Breeding of Salad Rocket (\textit{Eruca stiva} Mill.) Varieties for Healthy Functional Proposes

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Abstract

Breeding new variety of rocket with low nitrate content and high \(\alpha\)-glucosidase inhibitory can help the balance in human diet. Two outstanding lines which considered to be used as Ssam and green for Bibimbap were finally selected, and labeled as ‘Kangwondae Ssam’ and ‘Soyangkang Girl’ respectively. ‘Kangwondae Ssam’ is a new rocket line developed from a low nitrate content genotype (R131) with a nitrate content of 1050 mg/kg of fresh weight. The pungency of leaf is mild and its \(\alpha\)-glucosidase inhibitory is 14.9\%. The young plants of ‘Kangwondae Ssam’ have a big size of leaf. ‘Soyangkang Girl’ is a new rocket line breed from genotype R96 with a nitrate content of 1680 mg/kg of fresh weight. The young plants of ‘Soyangkang Girl’ have a nice tooth type of leaf and the pungency of leaf is stronger. \(\alpha\)-glucosidase inhibitory activity of ‘Soyangkang Girl’ is high (29\%).

Key words: Nitrate, Salad rocket, Selecting, \(\alpha\)-glucosidase inhibitory.

Introduction

Rocket (\textit{Eruca sativa} Mill.), which belongs to the \textit{Brassicaceae} family, was cultivated in southern Europe, north and north-east Africa, the Middle East, central Asia and north central India (Warwick et al., 2007). Rocket is important leafy vegetables and cultivated vegetable most of countries where they are used (either wild or cultivated) in many ways, as condiment in salads, cooked vegetables, medicinal and functional plants (Kim et al., 2004). Rocket is a nutrient rich plant, contains a range of health-promoting phytochemicals including carotenoids (pro-vitamin A), vitamin C, folate, fibres, polyphenols, flavonoids and glucosinolates (glucoerucin). It could be classifies as a functional foods (Milan et al., 1996). Research works had proved that rocket has the varies health functions including astringent, diuretic, digestive, emollient, tonic, stomachic, depurative, laxative, rubefacient, stimulant and aphrodisiac (Yaniv et al., 1998; Abu-Rabia, 2005). Rocket has the anthepatotoxic effect on alcohol induced liver injury and anti-cancer (Hussein et al., 2010).

Nowadays, overweight and obesity become a big issue in public health and economic development. Overweight and obesity are the result from unbalance of energy intake and energy expenditure of human body. Controlling the obesity, diabetes mellitus and hypertension through dietary management by consuming plant food and their ingredients could be a more effective strategy because of likelihood of high compliance and absence of side effects (Kwon et al., 2007). Vegetables are common in human dietary and have good nutrient benefits with low calories, it is a good strategy to consume the vegetables with additional function of inhibition to \(\alpha\)-glucosidase and \(\alpha\)-amylase. As glucosidase and amylase play a significant role in the digestive process of dietary complex carbohydrate, the blockage of carbohydrate etabolism by amylase or glucosidase inhibitors might benefit obesity treatment. With respect to suppression of glucose production from carbohydrate and glucose absorption from the intestine, the efforts to investigate and find potential inhibitors of \(\alpha\)-glucosidase and \(\alpha\)-amylase in...
natural products are increasing. The $\alpha$-glucosidase inhibitory had been fund in many medicine herbs, such as Cassia obtusifolia (Kang et al., 2012), Rubus coreanus (Ju and Han, 2010), Bergenia ciliata (Bhandari et al., 2008), Rosa gallica (Ochir et al., 2010), Toona sinensis (Zhao et al., 2009). In addition, Barillari et al. (2005) fund the direct antioxidant activity of purified glucoerucin, the dietary secondary metabolite contained in rocket seeds and sprouts.

Nitrate is one of the forms of nitrogen in nature, and it is found in the environment, such as air, soil, and water. Nitrate is also found in the meats, fishes and vegetables. The ferment vegetables, such as kimchi, have high nitrate contents (Santamaria, 2006; Burns et al., 2011). Nitrate itself is relatively non-toxic but its metabolites may produce a number of health effects (Santamaria, 2006; Mensinga et al., 2003). Vegetables in the families of Brassicaceae (rocket, radish, mustard), Chenopodiaceae (beetroot, Swiss chard, spinach) and Amaranthaceae, as long as Asteraceae (lettuce) and Apiacae (celery, parsley) accumulate high nitrate contents, that may exceed to 2500 mg/kg of fresh weight. The rocket, which exported from Italy to Switzerland and Germany, is required that the nitrate content should be not to exceed 2500-4000 mg/kg of fresh weight (Santamaria, 2006). Bianco et al. (1998) found that it is very difficult to achieve even a reduced amounts of nitrate is used in the cultivation. The nitrate content in rocket could reach up to 9300 mg/kg of fresh product according the survey in Italy (Santamaria et al., 1999).

Thus, the aim of this study is to evaluate the nitrate content and the $\alpha$-glucosidase inhibitory activities in 190 genotypes of rockets, and try to screen out the high activity of $\alpha$-glucosidase inhibitory and low nitrate content rocket genotypes.

Materials and Methods

A total of 190 genotypes of rocket were used in this breeding program. These genotypes were mainly original from Afghanistan, China, Egypt, India, Iran, Italy, Pakistan, and Turkey, and they are grouped as wild materials or cultivated materials, and further separated as genotypes from native area or non-native area (Table 1).

**First year selection:** A total of 190 genotypes of rocket were used in this study (Table 1). The seeds of some accessions were received from the North Central Regional Plant Introduction Station, Iowa, USA, and some were collected or breed by the authors. The seeds of rockets were sown in the nursing trays that was filled with commercial growing substrate and kept in glasshouse. When seedlings had 2-3 true leaves, 20 seedlings for each genotype were transplanted into a hydroponic system for further growth. The leaf samples of rockets were harvested for determination of $\alpha$-glucosidase inhibitory activity when plants have 6 to 7 leaves.

**Second year selection:** From first year experiment, all genotypes of rocket with high $\alpha$-glucosidase inhibitory activity ($\geq$10%) were used in this study. The seeds of rockets were sown in the 32-cell plug trays that was filled with commercial growing substrate and kept in glasshouse. When seedlings had 2-3 true leaves, the weak or deformed seedlings were removed and only one best plant was remained in each plug. Each genotype of rocket was sown in three trays, and kept in the glasshouse. Plants were daily irrigated with a complete nutrient solution containing 250 mg/L of N-NO3 with about 40% drainage to ensure all the plants received even and sufficient nutrients. The leaf samples of rockets were harvested for determination of nitrate content when plants have 6 to 7 leaves. In order to encourage the accumulation of nitrate, 10 days before taking samples, the plants were covered with a shade screen (70% shading) all day to reduce the light intensity. The leaf samples were harvested by hand with a knife in the morning and stored under -72°C to minimize the changes of physiological parameters.

**Third and final year selection:** From first and second year selection, genotypes of rocket which were best records in low nitrate content and high $\alpha$-glucosidase inhibitory activity were selected. In order to selecting best rocket lines, environmental stress study was conducted. The seeds of best rockets were sown in plug trays that was filled with commercial growing substrate and kept on in growth chamber at 40°C for heat tolerant evaluation. The best individuals of survivals were transferred to hydroponic

Table 1. The genotype numbers of rocket in this breeding program

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<thead>
<tr>
<th></th>
<th>Wild</th>
<th>Cultivated</th>
<th>Total</th>
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<tbody>
<tr>
<td>Native Area</td>
<td>5</td>
<td>123</td>
<td>128</td>
</tr>
<tr>
<td>Non-native Area</td>
<td>9</td>
<td>53</td>
<td>62</td>
</tr>
<tr>
<td>Total</td>
<td>14</td>
<td>176</td>
<td>190</td>
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