Registration of High Alpha-Acid Male Hop Germplasm USDA 21267M

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Abstract

USDA 21267M (Reg. No. GP-37, PI 559093) male hop (Humulus lupulus L. var. lupulus) germplasm was released to the public 6 Dec. 2011. This germplasm has been used by the USDA–ARS for hop cultivar development for several decades. Nonagency interest in this germplasm was expressed, and release of this material is considered beneficial for the US public. USDA 21267M arose from a cross, made in 1973 involving the hop cultivar Comet and the USDA–ARS hop male germplasm 21110M. The defining characteristics of USDA 21267M are its high alpha acid levels observed in lupulin glands as well as good breeding value for yield, alpha and beta acid levels, and high xanthohumol levels. It is expected that hop breeders will use this germplasm as a parent for high-yielding, high-alpha hop breeding material. The release of USDA 21267M will give both public and private breeders new germplasm for varietal development.

Hop (Humulus lupulus L. var. lupulus) is a dioecious, perennial crop cultivated worldwide primarily in the Northern and Southern Hemispheres between the 35th and 60th parallels in latitude. Hop is used primarily for bittering and flavoring in beer brewing, with several other uses identified and implemented in recent years (Cornelison et al., 2006; Pollach et al., 1996; Stevens and Page, 2004; Zanoli and Zavatti, 2008). The harvested portion of hop is the female floral structure called the strobiles—commonly called hop cones. The plant structural component containing almost all of the important chemical constituents for brewing are lupulin glands and are present on the surface of both male and female hop flowers.

The two major hop-producing nations in the world are the United States and Germany. Hop production in United States resides principally in the Pacific Northwest, with Washington being the main hop-producing state, followed by Oregon and then Idaho. New production regions for hop are found in Michigan, Minnesota, New York, and to a lesser extent, other states in the northeastern United States. Agronomic issues faced by producers are low yields of European landraces and US cultivars developed before the 21st century, control of fungal pathogens such as downy mildew [caused by Pseudoperonospora humuli (Miyabe & Takah.) G.W. Wilson] and powdery mildew [caused by Podosphaera macularis (Wallr.) U. Braun & Takam.], and damson hop aphid (Phorodon humuli Scrank) and two-spotted spider mite (Tetranychus urticae Koch).

Two primary hop cultivar types exist, based on end use. The traditional “aroma-type” hop cultivar has lower levels of the primary bittering component called alpha acids—a class of water-soluble chemicals that produce the bittering of beer during the fermentation stage of brewing. The other primary class of hop cultivars are those produced for their high concentration of alpha acids present in the resin of lupulin glands. The alpha acids in these cultivars are extracted by supercritical fluid extraction using liquid CO₂ (Gardner 1993) and are sold under the singular nomenclature “alpha-extract.”

The units for reporting alpha acids differ between male and female accessions based solely on the preponderance of lupulin glands found in female flowers in comparison to male flowers. Alpha acid content in female accessions are reported as the weight of alpha acid per weight of dried hop cone tissue, with...
values ranging from 2.0 to 18.0 (w/w). Alpha acid content in male accessions are reported as the volume of alpha acid per total volume of bittering acids (v/v). Alpha acid values for male hop accessions range from 20.0 to 65.0 (v/v).

Several cultivars are classified as “super-alpha” cultivars (average alpha acids greater than 15% v/v total bittering acids), with almost all being privately held, patented cultivars. The only publicly available super-alpha hop cultivar is ‘Nugget’ (Haunold et al., 1984). Three other super-alpha hop cultivars are proprietary, nonpatented lines (‘Columbus’, John I Haas, Inc.; ‘Tomahawk’, Select Botanicals, Inc.; and ‘Zeus’, Hopsteiner, Inc.), while the remaining are patented proprietary cultivars. The USDA–ARS released USDA 21742M germplasm line in 2009 as a breeding material for generating high alpha levels averaging above 18% (v/v alpha acid/total bittering acids). Unfortunately, this line exhibited low yields and was thus unsatisfactory for production purposes. It remains that high-yielding public super-alpha hop cultivars are needed by the US hop industry, particularly so after reports that Nugget was susceptible to all races of powdery and downy mildew in the Pacific Northwest (Wolfenbarger et al., 2014). While several female hop accessions are available for germplasm development of high alpha hop cultivars, only a few male lines currently fulfill this breeding niche. Hop breeding would greatly benefit from having access to male germplasm that can be used to produce offspring with the potential for high yields and high alpha-acid levels.

Methods

USDA 21267M (Reg. No. GP-37, PI 559093) originated from a cross performed by USDA–ARS personnel at Corvalis, OR, in 1973 between ‘Comet’ (Zimmermann et al., 1975) and USDA 21110M. USDA 21110M is a nonpatented, proprietary experimental male hop line within the USDA–ARS hop breeding program that resulted from a cross between ‘Bullion’ and ‘64035M’ (Haunold et al., 1985). The pedigree for USDA 21267M is Comet/Bullion/64035M. USDA 64035M is a nonpatented, publicly released male line possessing resistance to downy mildew derived from a cross between Hallertauer Mittelfruh and an unknown male accession. Thus, the genetic composition of USDA 21267M is 1/2 Comet, 1/8th Bullion, 1/8th Hallertauer Mittelfruh, and 1/8th unknown. This germplasm produces abundant pollen, is vigorous, and matures early in comparison to other male germplasm. Pollen is generally available at the beginning of July in Oregon.

USDA 21267M was first grown at the USDA–ARS hop research facility near Corvalis in 1974 as offspring 7303-138M and was given the USDA accession number 21267M in 1980. It was evaluated for phenotypic characteristics for several years before being saved as a male germplasm. Determination of alpha and beta acids, along with hop storage index (HSI; Nickerson and Likens, 1979), was performed using American Society of Brewing Chemists (1969, 1992) standard methods. Mating studies for heritability of six traits (yield, alpha and beta acid levels, cohumulone and colupulone content, and xanthohumol content) were performed in 1999 and 2000 under replicated field design and with methods reported by Henning and Townsend (2005). Downy mildew ratings were collected on naturally inoculated field-grown hop plants (M.S. Townsend, unpublished data, 2009). Powdery mildew ratings were obtained by inoculation and subsequent scoring of leaf infection under greenhouse conditions (D.H. Gent, unpublished data, 2016) using inoculation techniques as reported by Henning et al. (2011)

Characteristics

Chemical analyses of USDA 21267M lupulin glands indicate its primary use as a breeding parent for bittering-type hop cultivars where higher levels of alpha acids in offspring are desired. Phenotypic measurements of USDA 21267M showed alpha acids averaged 53.2% (v/v), while beta acids averaged 18.1% (v/v), with alpha acids comprising 74% of the total bittering acids (v/v). This germplasm also exhibited low cohumulone levels of 28% and has good storage capabilities with a HSI of 0.259.

Genetic tests of USDA 21267M (Henning and Townsend, 2005) among five males mated to five unrelated hop cultivars for five traits showed this germplasm as producing offspring with the highest average values for yield, alpha acid levels, beta acid levels, colupulone levels, and xanthohumol levels (Table 1). There were no statistical differences between USDA 21267M and USDA 21262M for offspring yield. However, USDA 21267M was statistically higher in alpha acid levels than all other male lines. This line also produced female offspring with high colupulone levels that were equal to offspring derived from USDA 21262M. Finally, offspring from USDA 21267M, along with those from USDA 21266M (full-sib brother), produced offspring with the highest level of xanthohumol.

Field-based observations of USDA 21267M showed it having susceptibility to downy mildew infection similar to the cultivars ‘Crystal’ (Haunold et al., 1995), Wye ‘Target’, and Comet. Under normal growing conditions, early downy mildew infections should be controlled with a combination of pruning and fungicides registered for use on hop. No research on the genetic transfer of susceptibility to downy mildew has been reported, and the breeding value for this trait is unknown.

No published experiments have been written on resistance levels of USDA 21267M to powdery mildew strains found in the Pacific Northwest. Greenhouse observations of inoculated plants (D.H. Gent, unpublished data, 2016) showed it as being susceptible to powdery mildew races found in the US Pacific Northwest. As a result, regular prophylactic spraying for this disease as well as downy mildew is recommended. No cases of Verticillium wilt (caused by Verticillium albo-atrum Reinke & Berthier) were observed in nursery plots. Growers are cautioned against growing USDA 21267M in known Verticillium-infested fields due to the potential susceptibility of this line based on its parentage. No published studies have been performed on USDA 21267M susceptibility to hop aphids, nor is there any information on its susceptibility to spider mite infestation, although no serious problems have been noted in nursery plots located in Oregon.

Discussion

Hop breeders from private breeding companies and new state breeding programs have requested access to USDA 21267M for development of high-yielding, high alpha-acid hop cultivars. The lack of publicly available male lines for developing
germplasm or cultivars with high yield and high alpha-acid levels suggests the need for such lines in burgeoning hop breeding programs across the United States. While disease resistance levels for USDA 21267M are not as good as other available male germplasm, these other male lines do not have the genetic potential for developing higher-yielding, higher alpha-acid levels. It is recommended that use of this breeding line be coupled with that of female lines possessing good resistance to both downy and powdery mildews.

USDA 21267M also showed high general combining ability for colupulone and xanthohumol. Colupulone is one of the components that make up beta acids found in hop bittering resins. As such, it is not readily soluble in water but slowly breaks down to form other non-bittering flavor compounds during boiling steps in the presence of oxygen (Bett et al., 1981). High levels of this compound are considered desirable by brewers. Xanthohumol is another component of the hop resins that is also nonsoluble in water. Numerous human health benefits have been attributed to the use of purified xanthohumol, including acting as an anti-oxidant, anti-microbial, anti-inflammatory, anti-cancer agent (Gerhäuser, 2005; Legette et al., 2013; Lupinacci et al., 2009; Miranda et al., 2013; Nozawa, 2005; Peluso et al., 1996; Xuan et al., 2010), as well as showing potential for therapeutic treatment of metabolic diseases including type 2 diabetes (Legette et al., 2014). Given that USDA 21267M has high general combining ability for colupulone and xanthohumol, its use in breeding for higher levels of these two compounds would be advantageous over other male lines.

**Conclusion**

The release of USDA 21267M provides a new male hop germplasm exhibiting high breeding value for yield, high alpha-acid levels, and high colupulone and xanthohumol levels in female offspring. Its use in hop breeding programs may result in the development of high-yielding, high alpha-acid superior extract type hop cultivars. USDA 21267M is released to the public at the request of multiple breeding programs.

**Availability**

Genetic material of USDA 21267M has been deposited in the National Germplasm System at the USDA–ARS National Clonal Germplasm Repository located near Corvallis, OR. This material will be available for research purposes, including the development and commercialization of new cultivars. It is requested that appropriate recognition be given if this germplasm contributes to the development of a new breeding line or cultivar. Requests should be made to Dr. Kim Hummer, Curator, USDA–ARS National Clonal Germplasm Repository, Corvallis, OR 97331 (http://www.ars.usda.gov/Main/docs.htm?docid=11308).

**References**


**Table 1. Maternal and paternal half-sib family averages for yield, alpha-acid concentration (ALP), beta-acid concentration (BET), cohumulone levels (COH), colupulone levels (COL), and xanthohumol concentration (XAN).†**

<table>
<thead>
<tr>
<th>Family</th>
<th>Yield</th>
<th>ALP</th>
<th>BET</th>
<th>COH</th>
<th>COL</th>
<th>XAN</th>
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<tr>
<td></td>
<td>kg h⁻¹</td>
<td>w/w</td>
<td>v/v</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Maternal family</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Challenger</td>
<td>668.4 b‡</td>
<td>4.85 c</td>
<td>1.71 a</td>
<td>30.6 a</td>
<td>51.9 a</td>
<td>0.18 a</td>
</tr>
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<td>4.63 c</td>
<td>1.73 a</td>
<td>31.5 a</td>
<td>53.5 a</td>
<td>0.18 a</td>
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<td>5.46 b</td>
<td>1.58 a</td>
<td>30.5 a</td>
<td>53.2 a</td>
<td>0.16 a</td>
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<td>Magnum</td>
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<td>6.45 a</td>
<td>2.13 a</td>
<td>30.2 a</td>
<td>51.9 a</td>
<td>0.22 a</td>
</tr>
<tr>
<td>Orion</td>
<td>556.3 c</td>
<td>5.80 b</td>
<td>1.81 a</td>
<td>29.8 a</td>
<td>52.9 a</td>
<td>0.20 a</td>
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<td>21262M</td>
<td>697.4 ab</td>
<td>5.74 b</td>
<td>1.99 a</td>
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<td>52.5 b</td>
<td>0.16 b</td>
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</table>

† Means within a column and subheading of male or female that have similar letters are not significantly different from one another based on Fisher’s LSD test.

‡ From Henning and Townsend (2005).


