Brassica villosa a Potential Tool to Improve the Insect or Disease Resistance of Brassica Crop Species

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Abstract

Outward growths on plant epidermal cells are known as trichomes or hair cells and they act as a barrier for plant predators. Brassica napus (canola) is the major oil seed crop in the world but lacks trichomes on them but a wild relative of canola, Brassica villosa has dense trichomes on them. Transcriptomic study of B. villosa indicated differential expression of trichome, photosynthesis light reactions, major carbohydrates, cellulose, lipid and amino acid metabolism, sulfur assimilation, metal handling/binding, hormones, biotic stress, redox, RNA regulation/ transcription, post-translational modification, signalling, cell vesicle transport, development, secondary metabolism and miscellaneous genes. And bio-chemical results from B. villosa trichomes confirmed accumulation of metals and a unique alkaloid-like compound in them. These results from B. villosa opened the doors for using this species as a potential tool for the improvement of insect or disease resistance in Brassica crop species.

Keywords: Trichomes; Hormones; Metabolites; Microarray

Short Communication

Outward growths on plant epidermal cells Levin [1], Mathur [2] are known as trichomes or hair cells. Trichomes can act as a barrier for plant predators, such as insects. As well, they provide a “blanket” that assists plants against dehydration Levin [1]. In nature, there are two types of trichomes, one with glands which excrete defensive or attracting metabolites and accumulate metals, and a second non-glandular type that mainly provides a physical barrier.

Brassica napus is one of the world’s major oil seed crops and has very sparse (i.e., semi-glabrous) trichome coverage and susceptibility to insect pests and disease. Its wild relative B. villosa is a perennial weed with an exceptionally dense coverage of trichomes (>2,172 per cm²) Gomez Campo and Tortosa [3] and resistance to many pests. Attempts at reciprocal crossing between B. villosa and B. napus have resulted in only a few flowers and empty pods (Nayidu, unpublished), but segments of the B. villosa genome have been introduced into B. oleracea (broccoli) using QTL tracking Mithen et al. [4].

Therefore, global gene expression data from the first emerging leaf of these two species was compared using a B. napus microarray to determine whether B. villosa could serve as a candidate species for characterizing trichomes in Brassica species. Preliminary data indicated that 78.5% of B. villosa paired-end RNA reads easily mapped to a B. oleracea reference genome, a value similar to mapped B. oleracea (81.9%) and B. napus (73%) young leaf transcripts Nayidu et al. [5], in preparation.

Although not all B. napus genes were represented on the microarray (since B. napus was not sequenced at the time), a total of 94 trichome genes showed differential expression, with 35 more strongly expressed and 59 more weakly expressed in B. villosa emerging leaves compared with B. napus leaves Nayidu et al.[6]. Trichome positive regulatory genes (GL1, GL2, and EGL3) and two negative regulators (TRY and ETC1) also had more abundant transcripts in B. villosa than in B. napus leaves, suggesting a different mode of regulation than in the Arabidopsis trichome model. Other non-trichome differentially expressed transcripts in trichome-bearing B. villosa leaves included those coding for photosynthesis light reactions, major carbohydrates, cellulose, lipid and amino acid metabolism, sulfur assimilation, metal handling/binding, hormones, biotic stress, redox, RNA regulation/transcription, post-translational modification, signalling, cell vesicle transport, development, other transporters, secondary metabolism (particularly phenolics, glucosinolates, and alkaloids), and miscellaneous genes. Although protocols for plant transformation are not available for B. villosa, regulatory genes for specific traits in this species can be studied indirectly by selection for strong differential expression relative to B. napus in leaf microarray data or leaf and cotyledon RNA sequencing data (“The latter has recently been deposited in NCBI databases”). Selected genes could then be used for identifying useful phenotypes in B. villosa expression lines featuring B. villosa transgenes or knockdown cassettes of homologous B. napus genes. Five of the major trichome regulatory genes from B. villosa Nayidu et al. [6], have been already introduced into B. napus. Alternatively, B. villosa mutant populations could be developed to study genes directly if a shorter method for generating large amounts of seed was in place. At present, B. villosa seeds are either collected from natural sources or produced in small quantities after repeated rounds of vernalization to induce B. villosa flowering.

Trichomes are considered to be organs that store metals and secondary metabolites or detoxify undesirable metal concentrations in plants. Isolated B. villosa trichomes were confirmed to accumulate metals, using ICP/MS analysis, and HPLC-MS and NMR confirmed a unique UV-absorbing 4-N alkaloid-like compound (C_{25}H_{26}N_{4}O_{2}; absolute mass 414.2046 Da) Nayidu et al.[5]. Metal accumulation in trichomes may assist B. villosa to manage the calcium-laden rocky slopes of its native habitat Snogerup et al. [7]. Heavy metals and alkaloids are known to impact negatively on insects van Ooik et al.[8]; Wink [9,10].

The firmly attached, flexible trichomes, their contents, and the presence of transcripts specifying trichome development and a host of other relevant

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genes indicate that *B. villosa* leaves have a variety of protection strategies. The data confirms this species as a biochemical system for studying genes, proteins and metabolites of importance to plants.

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