

Diplotaxis tenuifolia: Biology, Production and Properties

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ABSTRACT

Perennial wallrocket (*Diplotaxis tenuifolia* (L.) DC) is a perennial herbaceous plant belonging to the *Brassicaceae* native of the Mediterranean area and western Asia, but has now become cosmopolitan. Once harvested as a spontaneous herb, today it is a crop species whose importance is increasing especially in Europe after the diffusion of ready-to-use salads. Leaves present interesting nutritional properties depending on their content in glucosinolates and some antioxidant compounds, such as vitamin C and flavonoids, and their consumption is recommended in the prevention of cancer and cardiovascular diseases. Italy is the main producer country in the world with over 1100 hectares cropped in 2003. However, production in home gardens and harvesting from the wild are still quite significant, which makes figures concerning the overall production underestimated. It is cultivated both in open fields and under plastic tunnels, mostly in rotation with other vegetable crops in areas of intensive horticulture. Besides its relevance as a food crop, it is also thought to possess therapeutic properties that have stimulated its use in the traditional medicine of several peoples in the Mediterranean and the Near East areas. An oil rich in erucic acid can be extracted by the seeds, introducing possible relevance for industrial applications. Aspects concerning biology, diffusion, cultivation, crop protection, industrial processing, nutritional properties and uses of perennial wallrocket are considered in this review.

Keywords: *Brassicaceae*, nutritional properties, perennial wallrocket, ready-to-use salads

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INTRODUCTION

Diplotaxis tenuifolia (L.) DC is one of a group of taxonomically related species belonging to the *Brassicaceae* collectively known as 'rocket' or 'arugula'. In Italy the trivial name of '*rucola selvatica*' (wild rocket) is prevalent, in contrast to the name of '*rucola coltivata*' (cropped rocket), which refers to the species *Eruca sativa* Miller (syn. *E. vesicaria* (L.) Cav.). The specific trivial name of 'perennial wallrocket' is currently preferred to discriminate it from other species in the genus *Diplotaxis*, while the name of 'Lincoln weed' or 'sand rocket' is more commonly used in Australia where the species is mainly known as a problematic weed (Parsons and Cuthbertson 1992).

The economic interest in cropping perennial wallrocket is growing due to the progressive diffusion of ready-to-use salads, the so-called 'fourth generation of vegetables', better known in Italy as '*IV gamma*'. This commercialization technique preserves the freshness and typical scent of the leaves, thus increasing their shelf life and market availability. Until a few years ago *E. sativa* was the only rocket species to be cropped, while *D. tenuifolia* was mostly harvested as a spontaneous herb. Recently, in most cropping areas the latter is more and more replacing the former thanks to its smoother and more succulent leaves that have encountered a higher preference of consumers. Aspects

concerning biology, properties, cultivation, processing and uses of the product are reviewed in this paper.

BIOLOGY, TAXONOMY, DIFFUSION

The genus *Diplotaxis* (tribe *Brassicae*, subtribe *Brassicinae*), phylogenetically close to the genus *Brassica* (Warwick and Sauder 2005), includes about 30 species mainly originating from the Mediterranean area, especially from the western part of the basin which is considered its centre of diversity (Martinez-Laborde 1996). Species of *Diplotaxis* are quite similar morphologically, and all of them present leafy to subscapose stems bearing entire to pinnately lobed or toothed leaves. Distinctive features for *D. tenuifolia* and a few related species are represented by brochidrodromous yellow petals and siliques with a seedless beak. Moreover, unlike most species that are annual, *D. tenuifolia* is characterized by a perennial growth habitus which makes it more suitable as a crop plant in that novel foliage can develop after repeated harvestings by the adventitious buds scattered on its roots. The taxon *D. cretacea*, formerly considered a subspecies of *D. tenuifolia* more widespread in Ukraine and southern Russia (Sobrino Vesperinas 1996), is now recognized as a separate species (Martin and Sanchez-Yelamo 2000). The species *Diplotaxis muralis* (L.) DC is also edible, but it is mainly collected from the wild and has not spread

in cultivation so far; its trivial name of 'wall rocket' is indicative of a close similarity with *D. tenuifolia*, from which it can be distinguished by more oval cotyledons, thinner leaves, pale yellow flowers with smaller petals and the presence of bristly hairs on the lower part of the stem (Bianco 1995; Martinez-Laborde 1996). The two species also differ in their breeding systems (Eschmann-Grupe *et al.* 2004b), and their karyotype: *D. tenuifolia* is diploid ($n=11$), while *D. muralis* ($n=21$) is an allotetraploid deriving from hybridization between *D. viminea* (L.) DC ($n=10$) and *D. tenuifolia* itself (Eschmann-Grupe *et al.* 2004a). The allotetraploid condition is also supported by observations on the seed coat pattern (Koul *et al.* 2000) and by studies concerning the photosynthetic type, which is C3 in *D. viminea* and C3-C4 intermediate in *D. tenuifolia* (Ueno *et al.* 2006). The latter system is more efficient and allows an enhanced potential for CO₂ assimilation (Apel *et al.* 1996).

D. tenuifolia is well adapted to harsh and calcareous soils and in Italy, unlike *E. sativa* which is more widespread in the inner regions, it spontaneously thrives along coastal areas at altitudes below 400 m (Pignone 1996). Sensitivity to excess boron in alkaline soils has been reported, as symptoms of toxicity were observed at a concentration of 12.2 mg kg⁻¹ (Choi *et al.* 2006a). Data concerning diffusion of perennial wallrocket as a crop plant are fragmentary since it is often included in statistics concerning the quite heterogeneous category of 'salads', and rarely considered separately by *E. sativa*. Moreover, incidence on the overall production by cultivation in home gardens and small plots, other than harvesting from the wild, is still quite notable. Italy is the most important producer country in the world with 1,178 ha cropped in 2003, 1,100 of which are under plastic tunnels; Campania is the leading region with almost 3/4 of the total surface (850 ha), mostly concentrated in the Piana del Sele area, followed by Veneto with 120 ha (Pimpini *et al.* 2005).

Besides cropping reasons, the current cosmopolitan distribution of *D. tenuifolia* is the result of its capacity to adapt and ease of propagation, and its invasive behaviour has been regarded as a case study (Hurka *et al.* 2003). Its strong competitive ability in floristic communities is also related to the production of allelopathic substances, such as S-glucopyranosyl thiohydroximate (Giordano *et al.* 2005). In Australia the species has become particularly widespread in some areas and is now considered a noxious weed, as it has invaded and dominates poor pastures in such a way to be retained responsible for possibly poisoning livestock (Parsons and Cuthbertson 1992). Similar problems have also been reported from Argentina (Caso 1972; Ziller *et al.* 2005) and in north America, where its spread in New York city and vicinities in connexion with ship trading from Europe was reported since the nineteenth century (Brown 1879). The relevance as a weed also determines a reasonable concern for risks about the possible transfer of genes for herbicide resistance which have been introduced in some cultivated *Brassicaceae* known to be able to hybridize with *D. tenuifolia* (Rieger *et al.* 1999; Siemens 2002).

BIOCHEMICAL PROPERTIES

Leaves of perennial wallrocket are characterized by a bitter or pungent taste depending on their content of glucosinolates, and a strong acrid aroma which probably derives by the release of volatile isothiocyanates, such as methyl-thiobutyl-isothiocyanate (Delaveau and Paris 1958). Many other volatile compounds are known to contribute to the scent of leaves of *E. sativa* (Jirovetz *et al.* 2002), while no additional data are available for *D. tenuifolia*. The volatile compound 5-dimethylsulphonium pentanoic acid, characterized for the first time from flowers of *D. tenuifolia* (Larher and Hamelin 1979), has been actually proven to be an extraction artefact (Hanson *et al.* 1993).

Glucosinolates are a class of β -thioglucoside-N-hydroxysulfates typical of the *Brassicaceae* including about 120 compounds which differ by the chemical structure of their

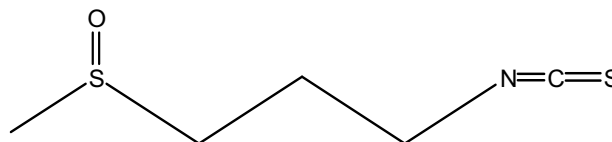


Fig. 1 Molecular structure of sulforaphane.

side chain and release isothiocyanates upon enzymatic hydrolysis (Fahey *et al.* 2001). *D. tenuifolia* is reported to contain high amounts of 4-mercaptobutylglucosinolate (glucosativin) and lower levels of 4-methylthiobutylglucosinolate and 4-methylsulfinylbutylglucosinolate, respectively also known as glucoerucin and glucoraphanin; the latter is more abundant in the flowers, while roots contain 4-hydroxybenzylglucosinolate (sinalbin) (Bennett *et al.* 2006, 2007). Other minor compounds are 3-butenylglucosinolate (glucoraphanin) and some indolylglucosinolates (Nitz and Schnitzler 2002). The aglycone breakdown product of glucoraphanin, named sulforaphane (Fig. 1), has a recognized cancer chemopreventive activity (Zhang *et al.* 1992). However, glucoerucin may also convert to glucoraphanin (Iori *et al.* 1999), and glucosinolates themselves typically possess some extent of anticancer activity (O'Hare *et al.* 2005). Glucosinolate content of rocket leaves depends on genetic traits, but it is also influenced by cropping practices. In particular the total content of glucoerucin and glucoraphanin increases significantly with the number of cuts (Nitz and Schnitzler 2002). Therefore the physiological nutrient value of the product evaluated under this aspect improves in the successive harvest rounds. Isothiocyanates released by the glucosinolates upon their enzymatic degradation are known to protect the plant against many biological adversities. This process has stimulated to exploit the potential of crop residues of cruciferous species as biofumigants (Kirkegaard and Sarwar 1998). In the case of *D. tenuifolia* these properties do not seem to be consistent, since there is an experimental evidence that suppression of mycelial growth of the pathogenic fungus *Rhizoctonia solani* only occurs when soil is amended with very high amounts of green manure (10% w/w) (Yulianti *et al.* 2006).

The nutrient content of perennial wallrocket is similar to other salads, except in that dry matter and fibre content are slightly higher. Leaves present high levels of potassium (4.7 g kg⁻¹ fresh product), calcium (3.1 g kg⁻¹) and iron (52 mg kg⁻¹) (Pimpini *et al.* 2005). The content of some vitamins, particularly vitamin A (7.4 g kg⁻¹) and ascorbic acid (vitamin C) (1.1 g kg⁻¹), is also notable (Bruno *et al.* 1980; Pimpini *et al.* 2005). The latter substance and flavonoids confer antioxidant properties; therefore nutritionists more and more recommend the inclusion of rocket leaves in diets as an aid for preventing cardiovascular and cancer diseases (Salvatore *et al.* 2005). With the exception of roots, all *D. tenuifolia* tissues contain polyglycosylated flavonoids whose core aglycon is quercetin, kaempferol, or isorhamnetin (Maldoni *et al.* 1988; Sanchez-Yelamo 1994; Bennett *et al.* 2006); besides a role in protection from UV-radiation, their biological function is thought to be primarily related to resistance to pathogens and pests (Treutter 2005). Therefore, increasing flavonoid content, that expectantly represents one of the primary objectives in perennial wallrocket breeding, is likely to have beneficial side effects for plant protection. There is a seasonal influence on flavonoid content, being higher in spring-harvested leaves (di Venere *et al.* 2000). Antioxidant properties also characterize the alkaloid sinapine (4-hydroxy-3,5-dimethoxy-cinnamic acid choline ester) contained in *D. tenuifolia* seeds, which however is better known as an antinutritional factor (Boucherau *et al.* 1991; Bennett *et al.* 2006).

Potential pharmaceutical interest relies on some compounds that have been recently extracted by perennial wallrocket leaves, such as some nortropane alkaloids derived from pseudotropine known as calystegins, especially calystegin A5 (Brock *et al.* 2006), and an essential oil containing 5-methylthiopentanenitrile possessing antifungal properties

(Rodriguez *et al.* 2006).

USES OF PRODUCT

The use of perennial wallrocket as a vegetable is known at least since the 19th century in France and Italy (Martinez-Laborde 1996). Besides being an ingredient of salads, it is now used for many dishes as a garnish, and as a pizza topping. Older leaves are too piquant to be consumed raw, but can be added to soups or sauces. As for other cruciferous species, breeding may allow the regulation of glucosinolate content to obtain a product that can adapt to the different consumer tastes. So far the number of cultivated varieties seems quite low if compared to the increasing market demand. Therefore some efforts have been made, or are in progress, to collect new germplasm throughout the Mediterranean area, in the awareness that a targeted genetic improvement may provide very profitable results (Pignone 1996). A liquor prepared in some areas of Campania is claimed to have digestive and diuretic properties and is also to be considered within the food uses.

Other potential uses of perennial wallrocket pertain to the oil extracted from its seeds in a yield of about 36%. It contains 12.6% oleic acid, 22.3% linoleic acid, 38.9% linolenic acid and 11.8% erucic acid (Dolya *et al.* 1972; Agullo *et al.* 1987). Although being lower than those reported in the oil extracted from other cruciferous crops, the high levels of erucic acid are counter to its use for human consumption. Properties are similar to the oil extracted by *E. sativa* (jamba oil) that, especially in the Indian subcontinent, is employed as a lubricant in industries, and in the preparation of soaps or as an illuminating agent (Pignone 1996).

Besides the previously discussed beneficial effect for human health related to the content in sulphoraphane, glucosinolates and flavonoids, the use of *D. tenuifolia* as a medicinal plant is widely documented in the tradition of several peoples and cultures, and even nowadays it is still recommended somewhere, mainly as an aphrodisiac or an anti-diarrhoeic herb (de Feo *et al.* 1992, 1993; Bianco 1995; Pignone 1996; Pieroni *et al.* 2004). Mucolytic effects are also reported (Caudron 2005).

CROPPING

D. tenuifolia can be cultivated in open fields, or more commonly under plastic tunnels (Fig. 2), even as a soilless or hydroponic crop. In open cultivation soil must be ploughed, or better spaded in the case of sandy soils, reaching a depth of about 30 cm. Harrowing usually completes the soil preparation, but excessive pulverization should be avoided. Soil preparation ends with the formation of ridges (1-3 m in width). A seed quantity of 4-6 kg ha⁻¹ is used, depending on the sowing period and modality. Rolling should follow seeding in light soils. The sowing period varies from spring to early autumn depending on commercial requirements. In summer, preliminary irrigation is suggested before starting the preparation which improves soil structure and facilitates rapid germination and uniform emergence, besides contributing to the control of weeds (false sowing). Alternatively, the crop can be transplanted (Fig. 3), with an ensuing reduction of the cycle that generally implies a better weed and pest management and an improved quality of the product. Plantlets are reared for a period of 20-30 days in alveolar containers filled with a peat-vermiculite seedling medium (about 10 seeds per hole). Transplanting can be operated after mulching with a black polyethylene film under which a sprinkler is placed in alternate rows. As for other vegetable crops, monosuccession is not advisable; plants such as bean, celery, cucurbits and solanaceous crops are also not considered a good precession since they have a detrimental effect on yield (Bianco 1995; Pimpini and Enzo 1996).

Nutritional requirements of perennial wallrocket are modest, as 100 kg of product remove estimated amounts of 0.28 kg of nitrogen, 0.11 kg of phosphoric anhydride and



Fig. 2 Cropping under plastic tunnel is the most widespread system in Campania.



Fig. 3 Plantlets 1 week after transplantation.

0.34 kg of potassium oxide (Pimpini *et al.* 2005). Of course fertilization must be operated considering the soil conditions, as well as its content of organic matter and macroelements, which can be also administered by fertirrigation. Several studies have shown that nitrogen supplying should never exceed 100 kg ha⁻¹ (Bianco and Boari 1996; Pimpini and Enzo 1996). In fact, leaves of *D. tenuifolia* can accumulate large amounts of nitrate, up to 10 g kg⁻¹ fresh weight (Santamaria *et al.* 2001), as their nitrogen use efficiency is quite low (Santamaria *et al.* 2002). Moreover, excessive nitrogen worsens the quality of the product during storage; therefore its administration must be strictly avoided in proximity of the harvesting time, especially in soilless cropping.

Perennial wallrocket is adapted to harsh and arid soils, and the presence of a well-developed taproot reaching up to 40 cm in depth suggests a limit to irrigation; in fact, excess water may worsen the sanitary status of the crop, particularly with respect to the incidence of fungal diseases. However, it must be also considered that a good availability of water is essential to ensure an optimum quality of leaves, which otherwise become too fibrous and less succulent. Irrigation is commonly based on a sprinkling system operating at medium volumes (120 L h⁻¹) and ranges (3-5 m), but dripping systems are more indicated to safeguard the foliage sanity and to avoid contamination by microbes dangerous for human health.

As discussed above, *D. tenuifolia* is known to be a good competitor; however weed development can be very detrimental to the quality of the product, as the absence of contamination by other herbs is regarded as a fundamental requisite. Therefore an onerous selection operation must be effected in case weed management is not adequate. Means



Fig. 4 Manual harvesting.

other than the use of herbicides should be preferred, since very few active principles are registered in all countries by reason of the short crop cycle, and their range of action and selectivity are not consistent. Treatments with benfluralin are usually effected before seeding in protected crops in Campania (Nicoletti *et al.* 2007). However the product is unable to control some problematic weeds such as shepherd's purse (*Capsella bursa-pastoris*), and a manual operation before harvesting is often necessary.

The cultivation cycle has a quite variable duration, and harvesting time cannot be established in advance, depending on various and independent factors, such as the climatic conditions and the necessity to fulfil the market demand. These factors, together with the phytosanitary status of the crop, also determine the number of possible successive harvests. Therefore it is quite difficult to make previsions concerning yield. In 2006 an average yield of 27 ton ha⁻¹ has been estimated in the Piana del Sele area, where four harvests were effected on average (Raimo and Miccio, unpublished). Leaves can be cut manually (Fig. 4) or by opportunely adapted mowers, and the most appropriate timing must be decided according to local conditions; in fact, morning harvests are preferable in spring and summer considering the lower temperature and the higher relative humidity, while afternoon is suggested in case there is a concern about nitrate concentration. After harvesting, the product is placed in plastic boxes to be carried to the processing plant. For the preparation of ready-to-use salads leaves must be refrigerated and conditioned in a due time to ensure an adequate shelf-life. Quality control is effected on pesticide residues, heavy metals and nitrate content, and microbial contamination; reference standards concerning the latter parameter are particularly restricted considering the possibility of a postponed consumption.

In soilless cropping a better uniformity of the foliage is obtained, and production can be programmed by opportunely managing the most relevant environmental parameters (temperature, humidity, fertilization, etc.). In Italy, floating system is the most widespread technique employed for *D. tenuifolia*; it allows a substantial reduction of the incidence of foliar diseases and weed contamination, and the product is almost clean of any kind of foreign bodies and may only require a quick washing before packaging. A thorough account concerning the various implications of this cropping system is beyond the scopes of this paper and we address the reader to appropriate publications for further documentation.

DISEASES AND PESTS

The development of the demand for ready-to-use vegetables has led to a remarkable intensification of cropping, with ensuing increasing problems concerning diseases and pests. Most reports on biological adversities of perennial

wallrocket concern observations effected in Italy; nevertheless they represent a valuable reference for other countries with similar climatic conditions where the crop has been introduced more recently.

Particularly, soil-borne pathogens represent a widespread problem in all Italian regions (Fig. 5). In fact, their inoculum accumulates in the soil and real outbursts may occur in a few years. Repeated cropping is regarded as the main causal factor for the epidemics of crown and root rot recently observed in the Piana del Sele area, caused by *Rhizoctonia solani* AG-4 (Nicoletti *et al.* 2004). Infected plants undergo either an acute (damping-off) or a chronic course, with necrotic lesions developing on one side of both crown and taproot, and leaf yellowing. *Sclerotinia sclerotiorum* is also a reported agent of crown and stem necrosis (Garibaldi *et al.* 2005; Minuto *et al.* 2005), but it can be easily recognized because infected tissues become soft and watery, and are covered by white mycelium and dark sclerotia. Unlike *R. solani*, this pathogen has a higher incidence at lower temperatures (15°C) and high relative humidity conditions. Outbursts of *Fusarium* wilt in plastic tunnels were recorded in northern Italy after 3 years during which crop yield had progressively declined (Garibaldi *et al.* 2002, 2003). Disease symptoms consist in reduced development, leaf chlorosis, epinasty and wilting, and a necrosis of vascular tissues of taproot is evident at an advanced stage. The causal agent, *Fusarium oxysporum*, is quite heterogeneous since isolates from diseased plants were found to belong to two different *formae speciales*, *conglutinans* and *raphani* (Catti *et al.* 2007). A similar onset of the disease was observed in distant farms in the same area that is indicative of the ability of the pathogen to propagate through the seeds (Garibaldi *et al.* 2002).

Seeds are also a vehicle for the diffusion of downy mildew caused by *Hyaloperonospora parasitica*, the fungal pathogen that is possibly to be regarded as a limiting factor for perennial wallrocket. *H. parasitica* commonly affects winter cruciferous crops in Italy without causing relevant damages. In the case of *D. tenuifolia*, the protected environment represents a favourable micro-climate that often determines its rapid spread (Minuto *et al.* 2004). Symptoms appear as small, irregular, dark brown speckling on the upper surface of the leaves. The speckled areas usually expand into larger spots and fruiting structures appear as a whitish mould on both the leaf surfaces. Leaves undergo a sudden yellowing, and rot in the case of high infestations. The disease may be quite underhand, as its signs can appear after harvesting on the market shelves; in fact the pathogen continues developing even at low temperatures that have the only effect to prolong its incubation period, otherwise lasting no more than 12 days in favourable conditions (Garibaldi *et al.* 2004). Therefore an economic threshold is often reached and control practices are recommended. Another foliar disease that



Fig. 5 Missing plants is one of the visible effects of damage caused by soil-borne fungal pathogens.

is quite common on winter cruciferous crops in temperate countries is white rust caused by *Albugo candida*. It is quite a heterogeneous pathogen (Choi et al. 2006b) that causes local infections on the leaves resulting in the appearance of white pustules on the lower surface corresponding to tan-yellow pustules at the upper surface. Tissues around the pustules may become necrotic and bring leaves to senescence. This species is particularly noxious in India on oil-seed crops since it can cause systemic infections of the inflorescence, often in association with *H. parasitica* (Gupta et al. 2004). Other fungal diseases occasionally observed in Italy are powdery mildew caused by *Erysiphe cichoracearum*, and *Alternaria* leaf spot (Nicoletti et al. 2007). The latter disease is mainly caused by *Alternaria brassicicola*, as some degree of resistance has been described toward the other species infecting *Cruciferae*, *A. brassicae* and *A. raphani* (Klewer et al. 2002; Sharma et al. 2002). Resistance has also been reported toward blackleg caused by *Leptosphaeria maculans* (Delourme et al. 2006), while some concern exists for another disease of general occurrence on cruciferous crops, clubroot caused by *Plasmodiophora brassicae* (Voorrips 1995). Its first visible symptom is usually wilting, particularly during hot-dry weather. Diseased plants are generally stunted, and the foliage may be different in colour from healthy plants. Infected roots show characteristic swellings or knots, and in case of severe infections taproots of young plants form a single-clubbed root. On *D. tenuifolia* the disease has been reported from New Zealand (Pennycook 1989), and more recently from Switzerland (Buser and Heller 2006); its possible occurrence is to be particularly considered in other central and northern European countries, where the pathogen is quite widespread.

Among bacterial diseases, a special consideration deserves black rot caused by *Xanthomonas campestris* pv. *campestris*, that is reported on most weedy crucifers (Westman et al. 1999) and has been recently found on *D. tenuifolia* in Campania (Raio and Giorgini 2005). Infected leaves typically stand out for their yellow colour at the upper surface, while the lower surface turns dark, almost black. Symptoms diffuse along the main vein, and necrosis may even reach the stem. The pathogen spreads through the seeds and irrigation. Phytoplasmas (Amici 1972) and a number of viruses, such as cucumber mosaic cucumovirus (CMV), turnip yellow mosaic tymovirus (TYMV), turnip mosaic potyvirus (TuMV) and cauliflower mosaic caulimovirus (CaMV) (Stavolone et al. 1998; Nicoletti et al. 2007), are also reported to be able to infect *D. tenuifolia*, but no substantial damage caused by these pathogens has been reported so far. Latent infections reported on other *Diplotaxis* species may confer to perennial wallrocket an epidemiological role in the spread of other viruses too, such as tomato spotted wilt tospovirus (TSWV) and *Pelargonium* zonate spot anulavirus (PZSV) (Lupo et al. 1991; Parrilla et al. 2003; Gallitelli et al. 2004).

After the recent phasing out of methyl bromide, control of cryptogamic diseases has become quite problematic on several vegetable crops. On perennial wallrocket the problem is even more complicated by the fact that no resistant cultivars have been selected toward any particular biological adversity yet. Chemical products currently available for soil disinfestations, such as metham sodium and dazomet, are reported not to be as effective. Dichloran is active against *S. sclerotiorum*, but treatments must be operated early in the cropping cycle considering its long persistence (20 days). Besides paying more attention to certain cropping practices, such as rotation and irrigation, farmers should consider any possible action to avoid the accumulation of pathogen inoculum in the soil. Solarization is an effective technique to be proposed in the subtropical and temperate countries, provided that it does not impair the farm organization and the rotation programs. The use of biological control products based on mycoparasites, such as *Coniothyrium minitans* against *S. sclerotiorum* and *Clonostachys rosea* against *F. oxysporum* or *R. solani*, is currently being proposed and is particularly convenient when

the crop is transplanted since these micro-organisms can be administrated in advance and, once established in the rhizosphere, may be able to prevent the pathogen infestation in the field. It may be advisable to use such products even in the case of a low disease incidence because the mycoparasites can exert their antagonism also in the absence of the crop against both the resting structures of the pathogens (e.g. *S. sclerotiorum* sclerotia) and their saprophytic development on the organic matter and crop residues (Nicoletti et al. 2007). Another environmentally friendly practice to be considered consists in the use of phosphites for leaf fertilization; in fact they are known to stimulate phytoalexin production whose effectiveness in plant protection against fungi is well-known (Guest and Grant 1991). Chemical control may be operated against downy mildew by using metalaxyl M + copper oxychloride, but iprovalicarb + copper oxychloride is generally preferred since it presents a very short interval (7 days, or slightly longer at low temperatures). Other products reported to have the same reduced persistence are tolylfluand and azoxystrobin. Copper salts, especially tetramic sulphate, present the shortest interval and are effective against the black rot agent, but their use is not suggested since stains persist on the foliage. Another product occasionally used is thiram, which is also effective against *Alternaria* leaf spot and can be used for seed treatment. Seed disinfection is not a routine practice; however, the definition of a protocol for their sterilization would be extremely useful, considering the number of pathogens which propagate by such means. Seed immersion in vinegar for 15 min followed by drying at 25-30°C is effective against downy mildew and recommended in Switzerland (Buser and Heller 2006).

Pest reports on *D. tenuifolia* generally refer to insect species common on other *Brassicaceae*, such as aphids (*Brevicoryne brassicae*, *Myzus persicae*, *Lipaphis erysimi*), flea beetles (*Phyllotreta* spp.), click beetles (*Agriotes* spp.), cabbage butterflies (*Pieris* spp.), and cutworms (*Mamestra brassicae*, *Autographa gamma*, *Spodoptera littoralis*) (Bianco 1995; Nicoletti et al. 2007). Other *Lepidoptera* that have recently caused economic damage in Italy are the diamondback moth (*Plutella xylostella*) (Ciampolini et al.



Fig. 6 Leaf riddling caused by springtails (*S. viridis*).

1998) and the cabbage webworm (*Hellula undalis*) (Ciampolini *et al.* 2001a). Flea beetles, also indirectly injurious as vectors of TYMV, can be particularly harmful in mild climates, where infestations start to occur early in spring. In fact leaf erosions are caused by adults, which represent the overwintering stage, while larvae only feed on roots without causing a consistent damage (Ciampolini *et al.* 2001b). The milder climatic conditions occurred in winter in the past few years in the Piana del Sele area have also determined a noticeable spread of springtails (*Sminthurus viridis*) (Raimo *et al.* 2005), whose feeding habit also causes riddling and erosion of leaves that become unmarketable (Fig. 6). Besides representing a direct economic loss, the presence of damaged leaves requires a quite onerous choice operation. Control practices must focus on an accurate weed management, especially in the surrounding ridges which represent an ideal micro-environment for springtail pullulation. As anticipated above, the use of pesticides on perennial wallrocket is quite problematic due to a very short interval between subsequent harvestings, and highly toxic or persistent chemicals cannot be used. Therefore pyrethroids (e.g. deltamethrin, bifenthrin), that have proved to be very efficacious against aphids, flea beetles, springtails and cutworms, are mostly employed. However their indiscriminate use can be deleterious, since it may favour the proliferation of spider mites (*Bryobia* spp.) that have been reported to be injurious to the crop in several Italian areas (Giorgini 2001; Laffi 2001). An alternative is represented by spinosad or etophenprox which present a 3- and a 7-day interval respectively. The low toxic product azadirachtin, also active against aphids and *Lepidoptera*, has showed a substantial antifeedant effect on flea beetles (Ciampolini *et al.* 2001b), and its use is suggested when they occur as the key pest. Products based on *Bacillus thuringiensis* are efficacious against *Lepidoptera*, and allow to respect some parasitoids, such as *Angitia tibialis* and *Apanteles* spp., whose ecological role as control agents may spontaneously establish (Ciampolini *et al.* 1998). Slugs (e.g. *Deroceras reticulatum*, *Arion* spp.) are also occasionally injurious and should be particularly considered in subtropical countries for their ability to transmit a number of human parasitic *Trematodes* (e.g. *Fasciola hepatica*). Baits treated with methiocarb or methaldehyde are usually efficacious against these pests (Nicoletti *et al.* 2007).

INDUSTRIAL PROCESSING

After harvesting, rocket leaves undergo a leakage of cell juices that support microbial growth. Many enzyme systems are activated which are responsible of the loss of nutritional properties and other modifications in flavour and consistency. Respiratory activity also increases, and CO₂ is produced at levels reaching 42 and 113 mL kg⁻¹ h⁻¹ at 0°C and 5°C respectively; ethylene develops in low amounts, that is less than 0.1 mL kg⁻¹ h⁻¹, but leaves are highly sensitive and soon undergo chlorophyll degradation and yellowing (Cornacchia *et al.* 2006). Therefore storage at low temperatures (at least 5°C) must be operated soon after harvesting, as any delay results in a reduction of the shelf life (Sinigaglia *et al.* 1999). Perennial wallrocket is resistant to coldness, and temperatures of 0-2°C can be borne for periods up to 10 days (Cornacchia *et al.* 2006).

As for other leaf vegetables, there is a great concern for possible microbial contamination. Microbiological analyses of the product provide for the absence of human pathogenic bacteria, such as *Listeria monocytogenes*, *Salmonella* spp., *Yersinia enterocolitica*, and a presence of *Staphylococcus aureus*, *Escherichia coli* and other coliforms under the limit of 100 cfu g⁻¹ (Galli and Franzetti 1998; Aruscavage *et al.* 2006). *Aeromonas* spp. have also been found on *D. tenuifolia* leaves (Villari *et al.* 2000). Therefore a greater attention must be paid to the quality of water for both irrigation and washing. Sanitization can be operated by accurate washing with cold tap water supplemented with a disinfectant. Chlorine products are prevalently used (Marchetti *et al.*

1992), but good effects have also experimentally resulted by treatments with lactic acid (20 mL L⁻¹) and peroxyacetic acid (300 mg L⁻¹), which inhibited the microbial growth throughout the shelf life (Martinez-Sanchez *et al.* 2006a).

As introduced above, the technology of the fourth generation of vegetables aims at preserving the visual quality of the product and its organoleptic properties for a prolonged period, that is at least a week. The product is packaged with appropriate films presenting differential permeability toward oxygen and carbon dioxide; in fact, low O₂ or high CO₂ levels are capable to reduce ethylene biosynthesis and chlorophyll loss (Kader 1986). A controlled atmosphere may be also artificially created in the plastic bags with even higher CO₂ and a lower O₂ concentrations; however, a weight loss is inevitable and has been documented for perennial wallrocket after 10 days storage under 3% O₂ and 15% CO₂ (Cornacchia *et al.* 2006). Experimental data concerning modifications in the nutritional value are controversial; in fact, a decrease in ascorbic acid, flavonoid and glucosinolate content was determined under a low O₂ (1-3 kPa) and high CO₂ (11-13 kPa) atmosphere (Martinez-Sanchez *et al.* 2006a), while the total flavonoid content was not affected, or even increased, under a controlled atmosphere of 5 kPa O₂ and 10 kPa CO₂ (Martinez-Sanchez *et al.* 2006b). Further studies are therefore necessary for a more definitive conclusion about the efficacy of storage of perennial wallrocket leaves under controlled atmosphere and the most appropriate levels of CO₂ and O₂.

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