietary fibre content, but an increased level of dietary fibre generally increases the amounts of healthpromoting bacteria (Yang et al., 2013) which means that the gut microbiota can be readily affected by changes in the diet (Li et al., 2009). All these arguments point towards a diet rich in dietary fibres being good for human health and wellbeing. However, most modern diets contain too low amounts of dietary fibre, which might have a harmful impact on health. In many Western countries, the average daily intake of dietary fibre is 15-25 g/day, compared with a recommended daily intake of 20-38 g/day (Stephen et al., 2017).

1.2.2 Phenolic compounds

Phenolic compounds are a diverse group of substances with antioxidative properties (Shen *et al.*, 2017) and with a structure that includes an aromatic ring (six carbons in a hexagonal ring) with at least one hydroxyl group (-OH) (Shahidi & Naczk, 2004). More than 8000 different types of phenolic

compounds have been identified in plants (Shen *et al.*, 2017), and of these, hundreds have been characterised in plant-based foods (Manach *et al.*, 2005; Cartea *et al.*, 2011). Depending on their chemical structure, the phenolic compounds are usually divided into groups, *e.g.* flavonoids, phenolic acids such as hydroxybenzoic acids stilbenes, tannins, lignans and lignins (Figure 3).



Figure 3: Examples of phenolic compounds commonly found in Brassica vegetables.

In plants, phenolic compounds have various functions, such as acting as an anti-predation mechanism by having a sharp taste, as an anti-pathogenic or as a protective agents (*e.g.* for UV light) (Shahidi & Naczk, 2004). Phenolic compounds also provide pigmentation in plants, serve as attractants for pollinators, make the cell walls impermeable to gas and water and contribute to the physical stability of the plant (Shahidi & Naczk, 2004). The phenolic compounds in plants are mainly found in conjugated form (with one or more mono- or polysaccharides bound to the phenolic groups) (Balasundram *et al.*, 2006).

In humans, phenolic compounds in the diet show antioxidative, antiinflammatory, anti-cancer, anti-diabetic, and cardioprotective properties, with suggested beneficial effects for human health (Perez-Jimenez *et al.*, 2009; Selma *et al.*, 2009; Ballard & Maróstica, 2019).The exact functions and target cells in the body are not determined in most cases, but the general results can be studied (Crozier *et al.*, 2009). Some suggested health benefits from ingesting phenolrich foods are an improvement of cardiovascular health (Wang *et al.*, 2011), a decreased risk of developing some forms of cancer (Kyle *et al.*, 2010) and a decreased mortality due to cancer (Ivey *et al.*, 2015) or in cardiovascular diseases (Manach *et al.*, 2005; Williamson 2017).

Many phenolic compounds have a low bioavailability, which means that they are not easily absorbed in the gastrointestinal tract in unaltered form. When the gut microbiota is fermenting the phenolic compounds, they produce smallersized phenolic acids, which have higher bioavailability and hence are more easily absorbed in the lumen of the colon (Selma *et al.*, 2009). Depending on the diversity of the gut microbiota, the fate of the phenolic compounds may differ. The enzymes needed for the transformation of the complex phenolic compounds into smaller molecules are largely species-dependent among the bacteria in the intestine and colon (Serreli & Deiana, 2019). These smaller molecules might act as signalling and regulating molecules that can have an impact on human health, even if their exact health benefits have yet to be determined (Serreli & Deiana, 2019).

The gut microbiota is also affected by phenolic compounds, as they change the amount and type of bacteria present in the gut (Saura-Calixto, 2011). Hence, phenolic compounds also have probiotic properties (Wang *et al.*, 2020).

1.2.3 Antioxidant dietary fibre - Together we stand?

Phenolic compounds have been found to bind easily to dietary fibre, and dietary fibre is believed to protect bound phenolic compounds from digestive enzymes, so that they reach the gut microbiota intact (Perez-Jimenez *et al.*, 2009; Palafox-Carlos *et al.*, 2011; Phan *et al.*, 2015). The complex consisting of dietary fibre and phenolic compounds is called *antioxidant dietary fibre* (Saura-Calixto, 1998, 2011). Phenolic compounds that move this way within the gastrointestinal tract, *i.e.* bound to the dietary fibre, may have a protective effect on the intestine by reducing the amount of free radicals in the lumen of the intestine before they are fermented by the gut microbiota (Saura-Calixto, 2011).

Some studies have shown that there may be complex interactions between phenolic compounds and other common compounds in foods, such as proteins (Foegeding *et al.*, 2017). These interactions stabilise the phenolic compounds and helps deliver them intact to the gut microbiota. For example, a recent study found that phenolic compounds bound to dietary fibre in kale entered the duodenum and small intestine, where the phenolic compounds were released from the dietary fibre in the presence of bile (Yang *et al.*, 2018). This increased the bioavailability of the phenolic compounds and potentially lowered the levels of cholesterol in the blood by trapping bile (which can be transformed into cholesterol by the liver) in the dietary fibre (Yang *et al.*, 2018). Pure phenolic compounds are digested by enzymes in the stomach, but studies examining pure phenolic compounds, either *in vitro* or *in vivo*, might not fully capture the complex ways in which plant-based material reacts in the gastrointestinal tract (Crozier *et al.*, 2009).

Since antioxidant dietary fibre is a new field, there is a need for more studies on how phenolic compounds bind to dietary fibre and how this can be of benefit to human health (Paper II).

1.3 Broccoli - an underutilised "superplant"

Broccoli (*Brassica oleracea* Italica group) belongs to the Brassicaceae family, together with *e.g.* cabbage, kale, cauliflower and Brussels sprouts. Broccoli most likely originated from the Mediterranean area (Maggioni, 2015), although the exact beginning of its cultivation is unclear (Gray, 1982). Some medieval sources mention a crop that might be broccoli, although it might also be a closely related species (Gray, 1989; Maggioni, 2015).

The usually eaten part of broccoli, the broccoli head with the florets, is also the most studied part of the plant. The florets are rich in glucosinolates, which is known to reduce the risk of chronic inflammation and the risk of some forms of cancer (Raiola *et al.*, 2018), and in sugars (Bhandari & Kwak, 2014, 2015). The florets also contain high levels of amino acids, iron, zinc and phosphorus (Liu *et al.*, 2018). The leaves of broccoli have been found to be rich in phenolic compounds (Bhandari & Kwak, 2014, 2015), as well as vitamin E and K, and the minerals magnesium and calcium (Liu *et al.*, 2018). The broccoli stems are rich in vitamin C, sugar (Bhandari & Kwak, 2014) and dietary fibre (Schäfer *et al.*, 2017). Hence, all the parts of the broccoli plant have a nutritional content that is interesting from a health perspective.



Figure 4: A broccoli plant divided into broccoli head (with florets), stem (with roots) and leaves. Photo: Emilia Berndtsson

As can be seen in Figure 4, the broccoli head constitutes only a minor part of the broccoli plant. Earlier studies have estimated that 70–90 % of the above-ground biomass (including florets, leaves and stem) of the broccoli plant is not

harvested, but instead becomes waste due to not reaching trading standards or because of a lack of market (Campas-Baypoli et al., 2009; Dominguez-Perles et al., 2010). An example of a harvested broccoli field in shown in Figure 5. Of the harvested broccoli florets, another 45-50 % becomes waste during processing (Campas-Baypoli et al., 2009). Some of the trading standards dictate that the broccoli head must be intact, clean, fresh in appearance and practically free from pests and pest damages, while the flower buds must be closed and tightly-grained and of uniform shape (UNECE, 2019). In terms of size, the head has to be maximum 20 cm in height and 6 cm minimum in diameter, and the ratio between head diameter and floral stem must be at least 2:1 (UNECE, 2019). Based on these standards, retailers can procure a products of a certain quality (e.g. colour, flavour and texture) without having to examine all units in detail (Mattsson, 2014). However, the trading standards are usually only the minimum requirements, as retailers and buyers may have higher expectations and requirements on the products. This increases the amount of waste, as products that would be safe to eat are discarded due to being the wrong size, shape or showing cosmetic damages. For other parts of the products, the consumer market may be more or less non-existent. If consumers were made aware of the possible



Figure 5: A harvested broccoli field, with remaining stems, leaves and discarded broccoli heads. Photo: Emilia Berndtsson

uses, they might also start to demand these product parts, which in turn would decrease the amount of field waste.

The standard regulations result in a huge amount of unnecessary waste of edible produce. Other reasons for field waste can be the weather conditions, *e.g.* the produce may be damaged by hailstorms or may fail to grow to the appropriate size due to sub-optimal temperature, and over-production, *i.e.* the producers grow a surplus to ensure that they have the contracted amount to deliver at the end of the season even if the growing conditions should prove to be sub-optimal (Strid *et al.*, 2014). Hence, there are considerable amounts of side streams that are available from broccoli production, and these could be used either as a raw material for functional food ingredients or as novel food products (Paper I).

In the year 2018, the total amount harvested in Swedish production of broccoli and cauliflower (combined into one group in data from FAOSTAT) was 9330 tonnes, as compared with 1.2 million tonnes in the US, 8.8 million tonnes in India, and 10.6 million tonnes in China. The world total was 26.5 million tonnes (*FAOSTAT*, 2020). Assuming that the biomass harvested and used as food is only 20 % of the total biomass, the annual waste in global broccoli and cauliflower production would amount to 106 million tonnes. This is a huge amount of biomass that could be used in a more sustainable way either as food or as raw material – if people only knew how to use it.

1.4 Side streams, food loss and food waste - confusing terms

In order to have a fruitful discussion about the possible uses of the different parts of the cultivated plant biomass, it is necessary to define commonly used terms. The terms *food loss* and *food waste* are sometimes used interchangeably and are unfortunately not always well defined in the literature, which makes it difficult to make comparisons between studies.



Figure 6: Expressions applied at different stages in the food supply to denote non-eaten produce.

Food loss can be defined as edible material removed in the beginning of the food supply chain (Figure 6), i.e. during production (at harvest), in post-harvest processing and in distribution (Gustavsson et al., 2011). This material will not reach the consumers. Food waste, on the other hand, can be defined as edible food discarded at the end of the food supply chain, *i.e.* at the retailer and consumer level (Gustavsson et al., 2011). Field waste can be defined as material left in the field after harvest. Side streams are defined as the parts of the cultivated plants that are not harvested for food uses but might have other potential uses (Figure 6). Examples of side streams in agricultural and horticultural production are straw from cereal production (where the grain for food production is the target material) and pomace after juice (e.g. apple) or oil (e.g. rapeseed) production. The term side stream can also cover overproduction or failure of the produce to reach cosmetic or safety standards (de Hooge et al., 2018). Although side streams are usually not used as food, they may have the potential to be valorised or refined into useful products. Side streams are usually not included in the calculations of food waste or food loss. This means that the total amount of biomass that could be used as food is considerably larger than earlier reported by e.g. FAO (2014).

1.5 Products from broccoli side streams?

Broccoli is known to be nutritious and healthy (Vasanthi *et al.*, 2009; Latte *et al.*, 2011), but is unfortunately not to everybody's taste. As a way of increasing the daily intake of broccoli, with associated potential health benefits, several studies have focused on the incorporation of broccoli into common, everyday food products (Paper I).

One study found that pasta with added broccoli florets (20 % w/w) had a higher content of glucosinolates than a durum wheat-based control pasta, and also had an acceptable taste according to a sensory panel (Silva *et al.*, 2013). Another study found that bread with an addition of broccoli florets (2 % w/w) had increased contents of protein and vitamin E, and also an increased antioxidant potential (Ranawana *et al.*, 2016).

Many gluten-free products are lacking vital nutrients and health-beneficial components, *e.g.* dietary fibre, vitamins, minerals and antioxidants (Han *et al.*, 2017). These products could benefit from incorporation of broccoli leaf powder, as it has been shown to increase the total phenolic content in wheat bread and to delay the staling process seven days post-baking, hence making the bread stay fresh longer (Lafarga *et al.*, 2019). Some research has shown that, when the bread-making process includes a thermal processing step, the phenolic compounds in broccoli leaves may be more resistant to bread-making than the

phenolic compounds found in broccoli stems (Lafarga *et al.*, 2019). Broccoli leaf powder has also been shown to increase the content of phenolic compounds in gluten-free sponge cake (Drabińska *et al.*, 2018).

Aside from food, broccoli side streams could be used as a component in animal feed. In a study where 20 % of dried broccoli side streams was added to the feed given to dairy cows, there was an improvement in the milk fat content, but no effect on the milk yield, milk protein content or total solids content (Yi *et al.*, 2015). Inclusion of broccoli stems and leaf meal in feed to laying hens has been shown to increase the carotenoid levels in the yolk, without any negative effect on the hens as the amounts of these components added to the diet is kept to a moderate level (Hu *et al.*, 2011; Pedroza *et al.*, 2018).

Side streams from broccoli cultivation can also be used as raw material for recovery of vegetable antioxidants (Aires *et al.*, 2017). These possess antioxidative activity comparable to that of synthetic antioxidants used in food products (Balasundram *et al.*, 2006).

Irrespective of the use of the broccoli side streams, *i.e.* as food, animal feed or as raw material for extraction of antioxidants, there is a need for more analyses of the material to find the most productive uses, but also the sustainability and profitability of the production and the health impacts. If a larger proportion of the biomass produced could be used as high-value products, this would increase profitability for the growers decrease the mass of waste generated.

2 Thesis aims

Today, only a minor part of the broccoli plant is used as food, namely the broccoli head or the florets. The remaining parts, stem and leaves, are not harvested but are instead left in the field. The florets are well-characterised in terms of content of vitamins, minerals, bioactive compounds and, to some extent, dietary fibre. This is not the case for the remaining parts of the plant. The overall aim of this thesis was thus to determine the content of dietary fibre and bioactive compounds in the side streams of broccoli.

Specific objectives of the work were to:

- Characterise the content of dietary fibre and phenolic compounds in broccoli leaves, and also to analyse the correlations between these two groups of compounds in matter of content in the leaves.
- Consider possible uses of broccoli side streams and the ethical implications of use of these side streams, especially since there is a need for a wiser management of limited resources such as arable land, water, fertilisers and energy.

3 Methods

3.1 Plant material

Broccoli was chosen as the study object in this thesis because of the large amounts of biomass left in the field in production and because of its high content of the health-beneficial compounds glucosinolates, which are being studied in other projects underway at SLU, focusing especially on the broccoli cultivar 'Beneforte'.

Broccoli leaves and stems of the cultivar 'Beneforte' from a commercial production site in north-western county of Scania (southern-most region of Sweden) were collected in two different fields in 2017 and two different fields in 2018. The leaves and stems were collected within 24 hours after the final harvest to minimise deterioration of the material. Each field is harvested by the grower 4-5 times, with a few days in between, in order to lift the broccoli heads at optimal size. The leaves and stems analysed in this thesis were collected within 24 hours of the final harvest, to minimise deterioration of the material. For this, three squares (1.5 m x 1.5 m) were randomly positioned on the field (excluding edges) as described previously (Strid et al., 2014), and 10 broccoli plants in each square were cut 2 cm above the ground to exclude the roots and the most woody section of the stem. The plants were then transported to the laboratory in plastic bags, washed under running water to rinse away any visible dirt, air-dried and separated into leaves and stems. The whole leaves (including midvein and petiole) were then placed pairwise in bags. The stems were divided into three parts (top, middle and lower) and placed in bags. The middle part of the stem was used in this project. Leaves and stem parts were stored at -80 °C, and were freeze-dried and milled into a powder prior to analysis (see Paper II).

3.2 Soluble and insoluble dietary fibre

There are several methods available for analysing dietary fibre. One feature all these methods have in common is that they try to mimic the conditions in the gastrointestinal tract in humans, due to the definition of dietary fibre.

Dietary fibre content was measured in this thesis according to one of the standard methods available (Theander *et al.*, 1995), with modifications according to Andersson *et al.* (1999) for separate analysis of soluble and insoluble dietary fibre components (Paper II). The broccoli powder was treated with enzymes to extract the dietary fibre, and the soluble and insoluble fibre were separately hydrolysed in order to digest the fibre to its building blocks, *i.e.* sugar residues, uronic acids and Klason lignin, before analysis with gas chromatography, colourimetry and gravimetry, respectively.

3.3 Quantification and identification of phenolic compounds

Phenolic compounds were extracted from the powdered broccoli samples using methanol, and the total phenolic content in broccoli leaves and stems was measured according to Singleton & Rossi (1965), with some modifications (Gao *et al.*, 2000; Dewanto *et al.*, 2002). The methanol extract of broccoli leaves and stems was diluted 10-fold to appropriate concentrations to fit the range of the standard curve.

To quantify and tentatively identify the phenolic compounds in broccoli leaves, the methanol extract and the methanol extract treated with alkaline hydrolysis (to liberate the phenolic acids) were analysed with a highperformance liquid chromatography-mass spectrometry (HPLC-MS) system according to Lin *et al.* (2008) (Paper II).

3.4 Relationships between phenolic compounds and dietary fibre

Dietary fibre and phenolic compounds were analysed in aliquots of the same samples to study the possible relationships between these two groups of compounds, *i.e.* whether there were high concentrations of phenolic compounds when dietary fibre was present in high concentrations, as some phenolic compounds have been suggested to bind to dietary fibre. Principal component analysis (PCA) was performed to visualise the relationship between the content of dietary fibre and content of phenolic compounds (Paper II).

3.5 Field waste in commercial broccoli production (pilot study)

Few studies have examined the amount of field waste in broccoli production. Some studies have examined waste in field and in greenhouse production (Campas-Baypoli *et al.*, 2009; Dominguez-Perles *et al.*, 2010), but no previous studies has measured broccoli field waste under Swedish conditions. Hence, in this thesis the amount of field waste in a commercial broccoli field was measured between two consecutive harvests in August 2018. For this, three squares (1.5 m x 1.5 m) were randomly positioned on the field (excluding edges) as described previously (Strid *et al.*, 2014), and 10 broccoli plants in each square were cut 2 cm above the ground, weighed, and then divided into different fractions (broccoli head, leaves and stem) which were individually weighed. The mean weight per 2.25 m² square for each fraction and for the whole plants was calculated.

4 Results and discussion

4.1 Dietary fibre and phenolic content in broccoli side streams

4.1.1 Dietary fibre

Broccoli stem and broccoli leaves both had a high content of dietary fibre (Table 1), with levels comparable to those reported for broccoli florets, cabbage leaves and kale leaves (Table 2). The content of insoluble dietary fibre (IDF), soluble dietary fibre (SDF) and total dietary fibre (TDF) in the stems and the leaves did not significantly differ between the study years. However, some of the dietary fibre constituents (the sugar residues insoluble (Insol) uronic acid (UA), Insol arabinose (ara), Insol mannose (man), soluble (Sol) fuctose (fuc), Sol xylose (xyl), Sol man and Sol glucose (glc)) differed significantly between the study years, with higher levels of the insoluble sugar residues and lower levels of soluble sugar residues in 2017 than in 2018 (Paper II). The summer of 2018 was an exceptionally warm and dry in Sweden, with a maximum temperature of 28.6 °C and mean temperature 16.4 °C, as compared with 20.8 °C and 14.3 °C respectively, in 2017 (Swedish Meteorological and Hydrological Institute (SMHI), online data). Weather conditions might thus have had affected the content of dietary fibre in the broccoli leaves and stems, but more studies are needed to confirm this.

Table 1: Content of dietary fibre (insoluble dietary fibre (IDF), soluble dietary fibre (SDF) and total dietary fibre (TDF) in different broccoli parts sampled in the two study years.

	IDF	SDF	TDF
	[% of dry weight]	[% of dry weight]	[% of dry weight]
Stem, 2017	$33.2\pm6.0^{\text{b}}$	2.3 ± 0.4^{ab}	35.2 ± 5.8^{b}
Leaves, 2017	$28.6\pm4.1^{\text{ab}}$	$2.0\pm0.5^{\rm a}$	30.1 ± 4.3^{ab}
Stem, 2018	34.4 ± 5.3^{b}	$2.5\pm0.3^{\text{b}}$	$37.0\pm5.1^{\text{b}}$
Leaves, 2018	$24.6\pm1.5^{\rm a}$	$2.1\pm0.2^{\rm a}$	$26.7\pm1.5^{\rm a}$

Data is expressed as mean \pm SD. Values within columns followed by different letters differs significantly (P < 0.05) according to the Tukey post hoc test

Source	Mean [% of dry weight]	References	
Onion	47.2	(Kalala et al., 2018)	
Kale leaves	42.7	(Thavarajah et al., 2019)	
Cabbage outer leaves	40.9	(Tanongkankit et al., 2012)	
Broccoli florets	36.0	(Kalala et al., 2018)	
Broccoli stem	35-36	Paper II	
Brussels sprout	34.1	(Kahlon et al., 2007)	
Broccoli leaves	26-32	Paper II	
Cauliflower (curd)	29.7	(Kalala et al., 2018)	
Spinach	27.1	(Kahlon et al., 2007)	
Carrot	24.1	(Theander et al., 1995)	
Green peas	16.7	(Theander et al., 1995)	

Table 2: Levels of total dietary fibre in some vegetables and vegetable parts.

4.1.2 Phenolic compounds

The total content of phenolic compounds, measured with Folin-Ciocalteu reagent as gallic acid equivalents (GAE) varied significantly between the different broccoli plant parts in the samples from 2018, with higher levels in the leaves than in the stem (Table 3). There was no significant difference between the fields, which were managed in the same way by the same grower.

	mg GAE/g dry weight
Field 1, stem	$4.29\pm0.93^{\rm a}$
Field 1, leaves	$5.71 \pm 1.11^{\text{b}}$
Field 2, stem	$3.73\pm0.13^{\rm a}$
Field 2, leaves	6.74 ± 0.87^{b}

Table 3: Total content of phenolic compounds (assessed as gallic acid equivalents (GAE) in broccoli leaves and stems collected from two different fields in southern Sweden in 2018.

Data expressed as mean \pm SD. Values followed by different letter differs significantly (P < 0.05) according to the Tukey post hoc test. Total phenolic content measured with Folin-Ciocalteu reagent.

The Folin-Ciocalteu method is a quick way to get an overview of the total amount of phenolic compounds, but it has been criticised because it measures all compounds that are reactive with the reagent (*i.e.* that have an antioxidative effect), *e.g.* vitamin C, and not only phenolic compounds (Everette *et al.*, 2010). Since broccoli is rich in vitamin C, this might influence the results when this method is used, giving higher values than with a method measuring only the phenolic compounds. The advantage of analysing phenolic compounds with the HPLC-MS system is the possibility to select the conditions in which the phenolic compounds are easiest to separate from other compounds, *e.g.* by altering separating column or solvent ratio.

The HPLC-MS analysis (Olsen *et al.*, 2009) showed high levels of phenolic compounds, mainly from the groups flavonoids and phenolic acids, in the broccoli leaves (Paper II). The levels found in broccoli leaves were higher than those found in broccoli florets and comparable to those reported for kale leaves (Table 4). Kale leaves have recently attracted attention for their high content of health beneficial compounds (Becerra-Moreno *et al.*, 2014; Šamec *et al.*, 2018). The phenolic content was significantly higher during 2017 than in 2018, which might have been due to differences in weather conditions between years.

Source	mg/g dry weight	Reference
Broccoli leaves, 2017	10.8-15.2	Paper II
Kale leaves	10.6	Olsen et al. (2009)
Broccoli leaves, 2018	6.3–7.5	Paper II
Broccoli florets	1.7–2.2	Torres-Contreras et al. (2017)

 Table 4: Content of phenolic compounds determined by methanol extraction in different broccoli
 plant parts and in kale leaves

The impact of the weather on the results was evident in the PCA plot (Figure 7). In the PCA score plot, samples clustered into three groups, with the samples from 2017 separated into two groups, whereas the samples from 2018 clustered quite closely together (Figure 7a). Samples from Field 3 and Field 4 showed positive values on the first principal component (PC1, x-axis), indicating higher levels of phenolic acids (orange triangles) and of the four soluble fibre compounds Sol xyl, Sol fuc, Sol glc, and Sol man when the loading plot and score plot are analysed together (Figure 7a and Figure 7b). Samples from Field 1 clustered together with negative values on the second principal component (PC2, y-axis), indicating lower content of phenolic acids and higher levels of phenolic compounds (green rectangles) and dietary fibre (blue circles), with larger variation in the content of the dietary fibre. Samples from Field 2 showed negative values on the second principal component, which indicates high levels of phenolic compounds and lower levels of dietary fibre (Figure 7a).

Since this study only measured the content of phenolic compounds extractable with an organic solvent, and not those that are strongly bound to the dietary fibre (Phan *et al.*, 2015, 2017), it was not surprising that there was no significant relationship between the dietary fibre and the complex phenolic compounds. However, after treating the complex phenolic compounds with an alkaline hydrolysis to free the smaller phenolic acids, the PCA loading plot indicated significant relationships between some phenolic acids and the soluble dietary fibre (Figure 7b).

Some of the soluble fibre compounds (Sol fuc, Sol xyl, Sol man, Sol glc) were positioned close together with phenolics acids in the bottom right-hand corner, indicating that these compounds can be found together in broccoli leaves. Two of the phenolic compounds from the methanol extract (I and K) also showed correlation with the same phenolic acids. For the insoluble dietary fibre (blue circles), most had high positive values on the first principal component and close to zero on the second principal component, showing no clear correlation to the phenolic compounds (Figure 7b). This indicates that the phenolic compounds

that are bound to the insoluble dietary fibre were not released during the methanol extraction.

Most of the phenolic compounds from the methanol extraction had negative values, indicating a negative relationship with the phenolic acids since they were located at opposite corners in the PCA loading plot (Figure 7b).



(b)

Figure 7: Principal component analysis of relationship between dietary fibre constituents and phenolic compounds in broccoli leaves (a) score plot and (b) loading plot. Sol: soluble dietary fibre constituent. Insol: insoluble dietary fibre constituent. Source: Paper II

The HPLC analysis of phenolic compounds in the broccoli stems gave no useable results because to the amounts of phenolic compounds extracted with methanol from the stem were too low. This might be because the levels of phenolic compounds in the plant material were too low, or because the phenolic compounds were too strongly bound to the dietary fibre in the stem cell walls. Phenolic compounds have been shown to bind readily to bacterial cellulose (Phan *et al.*, 2015), making it difficult to extract the phenolic compounds with only an organic solvent system. Some previous studies have employed enzymatic extraction of phenolic compounds from broccoli inflorescences (Wu *et al.*, 2015) which might be a more efficient method when analysing phenolic compounds in broccoli stem.

4.1.3 Possible uses

Broccoli stems and the broccoli leaves both proved to be a rich source of dietary fibre. Most consumers eat too little fibre in their everyday diet. At the same time, there is a huge market for kale leaves, with their associated health benefits. Since broccoli leaves have a comparable content of dietary fibre and phenolic compounds and are closely related to kale, it seems possible to use broccoli leaves in a similar way. If the broccoli leaves were to be used, it would improve the economic situation for the growers, as more of the biomass produced could be used as a high value product. Broccoli stem and leaves might also be used as a raw material for extracting health beneficial ingredients, *e.g.* in a biorefinery (Paper I).

One important question that has to be answered is how to harvest the leaves in an efficient way, and more studies are needed to gain knowledge in this research area.

4.2 Field waste in the broccoli fields (pilot study)

4.2.1 Amount left in the field at harvest

On measuring the different fractions of the broccoli plant left in a commercial field between harvests, it became clear that the broccoli plants differed in size even though they were planted at the same time and belonged to the same cultivar ('Beneforte'). Comparison of the mean weight of the different plant parts and the whole plant showed leaves and stems make up the majority of the weight (76-87%) in two of the three sampling squares studied, while heads made

up the major part of the weight (57 %) in the third square (Table 5). Thus a large proportion of the biomass produced was composed of leaves and stems. Considering the high levels of bioactive compounds and dietary fibre in broccoli leaves, it is indeed a misuse of resources not to use them to a larger extent.

 Table 5: Measured weight [kg] of separate parts of broccoli plant and of the whole broccoli plant the three sampling squares used in the pilot study on field waste.

	Head	Leaves	Stem	Whole plant
Square 1	0.14 ± 0.08	0.65 ± 0.14	0.17 ± 0.03	0.94 ± 0.18
Square 2	0.16 ± 0.07	0.14 ± 0.04	0.46 ± 0.21	0.82 ± 0.42
Square 3	0.79 ± 0.19	0.30 ± 0.19	0.28 ± 0.04	1.37 ± 0.34

Data expressed as mean \pm SD

However, it is important to remember that there might be consequences of removing too much crop biomass from the field. Any intensification of food production that leads to the removal of more plant products from the fields will decrease the content of the organic carbon content in the soil, which in turn may have a negative impact on soil biodiversity and productivity (Kopittke *et al.*, 2019). Hence, there must be a balance between the organic material removed from the field and the organic material returned to the field. One solution could be to harvest the broccoli leaves and use them as food or raw material for extraction of health beneficial compounds, and leave the stems on the field as a green fertiliser.

4.3 Ethics

Among the 17 sustainable development goals (SDG) established by the United Nations (https://sustainabledevelopment.un.org/), at least three have a clear connection to the use of side streams in food production:

- Zero hunger (SDG 2): food production today already provides enough food for the whole global population, without increasing the use of water, arable land and/or fertilisers. However, with the looming threat of a climate change and the consequences of that for the food production (Masson-Delmotte *et al.*, 2018), it is crucial to use as much of the food crop biomass as possible, instead of discarding food that could be eaten, especially since food insecurity is a problem for about 2 billion people in different parts of the world (FAO *et al.*, 2019).
- Responsible consumption and production (SDG 12): Sustainable development includes "doing more and better with less", and also reducing the use of resources while still achieving an increase in welfare

and the economy. To achieve this, there is a need for changes along the whole food supply chain, from the producers to the consumers, with regard to using all the biomass produced. There is possibly also to need to change the food supply chain into a more circular economy to use more of the crop biomass produced (HLPE, 2014; Imbert, 2017).

Climate action (SDG 13): One of the major tasks in preventing a global climate catastrophe is to reduce the emissions of greenhouse gases. Using more of the crop biomass produced as food might lead to a reduced need to expand the area used for food production and a reduced need for fertiliser and/or soil management (Kummu *et al.*, 2012). Hence, a reduction in food waste would have a reducing impact on the greenhouse gas emissions and on global warming (Kummu *et al.*, 2012; Oldfield *et al.*, 2016).

Further research is needed on crops such as broccoli to demonstrate that there are possible alternative uses for the biomass produced that might help meet different sustainable development goals, and to prevent valuable resources from becoming waste instead of becoming food.

But why is so much edible biomass becoming waste, at the same time as an enormous number of people do not have sufficient food of appropriate quality to eat? In industrialised countries, the main reasons for waste in the food supply chain are over-production, premature harvesting, high aesthetic standards, high cost for valorising trimmings, large amounts of produce on display in stores, and food waste in households (Gustavsson *et al.*, 2011). In developing countries, on the other hand, the main reasons for waste in the food supply chain are premature harvesting, poor storage facilities and lack of infrastructure, processing facilities and markets (Gustavsson *et al.*, 2011). In both types of countries, the man factors in waste generation need to be handled in order to reduce the amount of waste and loss in the whole food supply chain, from field to fork and bin.

5 Conclusions

- Broccoli leaves are rich in dietary fibre and in phenolic compounds, with levels comparable to those in kale leaves.
- There are correlations between dietary fibre and some phenolic acids in the broccoli leaves were found, indicating that these co-occur in the broccoli leaves and possibly also binds to one another.
- It is difficult to extract phenolic compounds from the broccoli stems using methanol as the solvent, possibly due to the fact that the phenolic compounds bind tightly to the insoluble dietary fibre.
- There are huge side streams in broccoli production. If more of the biomass in the broccoli production (leaves, stems) could be used, this would increase the economic gain for the growers and make more food available, without increasing the use of limited resources such as land, water and fertilisers.
- Potential future uses of broccoli leaves and stems are as functional food ingredients to increase the nutritional value or technological properties of food products, or as a raw material to extract health-beneficial compounds.

6 Future perspectives

Based on the information presented in this thesis, and reported in earlier studies, broccoli leaves seem to be a possible valuable resource that is currently underutilised. Broccoli stems are rich in dietary fibre, but need further evaluation in terms of their content of phenolic compounds.

The results in this thesis indicates that there are interactions between the content of dietary fibre and phenolic compounds in broccoli leaves, and thus that broccoli leaves might be a good raw material for making food ingredients or for extracting health-beneficial compounds. Hence, there are possibilities to use the broccoli leaves in the food industry, but first there are knowledge gaps to be filled. More characterisation work is needed on dietary fibre and phenolic compounds in broccoli plant parts and the interaction between these. More studies that combine biology/agronomy and medical expertise are also needed, to evaluate the health benefits of broccoli leaves and how they are affected by different factors during growing and post-harvest handling. It has been shown that the content of phenolic compounds in curly kale is dependent on cultivar and on climate factors such as temperature and solar radiation during the growing season (Schmidt et al., 2010) and also on the amount of nitrogen fertiliser used (Neugart et al., 2018). This indicates that the content of phenolic compounds might be different in other broccoli cultivars, in other fields and/or with other growing practices.

Appropriate methods for extraction of phenolic compounds from the broccoli stem needs to be identified, since phenolic compounds and dietary fibre interact and bind to each other inside the plant material.

The world we live in is changing. With future climate change there will be a need to alter the way in which we produce food and the way in which we handle food. Food waste, food loss and field waste are luxuries that we cannot afford if we want to reduce the number of undernourished people on Earth and use the limited resources available in a responsible way.

References

- Aires, A., Carvalho, R. & Saavedra, M.J. (2017). Reuse potential of vegetable wastes (broccoli, green bean and tomato) for the recovery of antioxidant phenolic acids and flavonoids. *International Journal of Food Science & Technology*, vol. 52 (1), pp. 98–107
- Alexandratos, N. & Bruinsma, J. (2012). World agriculture towards 2030/2050: the 2012 revision. (ESA Working Paper No. 12-03). Food and Agriculture Organization of the United Nations. Available at: http://www.fao.org/economic/esa
- Andersson, A.A.M., Merker, A., Nilsson, P., Sørensen, H. & Åman, P. (1999). Chemical composition of the potential new oilseed crops Barbarea vulgaris, Barbarea verna and Lepidium campestre. *Journal of the Science of Food and Agriculture*, vol. 79 (2), pp. 179– 186
- Balasundram, N., Sundram, K. & Samman, S. (2006). Phenolic compounds in plants and agriindustrial by-products: Antioxidant activity, occurrence, and potential uses. *Food Chemistry*, vol. 99 (1), pp. 191–203
- Ballard, C.R. & Maróstica, M.R. (2019). Health Benefits of Flavonoids. In: Campos, M.R.S. (ed.) *Bioactive Compounds*. Cambridge, UK: Woodhead Publishing, pp. 185–201.
- Becerra-Moreno, A., Alanis-Garza, P.A., Luis Mora-Nieves, J., Pablo Mora-Mora, J. & Jacobo-Velazquez, D.A. (2014). Kale: An excellent source of vitamin C, pro-vitamin A, lutein and glucosinolates. *Cyta-Journal of Food*, vol. 12 (3), pp. 298–303
- den Besten, G., van Eunen, K., Groen, A.K., Venema, K., Reijngoud, D.-J. & Bakker, B.M. (2013). The role of short-chain fatty acids in the interplay between diet, gut microbiota, and host energy metabolism. *Journal of Lipid Research*, vol. 54 (9), pp. 2325–2340
- Bhandari, S.R. & Kwak, J.-H. (2014). Seasonal variation in phytochemicals and antioxidant activities in different tissues of various Broccoli cultivars. *African Journal of Biotechnology*, vol. 13 (4), pp. 604–615
- Bhandari, S.R. & Kwak, J.-H. (2015). Chemical Composition and Antioxidant Activity in Different Tissues of Brassica Vegetables. *Molecules*, vol. 20 (1), pp. 1228–1243
- Brett, C. & Waldron, K. (1996). Physiology and biochemistry of plant cell walls. Second edition. London: Chapman & Hall.
- Bryngelsson, D., Wirsenius, S., Hedenus, F. & Sonesson, U. (2016). How can the EU climate targets be met? A combined analysis of technological and demand-side changes in food and agriculture. *Food Policy*, vol. 59, pp. 152–164

- Campas-Baypoli, O.N., Sanchez-Machado, D.I., Bueno-Solano, C., Nunez-Gastelum, J.A., Reyes-Moreno, C. & Lopez-Cervantes, J. (2009). Biochemical composition and physicochemical properties of broccoli flours. *International Journal of Food Sciences and Nutrition*, vol. 60, pp. 163–173
- Cartea, M.E., Francisco, M., Soengas, P. & Velasco, P. (2011). Phenolic Compounds in Brassica Vegetables. *Molecules*, vol. 16 (1), pp. 251–280
- Codex Alimentarius (2017). Guidelines on nutrition labelling, CAC/GL 2-1985, last revisioned in 2017. (Codex Alimentarius International Food Standards, CAC/GL 2-1985). Rome: Joint FAO/WHO. Available at: http://www.fao.org/fao-who-codexalimentarius/shproxy/es/?lnk=1&url=https%253A%252F%252Fworkspace.fao.org%252FSites%252Fcodex %252FStandards%252FCAC%2BGL%2B2-1985%252FCXG 002e.pdf
- Corrêa-Oliveira, R., Fachi, J.L., Vieira, A., Sato, F.T. & Vinolo, M.A.R. (2016). Regulation of immune cell function by short-chain fatty acids. *Clinical & Translational Immunology*, vol. 5 (4), p. e73
- Crozier, A., Jaganath, I.B. & Clifford, M.N. (2009). Dietary phenolics: chemistry, bioavailability and effects on health. *Natural Product Reports*, vol. 26 (8), pp. 1001–1043
- Dewanto, V., Wu, X., Adom, K.K. & Liu, R.H. (2002). Thermal Processing Enhances the Nutritional Value of Tomatoes by Increasing Total Antioxidant Activity. *Journal of Agricultural and Food Chemistry*, vol. 50 (10), pp. 3010–3014
- Dominguez-Perles, R., Carmen Martinez-Ballesta, M., Carvajal, M., Garcia-Viguera, C. & Moreno, D.A. (2010). Broccoli-Derived By-Products-A Promising Source of Bioactive Ingredients. *Journal of Food Science*, vol. 75 (4), pp. C383–C392
- Drabińska, N., Ciska, E., Szmatowicz, B. & Krupa-Kozak, U. (2018). Broccoli by-products improve the nutraceutical potential of gluten-free mini sponge cakes. *Food Chemistry*, vol. 267, pp. 170–177 (1st Food Chemistry Conference: Shaping the future of food quality, health and safety)
- Everette, J.D., Bryant, Q.M., Green, A.M., Abbey, Y.A., Wangila, G.W. & Walker, R.B. (2010). Thorough Study of Reactivity of Various Compound Classes toward the Folin–Ciocalteu Reagent. *Journal of Agricultural and Food Chemistry*, vol. 58 (14), pp. 8139–8144
- FAO (2014). *Mitigation of food wastage societal costs and benefits*. Rome: Food and Agriculture organization of the United Nation (FAO).
- FAO, IFAD, UNICEF, WFP & WHO (2017). The State of Food Security and Nutrition in the World 2017. Building resilience for peace and food security. Rome: Food and Agriculture organization of the United Nation (FAO).
- FAO, IFAD, UNICEF, WFP & WHO (2018). *The State of Food Security and Nutrition in the World 2018. Building climate resilience for food security and nutrition.* Rome: FAO.
- FAO, IFAD, UNICEF, WFP & WHO (2019). The State of Food Security and Nutrition in the World 2019. Safeguarding against economic slowdowns and downturns. Rome: FAO.
- FAOSTAT (2020-02-06). Available at: http://www.fao.org/faostat/en/#data/QC [2020-02-17]
- Foegeding, E.A., Plundrich, N., Schneider, M., Campbell, C. & Lila, M.A. (2017). Proteinpolyphenol particles for delivering structural and health functionality. *Food Hydrocolloids*, vol. 72, pp. 163–173

- Food and Nutrition Board (2001). *Dietary Reference Intakes Proposed Definition of Dietary Fiber*. Washington D.C: Food and Nutrition Board, Institude of Medicine.
- Gao, X., Björk, L., Trajkovski, V. & Uggla, M. (2000). Evaluation of antioxidant activities of rosehip ethanol extracts in different test systems. *Journal of the Science of Food and Agriculture*, vol. 80 (14), pp. 2021–2027
- Gray, A.R. (1982). Taxonomy and Evolution of Broccoli (Brassica oleracea var. italica). *Economic Botany*, vol. 36/1982 (4), pp. 397–410
- Gray, A.R. (1989). Taxonomy and evoluation of broccolis and cauliflowers. *Ithaca: L. H. Bailey Hortorium, New York State College of Agriculture and Life Sciences, Cornell University*, vol. 23 (1), pp. 28–46
- Gustavsson, J., Cederberg, C. & Sonesson, U. (2011). Global food losses and food waste: extent, causes and prevention ; study conducted for the International Congress Save Food! at Interpack 2011, [16 - 17 May], Düsseldorf, Germany. Rome: Food and Agriculture Organization of the United Nations.
- Han, W., Ma, S., Li, L., Wang, X.-X. & Zheng, X.-L. (2017). Application and Development Prospects of Dietary Fibers in Flour Products. *Journal of Chemistry*, p. 2163218
- Hipsley, E.H. (1953). Dietary 'fibre' and pregnacy toxaemia. *British Medical Journal*, vol. Aug, 1953 (22)
- HLPE (2014). Food losses and waste in the context of sustainable food systems. Rome: High Level Panel of Experts on Food Security and Nutrition of the Committee on World Food Security.
- de Hooge, I.E., van Dulm, E. & van Trijp, H.C.M. (2018). Cosmetic specifications in the food waste issue: Supply chain considerations and practices concerning suboptimal food products. *Journal of Cleaner Production*, vol. 183, pp. 698–709
- Hu, C.H., Zuo, A.Y., Wang, D.G., Pan, H.Y., Zheng, W.B., Qian, Z.C. & Zou, X.T. (2011). Effects of broccoli stems and leaves meal on production performance and egg quality of laying hens. *Animal Feed Science and Technology*, vol. 170 (1–2), pp. 117–121
- Imbert, E. (2017). Food waste valorization options: opportunities from the bioeconomy. Open Agriculture, vol. 2 (1). DOI: https://doi.org/10.1515/opag-2017-0020
- Ivey, K.L., Hodgson, J.M., Croft, K.D., Lewis, J.R. & Prince, R.L. (2015). Flavonoid intake and all-cause mortality. *The American Journal of Clinical Nutrition*, vol. 101 (5), pp. 1012–1020
- Kahlon, T.S., Chapman, M.H. & Smith, G.E. (2007). In vitro binding of bile acids by spinach, kale, brussels sprouts, broccoli, mustard greens, green bell pepper, cabbage and collards. *Food Chemistry*, vol. 100 (4), pp. 1531–1536
- Kalala, G., Kambashi, B., Everaert, N., Beckers, Y., Richel, A., Pachikian, B., Neyrinck, A.M., Delzenne, N.M. & Bindelle, J. (2018). Characterization of fructans and dietary fibre profiles in raw and steamed vegetables. *International Journal of Food Sciences and Nutrition*, vol. 69 (6), pp. 682–689
- Kim, Y. & Je, Y. (2016). Dietary fibre intake and mortality from cardiovascular disease and all cancers: A meta-analysis of prospective cohort studies. *Archives of Cardiovascular Diseases*, vol. 109 (1), pp. 39–54

- Kopittke, P.M., Menzies, N.W., Wang, P., McKenna, B.A. & Lombi, E. (2019). Soil and the intensification of agriculture for global food security. *Environment International*, vol. 132, p. 105078
- Kummu, M., de Moel, H., Porkka, M., Siebert, S., Varis, O. & Ward, P.J. (2012). Lost food, wasted resources: Global food supply chain losses and their impacts on freshwater, cropland, and fertiliser use. *Science of the Total Environment*, vol. 438, pp. 477–489
- Kyle, J.A.M., Sharp, L., Little, J., Duthie, G.G. & McNeill, G. (2010). Dietary flavonoid intake and colorectal cancer: a case-control study. *The British Journal of Nutrition*, vol. 103 (3), pp. 429–436
- Lafarga, T., Gallagher, E., Bademunt, A., Viñas, I., Bobo, G., Villaró, S. & Aguiló-Aguayo, I. (2019). Bioaccessibility, physicochemical, sensorial, and nutritional characteristics of bread containing broccoli co-products. *Journal of Food Processing and Preservation*, vol. 43 (2)
- Latte, K.P., Appel, K.-E. & Lampen, A. (2011). Health benefits and possible risks of broccoli -An overview. *Food and Chemical Toxicology*, vol. 49 (12), pp. 3287–3309
- Li, F., Hullar, M.A.J., Schwarz, Y. & Lampe, J.W. (2009). Human Gut Bacterial Communities Are Altered by Addition of Cruciferous Vegetables to a Controlled Fruit- and Vegetable-Free Diet. *Journal of Nutrition*, vol. 139 (9), pp. 1685–1691
- Lin, L.-Z., Chen, P. & Harnly, J.M. (2008). New Phenolic Components and Chromatographic Profiles of Green and Fermented Teas. *Journal of Agricultural and Food Chemistry*, vol. 56 (17), pp. 8130–8140
- Liu, M., Zhang, L., Ser, S.L., Cumming, J.R. & Ku, K.-M. (2018). Comparative phytonutrient analysis of broccoli by-products: The potentials for broccoli by-product utilization. *Molecules*, vol. 23 (4), p. 18
- Mackie, A., Bajka, B. & Rigby, N. (2016). Roles for dietary fibre in the upper GI tract: The importance of viscosity. *Food Research International*, vol. 88, pp. 234–238
- Maggioni, L. (2015). Domestication of Brassica oleracea L. (PhD Thesis). Swedish University of Agricultural Sciences. Available at: https://pub.epsilon.slu.se/12424/ [2017-12-13]
- Makki, K., Deehan, E.C., Walter, J. & Bäckhed, F. (2018). The Impact of Dietary Fiber on Gut Microbiota in Host Health and Disease. *Cell Host & Microbe*, vol. 23 (6), pp. 705–715
- Manach, C., Williamson, G., Morand, C., Scalbert, A. & Rémésy, C. (2005). Bioavailability and bioefficacy of polyphenols in humans. I. Review of 97 bioavailability studies. *The American Journal of Clinical Nutrition*, vol. 81 (1), pp. 230S-242S
- Mandimika, T., Paturi, G., De Guzman, C.E., Butts, C.A., Nones, K., Monro, J.A., Butler, R.C., Joyce, N.I., Mishra, S. & Ansell, J. (2012). Effects of dietary broccoli fibre and corn oil on serum lipids, faecal bile acid excretion and hepatic gene expression in rats. *Food Chemistry*, vol. 131 (4), pp. 1272–1278
- Masson-Delmotte, V.P., Pörter, H.-O., Roberts, D., Skea, J., Shukla, P.R., Pirani, A., Moufouma-Okia, W., Péan, C., Pidcock, R., Connors, S., Matthews, J.B.R., Chen, Y., Zhou, X., Gomis, M.I., Lonnoy, E., Maycock, T., Tignor, M. & Waterfields, T. (2018). Global Warming of 1.5°C. An IPCC Special Report on the impacts of global warming of 1.5°C above preindustrial levels and related global greenhouse gas emission pathways, in the context of strengthening the global response to the threat of climate change, sustainable development,

and efforts to eradicate poverty. In press: The Intergovernmental Panel on Climate Change (IPCC). Available at: https://www.ipcc.ch/sr15/

- Mattsson, K. (2014). Vi slänger frukt och grönsaker i onödan varför? (2014:5). Jönköping: Jordbruksverket.
- Miki, T., Eguchi, M., Kurotani, K., Kochi, T., Kuwahara, K., Ito, R., Kimura, Y., Tsuruoka, H., Akter, S., Kashino, I., Kabe, I., Kawakami, N. & Mizoue, T. (2016). Dietary fiber intake and depressive symptoms in Japanese employees: The Furukawa Nutrition and Health Study. *Nutrition*, vol. 32 (5), pp. 584–589
- Nawirska, A. & Kwasniewska, M. (2005). Dietary fibre fractions from fruit and vegetable processing waste. *Food Chemistry*, vol. 91 (2), pp. 221–225
- Neugart, S., Baldermann, S., Hanschen, F.S., Klopsch, R., Wiesner-Reinhold, M. & Schreiner, M. (2018). The intrinsic quality of brassicaceous vegetables: How secondary plant metabolites are affected by genetic, environmental, and agronomic factors. *Scientia Horticulturae*, vol. 233, pp. 460–478
- Oldfield, T.L., White, E. & Holden, N.M. (2016). An environmental analysis of options for utilising wasted food and food residue. *Journal of Environmental Management*, vol. 183, pp. 826–835
- Olsen, H., Aaby, K. & Borge, G.I.A. (2009). Characterization and Quantification of Flavonoids and Hydroxycinnamic Acids in Curly Kale (Brassica oleracea L. Convar. acephala Var. sabellica) by HPLC-DAD-ESI-MSn. *Journal of Agricultural and Food Chemistry*, vol. 57 (7), pp. 2816–2825
- Palafox-Carlos, H., Ayala-Zavala, J.F. & Gonzalez-Aguilar, G.A. (2011). The Role of Dietary Fiber in the Bioaccessibility and Bioavailability of Fruit and Vegetable Antioxidants. *Journal* of Food Science, vol. 76 (1), pp. R6–R15
- Pedroza, G., Famula, T. & King, A. (2018). Broccoli meal fed to laying hens increases nutrients in eggs and deepens the yolk color. *California Agriculture*, vol. 72 (4), pp. 243–247
- Perez-Jimenez, J., Serrano, J., Tabernero, M., Arranz, S., Diaz-Rubio, M.E., Garcia-Diz, L., Goni, I. & Saura-Calixto, F. (2009). Bioavailability of Phenolic Antioxidants Associated with Dietary Fiber: Plasma Antioxidant Capacity After Acute and Long-Term Intake in Humans. *Plant Foods for Human Nutrition*, vol. 64 (2), pp. 102–107
- Phan, A.D.T., Flanagan, B.M., D'Arcy, B.R. & Gidley, M.J. (2017). Binding selectivity of dietary polyphenols to different plant cell wall components: Quantification and mechanism. *Food Chemistry*, vol. 233, pp. 216–227
- Phan, A.D.T., Netzel, G., Wang, D., Flanagan, B.M., D'Arcy, B.R. & Gidley, M.J. (2015). Binding of dietary polyphenols to cellulose: Structural and nutritional aspects. *Food Chemistry*, vol. 171, pp. 388–396
- Raiola, A., Errico, A., Petruk, G., Monti, D.M., Barone, A. & Rigano, M.M. (2018). Bioactive Compounds in Brassicaceae Vegetables with a Role in the Prevention of Chronic Diseases. *Molecules*, vol. 23 (1), p. 15
- Ranawana, V., Campbell, F., Bestwick, C., Nicol, P., Milne, L., Duthie, G. & Raikos, V. (2016). Breads Fortified with Freeze-Dried Vegetables: Quality and Nutritional Attributes. Part II: Breads Not Containing Oil as an Ingredient. *Foods*, vol. 5 (3), p. 62

- Röös, E., Carlsson, G., Ferawati, F., Hefni, M., Stephan, A., Tidåker, P. & Witthöft, C. (2018). Less meat, more legumes: prospects and challenges in the transition toward sustainable diets in Sweden. *Renewable Agriculture and Food Systems*, pp. 1–14
- Šamec, D., Urlić, B. & Salopek-Sondi, B. (2018). Kale (Brassica oleracea var. acephala) as a superfood: Review of the scientific evidence behind the statement. *Critical Reviews in Food Science and Nutrition*, vol. 0 (0), pp. 1–12
- Saura-Calixto, F. (1998). Antioxidant dietary fiber product: A new concept and a potential food ingredient. *Journal of Agricultural and Food Chemistry*, vol. 46 (10), pp. 4303–4306
- Saura-Calixto, F. (2011). Dietary Fiber as a Carrier of Dietary Antioxidants: An Essential Physiological Function. *Journal of Agricultural and Food Chemistry*, vol. 59 (1), pp. 43–49
- Sawicki, C.M., Livingston, K.A., Obin, M., Roberts, S.B., Chung, M. & McKeown, N.M. (2017). Dietary Fiber and the Human Gut Microbiota: Application of Evidence Mapping Methodology. *Nutrients*, vol. 9 (2) (125)
- Schäfer, J., Stanojlovic, L., Trierweiler, B. & Bunzel, M. (2017). Storage related changes of cell wall based dietary fiber components of broccoli (Brassica oleracea var. italica) stems. *Food Research International*, vol. 93, pp. 43–51
- Schmidt, S., Zietz, M., Schreiner, M., Rohn, S., Kroh, L.W. & Krumbein, A. (2010). Genotypic and climatic influences on the concentration and composition of flavonoids in kale (Brassica oleracea var. sabellica). *Food Chemistry*, vol. 119 (4), pp. 1293–1299
- Selma, M.V., Espín, J.C. & Tomás-Barberán, F.A. (2009). Interaction between Phenolics and Gut Microbiota: Role in Human Health. *Journal of Agricultural and Food Chemistry*, vol. 57 (15), pp. 6485–6501
- Serreli, G. & Deiana, M. (2019). In vivo formed metabolites of polyphenols and their biological efficacy. *Food and Function*, vol. 10 (11), pp. 6999–7021
- Shahidi, F. & Naczk, M. (2004). Phenolics in Food and Nutraceuticals. Boca Raton, Florida: CRC Press.
- Shen, T., Han, X.-Z., Wang, X.-N., Fan, P.-H., Ren, D.-M. & Lou, H.-X. (2017). Protective Effects of Dietary Polyphenols in Human Diseases and Mechanisms of Action. In: Al-Gubory, K.H. & Laher, I. (eds.) *Nutritional Antioxidant Therapies: Treatments and Perspectives*. Cham: Springer International Publishing, pp. 307–345.
- Silva, E., Gerritsen, L., Dekker, M., van der Linden, E. & Scholten, E. (2013). High amounts of broccoli in pasta-like products: nutritional evaluation and sensory acceptability. *Food & Function*, vol. 4 (11), pp. 1700–1708
- Singleton, V.L. & Rossi, J.A. (1965). Colorimetry of Total Phenolics with Phosphomolybdic-Phosphotungstic Acid Reagents. *American Journal of Enology and Viticulture*, vol. 16 (3), pp. 144–158
- Stephen, A.M., Champ, M.M.-J., Cloran, S.J., Fleith, M., van Lieshout, L., Mejborn, H. & Burley, V.J. (2017). Dietary fibre in Europe: current state of knowledge on definitions, sources, recommendations, intakes and relationships to health. *Nutrition Research Reviews*, vol. 30 (02), pp. 149–190
- Strid, I., Eriksson, M., Andersson, S. & Olsson, M. (2014). Svinn av isbergssallat i primärproduktionen och grossistledet i Sverige. (2014:06). Jordbruksverket.

- Taiz, L., Zeiger, E., Møller, I.M. & Murphy, A.S. (2015). Plant physiology and development. 6. ed. Sunderland, Massachusetts, U.S.A.: Sinauer Associates.
- Tanongkankit, Y., Chiewchan, N. & Devahastin, S. (2012). Physicochemical property changes of cabbage outer leaves upon preparation into functional dietary fiber powder. *Food and Bioproducts Processing*, vol. 90 (3), pp. 541–548
- Thavarajah, D., Siva, N., Johnson, N., McGee, R. & Thavarajah, P. (2019). Effect of cover crops on the yield and nutrient concentration of organic kale (Brassica oleracea L. var. acephala). *Scientific Reports*, vol. 9 (1)
- Theander, O., Aman, P., Westerlund, E., Andersson, R. & Petersson, D. (1995). Total dietary fiber determined as neutral sugar residues, uronic acid residues, and Klason Lignin (The Uppsala method): Collaborative study. *Journal of Aoac International*, vol. 78 (4), pp. 1030– 1044
- Torres-Contreras, A.M., Nair, V., Cisneros-Zevallos, L. & Jacobo-Velázquez, D.A. (2017). Stability of Bioactive Compounds in Broccoli as Affected by Cutting Styles and Storage Time. *Molecules*, vol. 22 (4), p. 636
- UNECE (2019). UNECE STANDARD FFV-48 concerning the marketing and commercial quality control of broccoli. United Nations Publications. Available at: http://www.unece.org/fileadmin/DAM/trade/agr/standard/fresh/FFV-Std/English/48_Broccoli.pdf [2020-01-28]
- Vasanthi, H.R., Mukherjee, S. & Das, D.K. (2009). Potential Health Benefits of Broccoli- A Chemico-Biological Overview. *Mini Reviews in Medicinal Chemistry*, vol. 9 (6), pp. 749–759
- Wakeland, W., Cholette, S. & Venkat, K. (2012). Food transportation issues and reducing carbon footprint. In: Boye, J.I. & Arcand, Y. (eds.) *Green Technologies in Food Production and Processing*. Boston, MA: Springer US, pp. 211–236.
- Wang, S., Melnyk, J.P., Tsao, R. & Marcone, M.F. (2011). How natural dietary antioxidants in fruits, vegetables and legumes promote vascular health. *Food Research International*, vol. 44 (1), pp. 14–22
- Wang, Z., Li, S., Ge, S. & Lin, S. (2020). Review of Distribution, Extraction Methods, and Health Benefits of Bound Phenolics in Food Plants. *Journal of Agricultural and Food Chemistry*, vol. 68 (11), pp. 3330–3343 American Chemical Society.
- Williams, B.A., Mikkelsen, D., Flanagan, B.M. & Gidley, M.J. (2019). 'Dietary fibre': moving beyond the 'soluble/insoluble' classification for monogastric nutrition, with an emphasis on humans and pigs. *Journal of Animal Science and Biotechnology*, vol. 10, p. 45
- Williamson, G. (2017). The role of polyphenols in modern nutrition. *Nutrition Bulletin*, vol. 42 (3), pp. 226–235
- Wrick, K., Robertson, J., Vansoest, P., Lewis, B., Rivers, J., Roe, D. & Hackler, L. (1983). The Influence of Dietary Fiber Source on Human Intestinal Transit and Stool Output. *Journal of Nutrition*, vol. 113 (8), pp. 1464–1479
- Wu, H., Zhu, J., Yang, L., Wang, R. & Wang, C. (2015). Ultrasonic-assisted enzymatic extraction of phenolics from broccoli (Brassica oleracea L. var. italica) inflorescences and evaluation of antioxidant activity invitro. *Food Science and Technology International*, vol. 21 (4), pp. 306– 319

- Yang, I., Jayaprakasha, G.K. & Patil, B. (2018). In vitro digestion with bile acids enhances the bioaccessibility of kale polyphenols. *Food and Function*, vol. 9 (2), pp. 1235–1244
- Yang, J., Martinez, I., Walter, J., Keshavarzian, A. & Rose, D.J. (2013). In vitro characterization of the impact of selected dietary fibers on fecal microbiota composition and short chain fatty acid production. *Anaerobe*, vol. 23, pp. 74–81
- Yi, X.W., Yang, F., Liu, J.X. & Wang, J.K. (2015). Effects of Replacement of Concentrate Mixture by Broccoli Byproducts on Lactating Performance in Dairy Cows. *Asian-Australasian Journal of Animal Sciences*, vol. 28 (10), pp. 1449–1453

Popular science summary

Broccoli leaves to improve health and fight food insecurity

What if I were to tell you that there is a leafy vegetable that is as nutritious as curly kale, but it is thrown away in the fields at harvest time? That this leafy vegetable constitutes about half of the plant in question, and the part that we eat is only 20 % of the plant? This underutilised vegetable, broccoli leaves, is the subject of this thesis.

Dietary fibre and phenolic compounds (a type of antioxidant) are found naturally in fruits and vegetables and have beneficial effects on health, *e.g.* by reducing the risk of developing some kinds of cancer and cardiovascular diseases, lowering cancer mortality and cholesterol levels in the blood, improving the immune system and positively affecting the gut microbiota. Many of us consumers today eat too little fruits and vegetables, and hence do not get the sufficient amounts of these health-beneficial compounds in our daily diet.

This thesis showed that broccoli leaves are rich in dietary fibre, with similar levels as in broccoli florets, cabbage leaves and kale leaves. The total content did not vary between the years in this study, but the proportions of some of the dietary fibre constituents changed, possibly due to weather conditions. Broccoli leaves were also found to be rich in of phenolic compounds, with higher levels than those found in broccoli florets and similar to those found in kale leaves. Hence, there is nutritional value in eating the broccoli leaves, if we as consumers had access to them.

Combined, Swedish production of broccoli and cauliflower (heads only) was 9330 tonnes in 2018, compared with 1.2 million tonnes in the US, 8.8 million tonnes in India, and 10.6 million tonnes in China, while the world total was 26.5 million tonnes. If the biomass is harvested and used as food represents only 20 % of total biomass, the amount of waste in the broccoli and cauliflower production in the world is 106 million tonnes per year. Hence, the side streams in broccoli (and cauliflower) production are abundant resources, and more uses for them should be developed.

But why is it of interest to study broccoli leaves? How can it help fight the food insecurity?

The food situation in the world is currently unbalanced, with as many as 821 million people that are undernourished while the amount of food wasted could feed 1.9 billion people. Consequently, much of the food produced is not used as food, while the resources devoted to producing the wasted food can also be considered as wasted. Many of the resources that are used in food production, such as water, fertiliser, land and energy, are limited, and must be used in a more efficient way. Research on how to use broccoli leaves in new ways might also provide ideas about how to use other parts of vegetables that are thrown away today, hence increasing the amount of food in the world without increasing the use of water, land, fertilisers and energy.

This thesis provides new knowledge about the unharvested leaves and stems of broccoli, and their content of some known health-beneficial compounds compared with those in other common plant-based foods. With the novel information provided here about the levels of dietary fibre and phenolic compounds in broccoli leaves, new uses for the broccoli leaves can be developed, which in turn can improve consumer health.

Populärvetenskaplig sammanfattning

Broccoliblad för att förbättra hälsa och bekämpa osäker livsmedelsproduktion

Vet du om att det finns en bladgrönsak som är lika näringsrik som grönkål, men som slängs på fältet vid skörd? Att denna bladgrönsak utgör ungefär halva den aktuella plantan, och att den del som vi äter enbart är 20 % av plantan? Denna underutnyttjade grönsak, broccoliblad, är föremålet för denna avhandling.

Kostfibrer och fenoliska ämnen (en typ av antioxidanter) finns naturligt i frukt och grönsaker och har hälsofrämjande effekter, exempelvis genom att minska risken för att utveckla vissa former av cancer och hjärt-kärlsjukdomar, sänka dödligheten i cancer och nivåerna av kolesterol i blodet, förbättra immunsystemet och positivt påverka tarmfloran. Många av oss konsumenter äter för lite frukt och grönsaker, och får därmed inte i oss tillräckliga halter av dessa hälsofrämjande ämnen i vår dagliga kost.

Denna avhandling visar att broccoliblad är rika på kostfibrer, med halter som motsvarar halterna i broccolibuketter, vitkålsblad och grönkålsblad. Det totala innehållet varierade inte mellan åren i denna studie, men sammansättningen av några av kostfibrerna förändrades, möjligen på grund av väderförhållandena. Broccolibladen var också rika på fenoliska ämnen, med högre halter jämfört med de som finns i broccolibuketter och jämförbara med de som finns i grönkål. Det finns därmed ett värde ur näringssynpunkt i att äta broccolibladen, om vi konsumenter kan få tillgång till dem.

Sammanslaget så är den svenska produktionen av broccoli och blomkål (huvud enbart) 9330 ton under 2018, jämfört med 1,2 miljoner ton i USA, 8,8 miljoner ton i Indien och 10,6 miljoner ton i Kina, medan den globala mängden var 26,5 miljoner ton. Om biomassan som skördas och användas som mat enbart är 20 % av biomassan, då blir den totala mängden svinn i världsproduktionen av broccoli och blomkål 106 miljoner ton per år. Följaktligen är sidoströmmarna i

produktionen av broccoli (och blomkål) en tillgänglig resurs och fler användningsområden för dem borde utvecklas.

Men varför är det av intresse att studera broccoliblad? Hur kan det hjälpa till att bekämpa en osäker livsmedelsförsörjning?

Livsmedelssituation i världen är för närvarande obalanserad, med så många som 821 miljoner undernärda människor, samtidigt som mängden matsvinn och matavfall skulle kunna mätta 1,9 miljarder människor. Följaktligen används mycket av den mat som produceras inte som mat, medan resurserna som lagts på att producerade den slängda maten därmed kan räknas som svinn. Många av de resurser som används i matproduktionen, så som vatten, gödning, mark och energi, är begränsade, och behöver bli använda på ett mer effektivt sätt. Forskning kring nya användningsområden för broccoliblad kan även ge idéer kring ur man kan använda andra delar av växter som idag slängs, och därmed öka mängden mat i världen utan att öka användandet av vatten, land, gödning och energi.

Denna avhandling ger ny kunskap kring de oskördade bladen och stammarna av broccoli och deras innehåll av några kända hälsofrämjande ämnen jämfört med andra vanliga växtbaserade livsmedel. Med den nya information som ges om halterna av kostfibrer och fenoliska ämnen i broccoliblad, kan nya användningsområden utvecklas, vilket i sin tur kan förbättra konsumenters hälsa.

Acknowledgements

My time as a PhD student at Swedish University of Agricultural Sciences has been a journey, during which I have grown both in knowledge and as a person. I have crossed paths with many fellow travellers at different stages in the journey. These are some to whom I would like to express a special thanks:

- My supervisor Marie, thank you for inviting me into the World of Broccoli, and showing me the pros and cons of academic life.
- My supervisor Eva, thank you for helping me see connections where I only see chaos, and for being the captain that helped me sail through the storm.
- My supervisor Roger, thank you for all your patience whenever I came with yet another question about dietary fibre, phenolics or statistics, and for always taking the time to explain in a pedagogic way so that I could see the light.
- My supervisor Helena, thank you for helping me with difficult choices, making me understand when my text really didn't make any sense and offering your wisdom (sometimes in exchange for some sweets so that I didn't have to snack alone).
- Kalle, you are worth your weight in gold and all precious metals. Thank you for all the time you spent teaching me the difficult task of extracting and characterising phenolic compounds.
- Annica, thank you for showing me that the difficult method of analysing dietary fibre is actually quite fun the second time around, and not just terrifying.

- My close colleagues and friends Anna-Lovisa, Joel, Antonio, Sophie, Catja, Maja, Linnea, Alex, Evelyn, Vera, Sbatie. Without you, my time at Alnarp would have been much tougher, duller and a poorer experience than it was. You made me laugh through my lows and you have soared with me during my highs. No amount of cake from me can say thank you enough.
- My office mate and friend, Jonatan. A special thank you for putting up with all of my talking, all of my questions and all of my high and low feelings. I cannot image a nicer (or greener) office than the one that I have had with you. A special cake, or some vanilla hearts, to you.
- My heart, my love, my better half, Anton, thanks for reminding me to be human once in a while and also that sometimes I am damn good at what I do and as a person.
- My family, who have supported me in everything I do without hesitation and with a lot of encouragement. Skam den som ger sig!