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Advances in Breeding in Vegetable *Brassica rapa* Crops

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Abstract

Brassica rapa includes oil and vegetable crops having a variety of forms, such as oilseeds, leafy vegetables and turnips. Leafy types, which are called turnip greens and turnip tops, are popular crops in NW Spain, and they represent an important part of the diet. However, their cultivation is limited in southern areas or in the Mediterranean basin, probably due to a lack of adaptation. Still, they could occupy a prominent place in the Mediterranean diet, which is based on a high consumption of fruits and vegetables. In this review, we summarize the studies on the agronomical and nutritional value of these crops when grown under Mediterranean climate conditions. Data reported here might be useful for a deeper understanding of these crops for both nutritional quality and bioaccessibility, and for selecting varieties adapted to the two abovementioned Mediterranean conditions, as well as for organic farming systems, thus contributing to the diversification of traditional Brassica vegetable production systems.

Keywords: turnip greens, turnip tops, adaptation, bioaccessibility, nutritional quality

1. Introduction

1.1 Taxonomy and diversified morphotypes

Brassica rapa ($2n = 20$, synonymous with *B. campestris* L.) is an economically important species belonging to the *Brassica* genus, Brassiceae tribe, from the Brassicaceae family. The *Brassica* genus includes many important crops. Among them, relationship of six species formed the model of U's triangle, with three basic diploid species, namely *B. rapa* (A genome, $n = 10$), *Brassica oleracea* (C genome, $n = 9$) and *Brassica nigra* (B genome, $n = 8$), which gave rise to three amphidiploid species, namely *Brassica napus* (AC genome, $n = 19$), *Brassica juncea* (AB genome, $n = 18$) and *Brassica carinata* (BC genome, $n = 17$).

Brassica rapa is an important oil and vegetable crop in many parts of the world, whose seeds are used for oil, and leaves, flowers, stems and roots are used as vegetables. *B. rapa* vegetables are consumed worldwide and provide a large proportion of the daily food intake in many regions of the world. Cultivation of this species for many centuries in different parts of the world has caused a large variation in the plant organs that are consumed (roots, leaves, and flower buds), which has resulted in the human selection of different morphotypes, depending on local preferences

[1]. Based on their morphological appearance and on the organs used, *B. rapa* crops can be classified into two groups:

- i. Vegetable types used for their tubers (=hypocotyl), leaves and flower buds, which include the *rapa* (= *rapifera* or *ruvo*) group and the leafy vegetable forms. These vegetable types belong to six groups: *rapa*, *chinensis*, *pekinensis*, *parachinensis*, *nipposinica*, *perviridis* and *narinosa* [2].
- ii. Oleiferous types, of which canola is a specific form, having low erucic acid levels in its oil and low glucosinolate content in its meal protein.

Until recently, these groups were considered as separate species because of the wide range of variability they show and the fact that they evolved in isolation from each other.

The *oleifera B. rapa* group includes oilseed crops that are known in Europe as rapeseed or turnip rape. It is believed that European forms developed in the Mediterranean area and then they were distributed from Europe to China. In India, crops used for oil production belong to the *trilochularis* and *dichotoma* groups. Sarson and toria types belong to this group. There are three ecotypes: brown sarson, toria and yellow sarson. Out of these, brown sarson appears to be the oldest one [2]. Yellow sarson is characterized by its yellow colored seeds and self-compatibility. Many of the cultivars have 3–4 valved siliquae, and for this reason, it was named *trilochularis*. It is believed to have evolved from brown sarson as a mutant and has survived because of its self-compatible nature. It might have been selected by farmers for its attractive yellow-colored seeds and bigger seed size.

Vegetable *B. rapa* crops, including *rapifera* and leafy types, are important crops in European and Asian countries, particularly in China, Korea, and Japan. Their consumption varies widely around the world and they are consumed as raw or steamed vegetables. The largest and most diverse *B. rapa* group consists of crops belonging to the *pekinensis* type, which includes popular crops in Chinese cuisine such as pet-sai or Chinese cabbage (**Table 1**). They are characterized by having large leaves and forming heads of different shapes. Chinese cabbage, for example, is the cabbage used for preparing dishes such as sauerkraut and kimchi, the famous fermented dish favored by Koreans. Its seeds have also been used for the hot mustard favored in Chinese cuisine. Pak-choy or bok-choy (*chinensis* group) are also popular crops in Asian culture. They have been used for their leaves, which do not form heads and are smooth. It is assumed that pak-choy types with narrow or wide green-white petioles were the first *B. rapa* crops to evolve in Central China. Another group of cultivars that is characterized by many narrow leaves belong to the *perviridis* group, which includes neep greens from Europe and the Japanese cultivar Komatsuna. Finally, we have the *nipposinica* group, which includes Japanese crops like mizuna or mibuna, which can be eaten raw or cooked at any stage, from seedling to mature plant (**Table 1**).

The *rapa* or *rapifera* group is characterized by the thickening of the hypocotyls, which can show different colors and shapes, and has a mainly horticultural and forage use. Turnips are both cultivated as fodder crops or as vegetables, and depending on the region, the tubers, leaves and shoots are used. Turnip greens are the young leaves harvested in the vegetative growth period. Turnip tops are the fructiferous stems with flower buds and the surrounding leaves that are consumed before opening and while still green (**Table 1, Figure 1**). In Europe, they are notably popular in Portugal, Italy and Spain, where they play an important role in traditional farming and in the diet. In these countries, *B. rapa* includes two main crops, turnip greens and turnip tops, as vegetable products. They are commonly consumed as boiled

Group	Crops	Distribution	Plant part used
Vegetable types			
<i>rapa</i> (= <i>rapifera</i>)	Turnip, turnip greens, turnip tops, rapini, broccoletti di rape, brocoletto, turnip broccoli, cima di rapa, Italian turnip	Europe	Leaves, flower buds and hypocotyl
<i>chinensis</i>	Pak-choy, Bok-choy, celery mustard	China	Leaves
<i>pekinensis</i>	Chinese cabbage, napa cabbage, celery cabbage, pet-sai, napa, wong-bok, chihli	China, Korea, Taiwan, Japan	Leaves
<i>parachinensis</i>	Choi-sum, caixin, caitai	China	Leaves and flower buds
<i>nipposinica</i>	Mizuna, mibuna, curled mustard, Japanese greens	Japan	Leaves
<i>perviridis</i>	Komatsuna, spinach mustard, tendergreen, neep greens	Japan, Korea, Taiwan	Leaves
<i>narinosa</i>	Wutacai or heibaicai	China	Leaves
Oleifera types			
<i>oleifera</i>	Turnip rape, rapeseed	China	Seeds
<i>dichotoma</i>	Brown sarson, toria	India	Seeds
<i>trilochularis</i>	Yellow sarson	India	Seeds

Table 1.
 Taxonomic groups in *Brassica rapa* species.

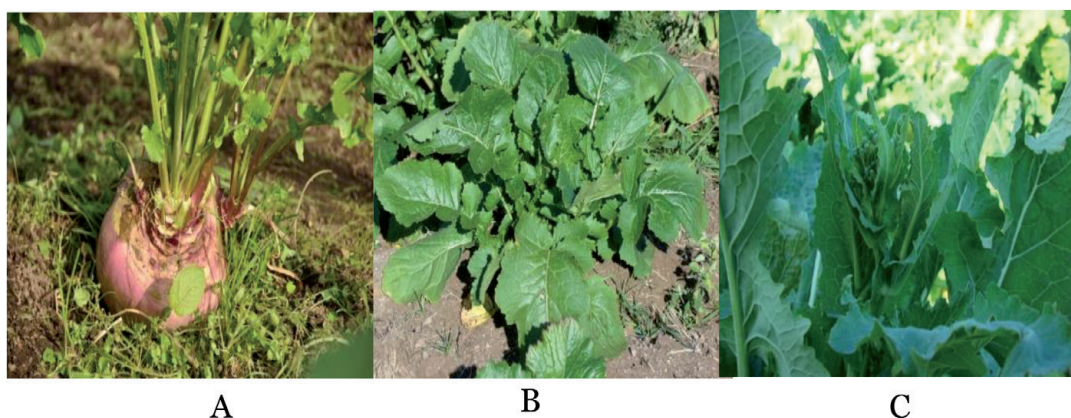


Figure 1.
 Leafy vegetable crops from the *Brassica rapa* group: Turnip (A), turnip greens (B) and turnip tops (C).

vegetables, being used in the preparation of soups and stews and they have a slightly spicy flavor like mustard greens [3]. Turnip greens and turnip tops have good commercial prospects in both countries and, the number of companies selling *B. rapa* canned products has been increasing in the last years.

1.2 Origin of *Brassica rapa* crops

The origin of cultivated *B. rapa* crops is still unknown. This species was probably the first domesticated Brassica several millennia ago, as a multipurpose crop [4]. It is believed that the most likely explanation for the wide variation within this species is that cultivated forms arose independently in different places of the world from wild *B. rapa* [1]. It seems to have spread naturally to the Western

Mediterranean region and to Central Asia, with secondary centers of diversity in Europe, Western Russia, Central Asia, and the Near East [5].

According to the studies based on morphology, geographic distribution, isozymes and molecular data, cultivated subspecies of *B. rapa* most likely originated independently in two different centers—Europe and Asia. Europe should be one primary center of origin for oil and turnip types [4], whereas East Asia should be another primary center for Indian oil types and leafy vegetables [1, 6]. Today, it is well established that Asia represents the main area of diversification for vegetable *B. rapa* crops. Leafy vegetables such as Chinese cabbage, pak-choi and narinosa may have been first domesticated in China. China is also the center of origin of Chinese turnip rape (var. *oleifera*). Other accessions of *B. rapa* most likely derived from different morphotypes in the two centers of origin and subsequently evolved separately.

It is believed that *B. rapa* was introduced into China through Western Asia or Mongolia as an agricultural species. In fact, *B. rapa* is also recognized as the ancestor of many oriental Brassica vegetables. Its introduction into Japan could have occurred via China or Siberia. In India, *B. rapa* is cultivated as an oilseed, but no wild forms are known in this country. In East Asia, leafy types such as Chinese cabbage, bok choy, pak-choi, mizuna, celery mustard, and Chinese kale, among others, are used extensively as vegetables [6]. In China, flowers of the crop called choy-sum (*parachinensis* group) are also consumed, and these inflorescences are known as caixin or caitai.

In Europe, broccoleto types, turnip rape and turnips are the predominant forms [7] and they can be used for both as food and feed. Other *B. rapa* accessions most likely derived from different morphotypes in the two centers of origin and subsequently evolved separately. The *rapa* or *rapifera* group is believed to have evolved in Europe. It is supposed that it was first used for its nutritious root around 2,500–2,000 B.C. and spread to other parts of the world afterwards. The expansion of vegetable crops within this group such as turnip greens and turnip tops took place later on and independently from the origin of leafy forms in Asia [7].

1.3 Breeding for turnip greens and turnip tops

This review will be focused on two *B. rapa* crops: turnip greens and turnip tops. In Northwestern Spain, Portugal and Southern Italy, both crops have a long tradition and they represent two important commodities, being part of very traditional recipes. Like other Brassica vegetable crops, they are generally either eaten after being cooked or they can also be processed as canned foods. Turnip greens and turnip tops have good commercial prospects and their consumption, both fresh and processed, has increased considerably in the last years. New uses and new markets for these crops (canned, frozen, fourth range-foods, ...) have been grown lately.

A collection of local varieties of turnip greens and turnip tops from Northwestern Spain is currently kept at the Misión Biológica de Galicia (CSIC) in Pontevedra, Northwestern Spain. These landraces are a valuable resource, since they are adapted to the climatic conditions of this area. Agronomical and nutritional evaluations of this collection were previously performed by [8–10]. Authors reported a high genetic diversity for several agronomic traits and found that some varieties are a valuable source of bioactive compounds such as glucosinolates and phenolic compounds. However, their cultivation is limited in southern areas or in the Mediterranean basin, probably due to a lack of adaptation. Still, these crops could occupy a prominent place in the Mediterranean diet, which is based on a high consumption of fruits and vegetables. The evaluation of *B. rapa* varieties with wide adaptability across diverse farming environments becomes essential for selecting

varieties for future breeding programs based on producers' and consumers' preferences. With this goal in mind, a breeding program in turnip tops and turnip greens was started at IAS-CSIC in Córdoba (South of Spain) in recent years. The goal was to achieve varieties adapted to the environmental conditions of this area but preserving similar nutritional properties to those produced in their original region.

In this review, we summarize the studies on the agronomical and nutritional value of these crops grown under Mediterranean climate conditions. Data reported here might be useful for deeper understanding of these crops for both nutritional quality and bioaccessibility, resistance to biotic stress, and for selecting varieties adapted to Mediterranean conditions, thus contributing to the diversification of traditional Brassica vegetable production systems.

2. Characterization, evaluation and selection of *Brassica rapa* germplasm under Mediterranean conditions

2.1 Introduction

It is well known that the change from a Western dietary pattern (high consumption of calories, animal products and sugars) to a Mediterranean diet (high consumption of fruits, vegetables, grains, legumes and reduced amounts of animal products, with the use of olive oil as the preferred fat) reduces the risk of diabetes by 7%, heart disease by 10%, and total mortality by 8% [11, 12]. It is therefore clear that increasing and promoting the consumption of locally produced foods of plant origin (km. 0), and, in particular, those with nutraceutical properties, is one of the crucial factors for the well-being and health promotion, hence allowing the prevention of various diseases, such as cancer, and cardiovascular and neurodegenerative diseases [13].

For several years now, the IAS-CSIC research group in plant breeding has been studying the possibilities of producing turnip greens and turnip tops in Southern Spain for incorporation into the Mediterranean diet. Under these conditions, they could be considered as a new regional crop that provides vegetables with more interesting nutraceutical and organoleptic properties than Brassica species, such as cauliflower, broccoli or Brussels sprouts, which have seen their consumption reduced mainly in children due to their strong and peculiar smell and taste.

The goal of this work was to study the adaptation and cultivation of a collection of germplasm and cultivars of *B. rapa* harvested in Galicia (Northwestern Spain) in the Guadalquivir Valley, and select the lines with better agronomic and nutritional characteristics, hence expanding the usual consumption area. In the evaluation and selection process, the turnip greens and turnip tops production capacity, as well as the glucosinolate content of the harvested products as a quality criterion for the final product, were studied.

2.2 Plant material

The *B. rapa* L. var. *rapa* germplasm used for this work came from the Brassica Germplasm Bank at Misión Biológica de Galicia (Pontevedra, Northwestern Spain), where it had been characterized by its agronomic characteristics and its aptitude for turnip greens and turnip tops production.

2.3 First trials of *Brassica rapa* cultivation in Córdoba (2009-2012)

The effect of different sowing dates on the production and quality of turnip greens and turnip tops was studied during the first stage. For this purpose, five

B. rapa accessions from the MBG-CSIC Germplasm Bank selected by their differences in phenological growth cycle (early and late) were used. These five accessions were cultivated in Córdoba (Southern Spain, Guadalquivir Valley) during the 2009/10, 2010/11 and 2011/12 agricultural seasons. Different sowing dates were tested for each agricultural season in order to cover the largest potential period of turnip greens and turnip tops production. During the 2009/10 season, the same entries were grown in Pontevedra, being used as a control.

During the first season that the five *B. rapa* accessions were sown in Córdoba (2009/10), the low turnip greens production for all entries highlighted the inadequacy of the sowing dates chosen and the need to bring them forward in successive seasons. In the first sowing, turnip greens production was low and turnip tops of acceptable quality were not obtained. The second sowing was lost due to the unusually high rainfall that caused root asphyxiation and plant death. In the third sowing, a good turnip greens production was achieved but the increase in spring temperatures caused them to rise quickly, thus obtaining low-quality turnip tops.

These results determined that all sowing dates would have to be brought forward in the following seasons (2010/11 and 2011/12), starting in September. This change notably favored the crop adaptation in Córdoba, improved plant development in the field and improved the turnip greens and turnip tops production. The existence of accessions, that did not form quality turnip tops in Córdoba, revealed the need to extend the germplasm collection to be studied, in order to be able to select the most suitable genotypes for turnip tops production in Mediterranean edaphoclimatic conditions (Table 2).

2.4 Characterization of a *Brassica rapa* germplasm collection (2013–2014)

Once the optimal sowing date was adjusted, in the next stage (2013/14 agricultural season), characterization and evaluation of 19 *B. rapa* accessions also from the MBG-CSIC Germplasm Bank was carried out. The selection of these entries was made according to the agronomic characteristics and phenological cycle in their origin area. A randomized block design with 3 replications was used in all trials. Glucosinolate analysis of was carried out in accordance with the European standard for this determination [14].

Location	Season	Transplanting date	Turnip greens harvest	Turnip tops harvest
Pontevedra (Control)	2009/10	September	December	January to April
Córdoba	2009/10	January	April	No
		March	No	No
		April	June	July
	2010/11	September	December	January
		November	April	April
		January	No	No
	2011/12	September	November	December
		November	February	February
		January	No	No

Table 2. *Transplanting and harvesting dates of Brassica rapa accessions in each localition by season and sowing date.*

The cultivation of these entries in Córdoba was successful, and turnip greens and turnip tops harvest was abundant for almost all the entries (**Figure 2**).

In addition to the agronomic evaluation, the glucosinolate content of the turnip greens and turnip tops harvested for each of the entries was analyzed. In general, the average glucosinolate content of turnip greens (27.98 $\mu\text{mol/g}$ dry matter) was lower than that of turnip tops (30.25 $\mu\text{mol/g}$ dry matter), which highlights the high variability in glucosinolate content between the different accessions and within each accession. The glucosinolate pattern was similar in turnip greens and turnip tops, with gluconapin being the major glucosinolate (representing about 80% of total glucosinolates), followed by progoitrin, glucobrassicinapin, gluconapoleiferin, glucobrassicin, 4-metoxi-glucobrassicin and neoglucobrassicin. Similar results were found in previous works on the glucosinolate content in vegetable *B. rapa* crops [10, 15]. No differences were found between the glucosinolate profile of the samples collected in Córdoba and that of samples collected in Pontevedra. Some accessions cultivated in Córdoba stood out for their ability to produce turnip greens and/or turnip tops with a total glucosinolate content equal or greater than those produced at their usual cultivation place (**Figure 3**).

2.5 Evaluation of selected *Brassica rapa* accessions

In the third stage (2014/15 season) six accessions were cultivated in Córdoba (from the 19 studied in the previous season), which were selected based on their homogeneity and their turnip tops production in Córdoba. The entries chosen were evaluated in terms of their agronomic characteristics, productivity and glucosinolate content of the harvested turnip greens and turnip tops.

Agronomic evaluations were carried out throughout the entire cultivation cycle in Córdoba, and it was possible to harvest quality turnip greens and turnip tops in all cultivated accessions (**Figure 4**). In general, we obtained turnip greens in Córdoba with lower fresh weight than that of turnip greens produced in Pontevedra. The opposite occurred with the fresh weight of turnip tops and the number of turnip tops/plant, which was higher in the entries cultivated in Córdoba (**Table 3**).



Figure 2.
Brassica rapa cultivation at the IAS-CSIC experimental farm, Córdoba (season 2013–2014).

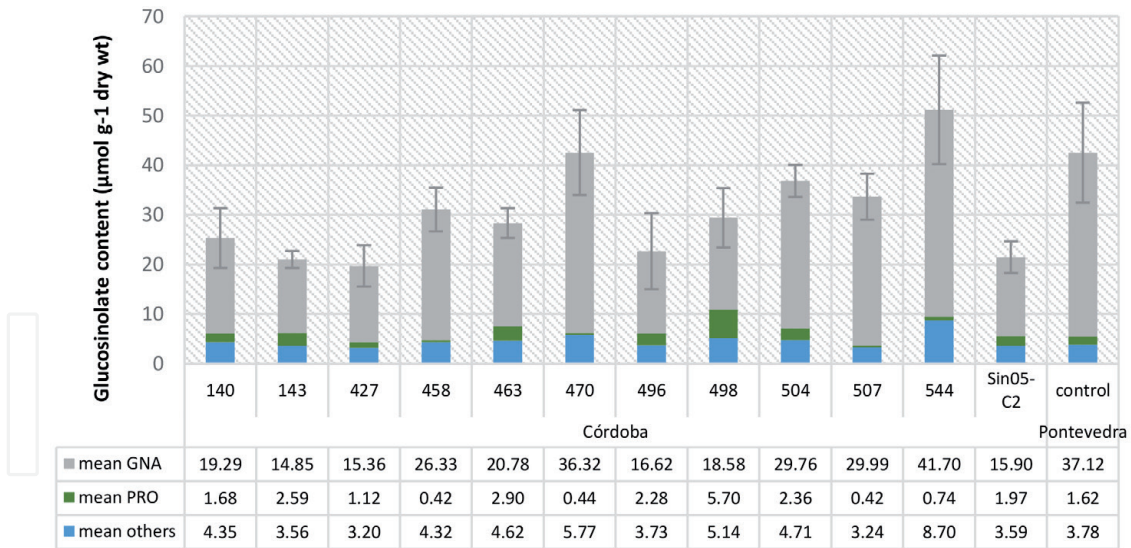


Figure 3. Gluconapin (GNA), progoitrin (PRO) and other glucosinolate contents (mean ± standard deviation) in turnip tops of *Brassica rapa* accessions cultivated in Córdoba and Pontevedra. Error bars represent the standard deviation of total glucosinolates.



Figure 4. Samples of turnip tops harvested in Córdoba (season 2014–2015).

	Turnip greens		Turnip tops			T ^c
	FW ^a (g)	M ^b (%)	Fw (g)	M (%)	Stems (n°)	
BRS0143	6.04	80.94	49.46	90.85	20.58	120
BRS0427	5.37	80.04	47.14	90.27	17.80	98
BRS0496	5.85	80.69	33.44	90.33	12.14	98
BRS0498	7.33	79.09	113.87	92.88	19.94	134
BRS0504	7.55	81.80	107.11	93.09	19.39	134
BRSin05-C2	5.73	79.70	45.36	85.58	21.84	106
Mean Cordoba	6.43	80.38	66.06	90.50	18.62	115
* Mean Pontevedra	22.12	90	63.02	91	12.13	162.3

^aSource: Francisco et al., [9].

^aFW: fresh weight.

^bM: moisture.

^cD: days from turnip tops sowing to harvest.

Table 3. Agronomic characteristics of turnip greens and turnip tops harvested in Córdoba (season 2014–2015).

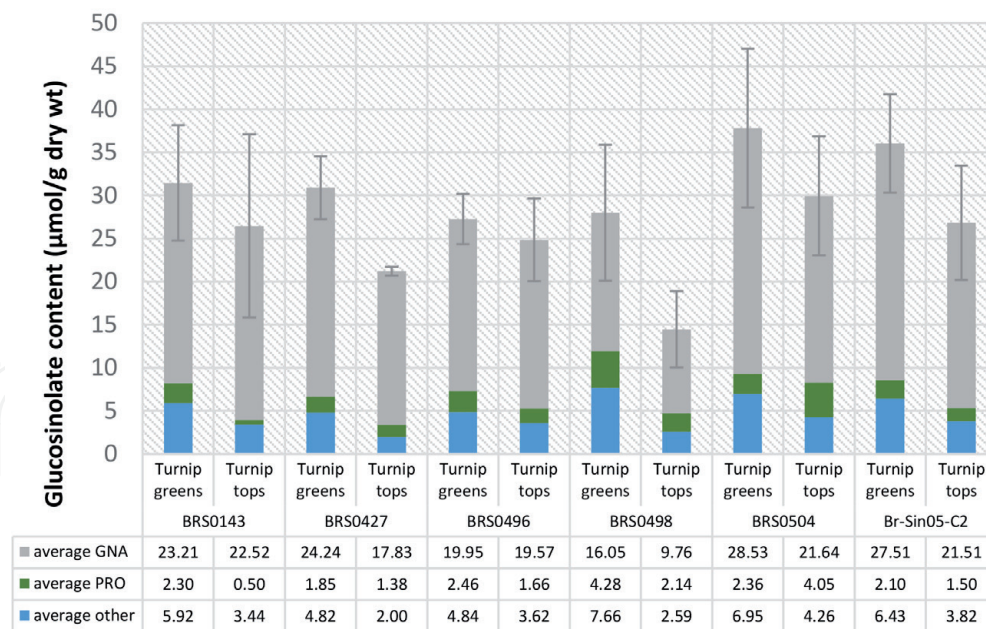


Figure 5. Gluconapin (GNA), progoitrin (PRO) and other glucosinolate contents (mean \pm standard deviation) in turnip greens and turnip tops of Brassica rapa selected accessions cultivated in Córdoba (season 2014–2015).

The total glucosinolate content was significantly higher in turnip greens than in turnip tops. Accessions BRS0143, BRS0504 and BRSin05-C2 stood out for their capacity to produce turnip greens and turnip tops with high gluconapin content (Figure 5). There are numerous studies that indicate that gluconapin is beneficial for health, since its degradation product (3-butenyl isothiocyanate) is capable of producing cell death induced mainly through tumor cell necrosis [16–18]. These results indicate the potential of these selected accessions to obtain varieties that are capable of producing turnip greens and turnip tops with high levels of beneficial glucosinolates (gluconapin) and low levels of glucosinolates with anti-nutritional potential (progoitrin).

3. Glucosinolate bioaccessibility

Bioavailability can be defined as being the micronutrient or bioactive compound fraction, originally present in the food, which is solubilized and absorbed in the intestinal lumen, metabolized by typical routes, and finally used for typical physiological functions or deposited in storage compounds [19]. As glucosinolates are hydrolyzed by the enzyme myrosinase, into glucose and a wide variety of unstable aglycones such as isothiocyanates, thiocyanates, nitriles, indoles, thiones and epithioalkanes among others, knowing the beneficial physiological effect of all these compounds requires a wide variety of further in vivo studies.

A first step could be to focus on the amount of glucosinolates that come into contact with enterocytes. Thus, bioavailability studies can be partly replaced by bioaccessibility ones. This term refers to the fraction of the micronutrient or bioactive compound that is soluble in the intestinal lumen and therefore will be capable of being absorbed by the enterocytes of the small intestine [20]. Bioaccessibility studies are based on a simulated gastrointestinal food digestion formed by an oral phase with salivary amylase, a gastric phase with pepsin-HCl at pH 2, and later by an intestinal phase with pancreatin-bile salts [21, 22]. Finally, the digest is centrifuged and glucosinolate fraction is determined, as the amount of this compound present in the supernatant.

Several studies have shown that around 85% of the initial glucosinolate dose in a rapeseed meal is capable of resisting the physiological conditions of the stomach,

and around 63–75% remains intact after *in vitro* simulation of a 4 h digestion in the small intestine [23]. Another study [24], using simulated *ex vivo* gastrointestinal digestion, also gave bioaccessibility values of 71 and 29% for two glucosinolates (glucoraphenin and glucoraphasatin) of *Matthiola incana*. The presence of the sulphate group and thioglucose moiety confers the glucosinolate molecule with high water solubility [23]. However, this bioaccessibility percentage will depend on the structure of the glucosinolate molecule and its ability to bind non-specifically to macromolecules (mainly proteins, peptides and small glycoproteins).

Thus, a previous study [25] with five plant species belonging to the Brassicaceae family (*B. rapa*, *B. oleracea*, *B. carinata*, *E. vesicaria* and *S. alba*) showed that over 30% of the glucosinolates initially present in the leaves of this plant species would be capable of reaching human enterocytes, hence resisting the degradation processes of digestive enzymes, including its own myrosinase enzyme (Figure 6). In that study, the highest bioaccessibility percentages corresponded to indolic glucosinolates such as glucobrassicin (70%) and neoglucobrassicin (around 56%), followed by aliphatic ones such as progoitrin (49%) and sinigrin (32–43%). The lowest bioaccessibility percentages corresponded to aromatic glucosinolates, with a percentage of 25% for sinalbin.

Another similar study conducted by [26] also showed the highest percentage of bioaccessibility for an indolic glucosinolate like glucobrassicin (around 42%) in broccoli “Parthenon” and Savoy cabbage “Dama” brassicas. According to these results, the presence of a five-membered pyrrole ring fused to a benzene ring seems to confer the glucosinolate molecule a higher solubility and less uptake to other molecules from the enzymatic digestion of food than the aromatic glucosinolate group.

It is suggested that intact glucosinolates must pass through the gut epithelium by passive, facilitated or active transport [23], although the real path way remains unknown. It is also important to emphasize that several glucosinolate hydrolysis-derived products, such as isothiocyanates and indoles, can also be found in the small intestine, likely arising by the enzymatic processing mediated by the plant

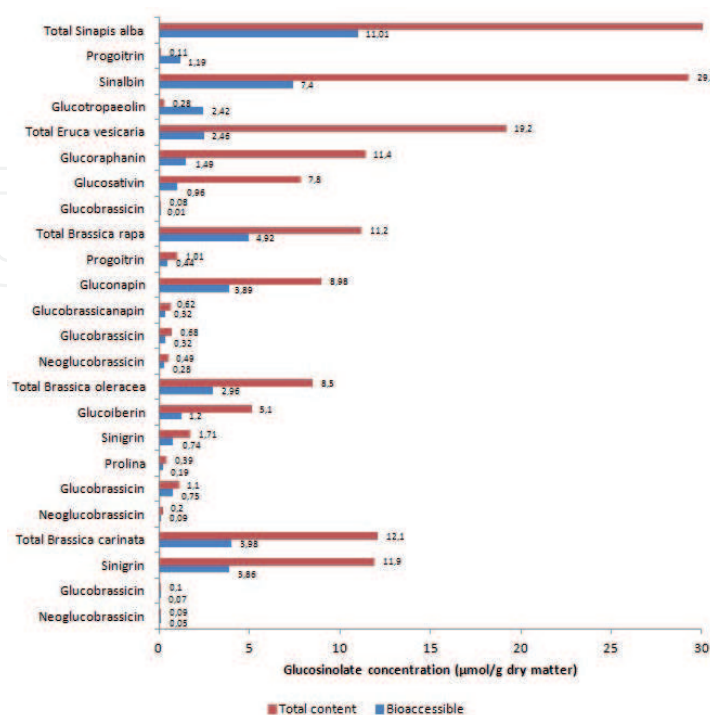


Figure 6. Glucosinolate concentration (total and bioaccessible) in five plant species belonging to the Brassicaceae family (expressed as $\mu\text{mol/g dw}$).

myrosinase. Bioaccessibility studies should also include these compounds because both glucosinolates and their derivatives provide beneficial effects on human health. Thus, studies with radioisotopes in rats [27, 28] have shown a high absorption of isothiocyanates, with a blood peak observed 3 h after ingestion. De la Fuente et al. [29] have reported bioaccessibility percentages ranging between 31 and 63% for total isothiocyanates of Brassica microgreens. Among all the isothiocyanates, one of the most studied is sulforaphane, which is produced by the hydrolysis of the glucosinolate glucoraphanin present in broccoli. A bioaccessibility study conducted by [30] has shown a concentration for sulforaphane and sulforaphane nitrile of 10.4 and 49.9 $\mu\text{mol}/100\text{ g}$ of fresh broccoli after the gastric phase and 28.6 and 113 $\mu\text{mol}/100\text{ g}$ of fresh broccoli after the intestinal phase. However, there is a wide variety of isothiocyanates coming from enzymatic hydrolysis of other glucosinolates whose bioavailability has not been studied yet. More research is needed in this field in order to know the nutritional role of all these compounds.

Finally, glucosinolates that are not absorbed in the small intestine reach the colon, where they could be hydrolyzed with bacterial myrosinase in nitriles and other unspecified products [31]. Formation of products from glucosinolates by intestinal microbiota is also still poorly documented and further studies are equally necessary.

4. Insect pests and diseases

Among the insect pests affecting *B. rapa* crops, the diamondback moth, *Plutella xylostella* L. (Lepidoptera: Plutellidae) and cabbage root flies, *Delia* spp. (Diptera: Anthomyiidae) are considered the most damaging pests [32–34]. Other important insect pests include *Phyllotreta* spp. (Coleoptera: Chrysomelidae) flea beetles, cabbage aphid, *Brevicoryne brassicae* L. (Hemiptera: Aphididae), cabbage butterflies, *Pieris* spp. (Lepidoptera: Pieridae), cabbage moth, *Mamestra brassicae* L. (Lepidoptera: Noctuidae), pollen beetle, *Meligethes aeneus* F. (Coleoptera: Nitidulidae), cabbage seed pod weevil, *Ceutorhynchus obstrictus* Marsham (Coleoptera: Curculionidae) [35–37]. At present, turnip greens/tops resistant varieties to major pests are scarce and chemical control is the most used method to protect these crops. Because of its attractiveness to insects, *B. rapa* has also been proposed as a trap crop and insectary plant [38].

The role of glucosinolates on pest resistance has been extensively studied in Brassica crops. Glucosinolates are considered a source of resistance to the cabbage moth, *M. brassicae*, and to the specialist *Pieris rapae* [39]. The yellow flowers of *B. rapa* are very attractive to pollen beetle, *Meligethes aeneus*, and the glucosinolate content in the inflorescence is positively correlated with *M. aeneus* incidence [40]. The content of certain glucosinolates is associated to an increased developmental time and reduced weight in the cabbage seedpod weevil, *Ceutorhynchus obstrictus* [41]. Since glucosinolate content can increase susceptibility to *P. xylostella*, breeding programs leading to increased glucosinolate content can result in higher damage by this insect [42]. Varieties with less wax on their leaves can be partly resistant to *P. xylostella* and *B. brassicae* damage [43]. However, an increase in leaf epicuticular waxes diminishes plant damage by *Phyllotreta* spp. [43].

Although *B. rapa* tends to be quite susceptible to *D. radicum*, some turnip greens/tops accessions have been identified by our group at MBG-CSIC, as they show some resistance to this pest [32]. We noticed that direct damage, as a result of *D. radicum* larvae feeding on root tissue, and indirect damage, by facilitating the entry of secondary root pathogens, reduce both yield and quality of these vegetables and eventually induce plant death (Figure 7).

The main diseases affecting *B. rapa* crops include fungal, bacterial and viral diseases. The most important are downy mildew (*Hyaloperonospora parasitica* (Pers.) Constant.), Turnip mosaic virus (TuMV), clubroot (*Plasmodiophora brassicae* Woronin), and soft rot caused by the bacterium *Pectobacterium carotovorum* (Jones) Waldee (syn. *Erwinia carotovora*) and *Pseudomonas marginalis* (Brown) Stevens, black rot (*Xanthomonas campestris* pv. *campestris* (Pammel) Dowson), (Xcc) and Fusarium wilt (*Fusarium oxysporum* f. sp. *conglutinans/rapae*) [44].

Among these, black rot of crucifers caused by Xcc is considered one of the most important diseases affecting crucifers worldwide. It is particularly destructive to *B. oleracea* vegetables because it causes reduction in yield and quality but it can also attack all other *Brassica* spp. In *B. rapa*, the disease has been reported in Chinese cabbage and other oriental *B. rapa* vegetable crops, and it can also be serious in turnip and turnip greens [45] (**Figure 8**).

A variety of resistance genes and QTLs to different diseases have been identified to develop disease resistance in *B. rapa* [44, 46]. The role of glucosinolate content against Brassica-pathogenic bacteria and fungi has been also reported from *in vitro* and *in vivo* studies, supporting the fact that they can be used as a means of disease resistance [47, 48].



Figure 7. Aspect of turnip greens damaged by cabbage maggot (*Delia radicum*) larvae (left). Turnip tops plants died by the attack of *Delia radicum* under natural infestation. Plants show the most common feeding symptoms with plant yellowing, stunting and slow growth (right).



Figure 8. Black rot, caused by bacterium *Xanthomonas campestris* pv. *campestris* (Pammel) Dowson (Xcc), is considered one of the most serious diseases for crucifers worldwide. The pathogen produces V shaped necrotic lesions from leaf margins, which decrease the quality of product quality for fresh-market sale and cause a decrease in the quality trade for the food industry.

5. Conclusions

In summary, the field and laboratory work carried out at the Institute of Sustainable Agriculture (Cordoba) in collaboration with the Misión Biológica de Galicia from 2009 to date has demonstrated the possibility of producing turnip greens and turnip tops in the Guadalquivir Valley with a performance and quality similar to those of the traditional farming area. The screening and evaluation of a collection of germplasm from the Misión Biológica de Galicia has allowed us to select the most suitable entries to obtain turnip tops with high glucosinolate content, which are beneficial to health and have organoleptic properties similar to those harvested in Galicia. The introduction of *B. rapa* cultivation in Andalusia and other similar regions would increase the diversification of horticultural products and stimulate the consumption of healthy products among the Spanish population. Data reported here might be useful for a deeper understanding of these crops for both nutritional quality and bioaccessibility, resistance to biotic stress, and for selecting varieties adapted to the Mediterranean conditions mentioned in this work.

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Conflict of interest

The authors declare no conflict of interest.

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