Genetic Analyses of Epicuticular Waxes Associated with the Glossy Foliage of ‘White Persian’ Onion

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ABSTRACT. Onion (*Allium cepa*) plants with lower amounts of epicuticular waxes on foliage suffer less damage from the insect pest *Thrips tabaci* (onion thrips). Glossy onion accumulates significantly less epicuticular wax compared with wild-type “waxy” onion, and a single recessive locus (*gl*) has been proposed to condition this phenotype. Genetic analyses of types and amounts of epicuticular waxes were completed using two segregating families from the cross of the glossy inbreds B9885 and B9897 (both originally selected from the onion cultivar White Persian) with waxy inbred B8667 and semiglossy (intermediate amounts of waxes) inbred B5351, respectively. F₂ progenies were grown in greenhouses and scored visually for foliar phenotypes, and amounts and types of epicuticular waxes were determined using gas chromatography-mass spectrometry (GCMS). For one F₂ family from the cross of glossy B9885 by waxy B8667, visually scored glossy vs. waxy foliage fit a 1:3 ratio and the phenotype mapped to chromosome 8 of onion. This same region on chromosome 8 was significantly associated with amounts of the ketone hentriacontanone-16 (H16) and fatty alcohols 1-octacosanol (Oct1) and 1-triacontanol (Tri1). Visually scored F₂ progeny from the cross of glossy B9897 × semiglossy B5351 did not fit expected models for one or two recessive loci. Significant quantitative trait loci (QTL) were revealed on chromosomes 5 and 8 controlling amounts of H16. Epistasis was detected between regions on chromosomes 1 and 8, and a 100-fold increase of H16 was conditioned by homozygous genotypes for the B5351 region on chromosome 1 and the B9885 region on chromosome 8. The three QTL model explained 41% of the phenotypic variation for amounts of H16 at logarithm of odds of 16.6. Amounts of Oct1 and Tri1 in the B9897 × B5351 family were associated with a major QTL on chromosome 1, explaining 37% to 46% of the phenotypic variation, respectively. This research demonstrates that glossy foliage of ‘White Persian’ onion is conditioned by a recessive locus on chromosome 8 for which we propose the name *gl^{wp}*. These results are important for selection of onion with unique profiles of epicuticular waxes to reduce losses resulting from onion thrips.

Waxes accumulate on the surface of plant leaves and are important for avoidance of abiotic stresses such as aerial desiccation (Cameron et al., 2006; Kim et al., 2007), water deprivation (Aharoni et al., 2004; Bourdenx et al., 2011), and damaging wavelengths of light (Holmes and Keiller, 2002). Wax composition varies among plants (Bernard and Joubès, 2013; Jenks et al., 1995), and in arabidopsis (*Arabidopsis thaliana*), the predominant waxes are C29 alkanes, ketones, and secondary alcohols (Lee and Suh, 2013). In arabidopsis, the *eceriferum* (*cer*) mutants have brighter green leaves compared with wild-type plants and have been used to study wax biosynthesis. Production of leaf waxes starts in the leucoplast with synthesis of C16 to C18 fatty acyl-coenzymes A (CoAs) by long-chain acyl-CoA synthetase 1 (LACS1) and 2 (LACS2) enzymes (Lü et al., 2009), follow by carbon elongation by the

fatty acid elongase (FAE) complex to produce very long-chain (>C18) fatty acyl-CoAs (VLFA-CoAs) (Yeats and Rose, 2013). The FAE complex in arabidopsis consists of four enzymes that extend fatty acids by two carbons per cycle: β -keto-acyl-CoA synthase (KCS), β -ketoacyl-CoA reductase, 3-hydroxyacyl-CoA dehydratase, and trans2,3-enoyl-reductase (Fiebig et al., 2000; Haslam et al., 2012, 2015). The KCS genes have shown substrate specificity for acyl-CoAs of different carbon chain length (Haslam and Kunst, 2013; Samuels et al., 2008). Hydrolysis of VLFA-CoAs by a thioesterase generates very long-chain fatty acids that can enter two pathways: the acyl-reduction pathway, forming primary alcohols (Rowland et al., 2006) and wax esters (Li et al., 2008); and the decarbonylation pathway, which forms secondary alcohols, aldehydes, alkanes, and ketones (Samuels et al., 2008).

Wax coverage and composition are important for biotic interactions providing stimuli that attract or repel insects (Eigenbrode and Espelie, 1995). The glossy phenotype of onion has light-green leaf color, accumulates significantly less epicuticular waxes on its foliage compared with wild-type “waxy” onion, and suffers less damage by onion thrips (Damon et al., 2014; Jones et al., 1934; Molenaar, 1984; Munaiz et al., 2019). The main epicuticular wax on the leaves of wild-type onion and leek (*Allium ampeloprasum*) is the ketone H16, followed by lower amounts of fatty alcohols and alkanes (Damon et al., 2014; Rhee et al., 1998). Scanning electron microscopy of epicuticular waxes on onion leaves revealed crystals associated with the

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phenotypic appearance of leaves (Damon et al., 2014; Molenaar, 1984; Yang et al., 2017). Specific wax constituents may form crystals on the leaf surface, and different components produce unique shapes (Gülz et al., 1992; Jenks et al., 1995). Visual crystals are produced on the surface of waxy onion leaves when the amount of H16 represents more than two thirds the total wax load, and they are responsible for the blue-green foliar color of waxy onion (Damon et al., 2014; Molenaar, 1984).

Jones et al. (1944) crossed glossy and waxy plants from the onion cultivar Australian Brown and completed genetic studies revealing that a single locus (*gl*) controlled glossy foliage. Crosses involving glossy plants from the onion cultivar White Persian (Jones et al., 1934) did not fit a single recessive model because of too few glossy segregants (Jones et al., 1944). Molenaar (1984) studied the glossy phenotype from 'White Persian' and proposed that it was conditioned by a single recessive locus, which he assumed to be *gl*. However, Jones and Mann (1963, p. 81) reported that crosses between glossy plants from 'Australian Brown' and 'White Persian' produced waxy hybrids, indicating that different loci condition the glossy phenotype in these two populations. In bunching onion (*A. fistulosum*), the glossy phenotype is conditioned by a single recessive locus (Yang et al., 2017). The goal of this study was to determine the genetic basis of the glossy phenotype from 'White Persian', which has been shown to suffer less damage by onion thrips in numerous studies (Damon et al., 2014; Jones et al., 1934; Molenaar, 1984; Munaiz et al., 2019).

Materials and Methods

PLANT MATERIALS. Glossy inbreds B9885 [U.S. Department of Agriculture (USDA) plant introduction (PI) 546303] and B9897 (PI 546305) of onion were developed by backcrossing glossy foliage from 'White Persian' (Jones et al., 1934) to long-day storage germplasm with yellow bulb color and were released in 1983 by the USDA and the experiment stations of Iowa, Michigan, and Wisconsin (Goldman et al., 2001; Molenaar, 1984). Inbred B8667 has waxy foliage and red bulbs (Havey and Bohanec, 2007); inbred B5351 has semiglossy foliage (Damon et al., 2014). Seeds of these inbreds were planted in field plots at the Dean Kincaid Farm (Palmyra, WI) under normal production conditions and were scored visually to confirm foliar phenotypes. Bulbs were harvested at maturity, stored for 6 months at 7 °C, and planted in a greenhouse in a soilless medium (Metro-Mix; Sun Gro Horticulture, Agawam, MA). Plants were grown under 14-h days and temperatures of 27 °C days and 22 °C nights, and fertilized every week with 20N–8.7P–16.6K (Peters Professional, Allentown, PA). Leaves were sampled for analyses of epicuticular waxes by GCMS as described later.

Glossy B9885 was crossed as the female parent with B8667, and hybrids identified by waxy foliage and red bulbs. Individual hybrid plants were self-pollinated to produce segregating F₂ families. Seeds from F₂ families were planted in field plots as described earlier and were scored visually for glossy vs. waxy foliage. Bulbs from one F₂ family were harvested at maturity, stored for 90 d at 7 °C, planted, and grown in a greenhouse as described previously. A second family was developed by crossing glossy B9897 as the female with semiglossy B5351. Hybrids were identified by waxy foliage and self-pollinated. Seeds from the parental inbreds and one F₂ family were planted in a greenhouse as described previously, and the plants were grown for 8 weeks as described earlier. For both families, foliage was scored visually as waxy vs. nonwaxy and sampled using GCMS as described later.

DNA EXTRACTIONS AND GENOTYPING OF SINGLE NUCLEOTIDE POLYMORPHISMS (SNPs). Leaf tissue was collected from at least five plants of the parental inbreds and each segregating progeny, frozen in liquid nitrogen, lyophilized, and stored at –20 °C. Samples were ground by adding metal spheres and shaking for 45 s until the leaf tissue was pulverized. Genomic DNA was extracted (NucleoSpin Plant II Midi Kit; Macherey-Nagel, Düren, Germany), and purity and concentration were evaluated spectrophotometrically (NanoDrop; Thermo Fisher Scientific, Waltham, MA).

DNAs from 93 F₂ progenies of the cross B9885 × B8667 were genotyped using the 1692 SNP array (Havey and Ghavami, 2018). Goodness-of-fits to the expected 1:2:1 segregation for codominant markers were tested using chi-square analyses, and the linkage map was created using JoinMap 4.0 (Van Ooijen, 2006) with regression mapping and the Kosambi function. DNAs of 98 F₂ plants from the B9897 × B5351 cross were genotyped for 232 SNPs (Duangjit et al., 2013) using the KASPar assay (LGC Genomics, Boston, MA). Genotypic scores were verified using SNP viewer 2 version 4.0.0 software (LGC Genomics). Segregation analysis and construction of the linkage map were completed as described earlier. Linkage groups from both maps were assigned to chromosomes using common SNPs that segregated in the mapping populations OH1 × 5225, BYG15-23 × AC43, or DehyA × B5351 (Damon and Havey, 2014; Duangjit et al., 2013).

SAMPLING FOR GCMS ANALYSES OF EPICUTICULAR WAXES. Two samples from one fully extended leaf were collected from five plants of the parental inbreds and from each F₂ progeny from the B9885 × B8667 family at 45 d after planting of bulbs in a greenhouse. For the B9897 × B5351 family, two samples from one fully extended leaf from each F₂ progeny were collected 8 weeks after planting of seed in the greenhouse. For all samplings, the chosen leaf was fully extended and equidistant between the oldest and youngest leaves. Individual leaf samples were weighed, and docosane (Sigma-Aldrich, St. Louis, MO) dissolved in high-performance liquid chromatography (HPLC) reagent-grade chloroform (Fisher Scientific, Hampton, NH) was added to leaf pieces at a rate of 100 µg·g^{–1} fresh weight. Each leaf segment was then submerged in HPLC-grade chloroform (Fisher Scientific) for 1 minute and discarded. The chloroform was allowed to dry in a fume hood for ≈5 d. In preparation for GCMS, wax extracts were dissolved in 500 µL HPLC-grade chloroform, 600 µL acetonitrile (Fisher Scientific), and 210 µL HPLC/GC-grade N,O bis(trimethylsilyl)trifluoro-acetamide (Sigma-Aldrich), and incubated for 30 min at 80 °C. Identification and quantification of waxes used GCMS conditions as described previously (Damon et al., 2014).

STATISTICAL ANALYSES. Peak areas of individual waxes were adjusted using areas of the docosane peaks in each run and therefore represent amounts of individual waxes on a gram fresh weight basis. Adjusted peak areas from the two samples from the same leaf were averaged and used for all analyses. Analyses of variance and Spearman correlations for amounts of individual waxes were calculated using RStudio (R Foundation for Statistical Computing, Vienna, Austria). Visually scored foliar phenotypes and amounts of individual waxes were analyzed using a binary model and composite interval mapping (CIM), respectively, with the R/qtl package (Broman and Sen, 2009; Broman et al., 2003). Haley Knott regression with 10 marker covariates within a window size of 10 cM was used to determine significant associations (Haley and Knott, 1992), and a 95% significance

Table 1. Foliar phenotypes and mean $\pm$ SD for peak areas adjusted to the internal docosane standard as detected by gas chromatography for the three most prevalent epicuticular waxes on the foliage of onion inbreds used to develop segregating families.

Inbred	Visual phenotype	Hentriacontanone-16	Octacosanol-1	Triacontanol-1
		(mean $\pm$ SD)		
B9897	Glossy	0.025 $\pm$ 0.017 a ^z	0.247 $\pm$ 0.066 a	0.129 $\pm$ 0.030 a
B9885	Glossy	0.166 $\pm$ 0.056 a	0.400 $\pm$ 0.202 a	0.183 $\pm$ 0.066 a
B5351	Semiglossy	0.871 $\pm$ 0.313 b	0.274 $\pm$ 0.200 a	0.173 $\pm$ 0.072 a
B8667	Waxy	1.925 $\pm$ 0.719 c	0.363 $\pm$ 0.222 a	0.230 $\pm$ 0.117 a

^zMeans followed by the same letter are not significantly different using Tukey's honestly significant difference at $P < 0.05$.

Fig. 1. Waxy (left) vs. glossy (right) foliage on F₂ progenies from the cross of B9885 $\times$ B8667 of onion.

logarithm of odds (LOD) threshold was used after 1000 permutations. Multiple QTL were detected using the stepwiseqtl function, dropping one marker at a time, with parameters of 0.05 significance level and 1.5-LOD score. Effects of the candidate QTL were refined using makeqtl, refineqtl, and fitqtl, and the estimated effects provided the percentage of the phenotypic variation explained by the QTL. Epistatic interactions among loci were evaluated using scanonnetwo, sim.geno, and the penalized LOD score.

Results

EPICUTICULAR WAXES ON FOLIAGE OF PARENTAL INBREDS. GCMS analyses of the parental inbreds revealed that the three most predominant waxes were the ketone H16 and fatty alcohols Oct1 and Tri1 (Table 1), which is in agreement with Damon et al., (2014). Glossy inbreds B9885 and B9867 had significantly lower amounts of H16 than semiglossy B5351 and waxy B8667, and B5351 had significantly less H16 than waxy B8667 (Table 1). Amounts of Oct1 and Tri1 were not

significantly different among these inbreds (Table 1). The Spearman correlation between the two fatty alcohols (Oct1 and Tri1) was highly significant at 0.92 ($P < 0.001$), and correlations between amounts of H16 and the two fatty alcohols (Oct1 and Tri1) were also highly significant ($P < 0.001$) at 0.77 and 0.78, respectively.

GENETIC ANALYSIS OF THE GLOSSY $\times$ WAXY FAMILY. The cross of glossy B9885 by waxy B8667 produced hybrids with waxy foliage, and self-pollination of six hybrids yielded F₂ families segregating for foliar glossiness. Visual scoring of glossy vs. waxy foliage (Fig. 1) yielded 20 glossy and 191 waxy F₂ progenies for segregations combined across families after confirming homogeneity of errors, which did not fit the 1:3 ratio for a single recessive locus ($P < 0.001$). Jones et al. (1944) also reported too few glossy progenies in F₂ families from crosses of glossy plants from 'White Persian' with waxy plants; however, Molenaar (1984) reported acceptable fits to a single recessive locus in crosses between B9885 and waxy inbred MSU611. Segregations for foliar glossiness in one F₂ family from B9885 by B8667 fit the 1:3 ratio ($P = 0.355$), and genotyping revealed 499 SNPs segregating in this family. One SNP was discarded because there were no maternal homozygotes and segregations did not fit the expected 1:3 ratio for a dominant locus ($P < 0.001$). All the remaining 498 SNPs showed goodness-of-fits at $P > 0.002$ and were used for genetic mapping. A total of 490 SNPs segregated into eight linkage groups at LOD 4.0, with a total map length of 8.98 Morgans (Supplemental Table 1). Eight SNPs remained unlinked and were not used for quantitative analyses.

Mapping of glossy vs. waxy foliage as a binary trait in the F₂ family from B9885 by B8667 revealed a major QTL on chromosome 8 at 46.2 cM explaining 65% of the phenotypic variation (Table 2). A significant QTL was revealed by CIM controlling amounts of H16 on chromosome 8 at 41 cM (LOD 15.8) and explained 46% of the phenotypic variation (Table 2). The chromosome region from the waxy parent B8667 had the additive effect of increasing adjusted peak areas for H16 by 0.57, with a dominance effect of 0.33. The most significant SNP on chromosome 8 associated with amounts of H16 was isotig19082_1721, which mapped very closely to visually scored glossy vs. waxy phenotypes (Table 2). Although amounts of Oct1 and Tri1 were not significantly different between B9885 and B8667 (Table 1), CIM revealed a QTL on chromosome 8 at 46.2 cM (LODs 3.7 and 5.5 for Oct1 and Tri1, respectively) associated with amounts of these fatty alcohols and mapping to the same location on chromosome 8 affecting amounts of H16 (Table 2). The QTL explained 17% of phenotypic variation for amounts of Oct1 (Table 3), and the chromosome region from B8667 showed an additive effect of 0.36 and a dominance effect of 0.31 (Table 2). For Tri1, this same QTL explained 24% of the phenotypic variation, with the region from B8667 increasing amounts of Tri1 with an additive effect of 0.30 and a dominance effect of 0.19 (Table 2). These results demonstrate that a region on chromosome 8 from B9885 is associated with the visually glossy phenotype and reduced amounts of the ketone H16 and fatty alcohols Oct1 and Tri1.

GENETIC ANALYSES OF THE GLOSSY $\times$ SEMIGLOSSY FAMILY. The cross of glossy B9897 by semiglossy B5351 produced waxy

Table 2. Chromosome (Chr) and position (Pos) of the most significant single-nucleotide polymorphism (SNP), SNPs flanking the 1.5 logarithm of odds (LOD) confidence interval, percent variation (Var) explained, LOD with threshold (Thresh) values from permutation analysis, and allelic effects for quantitative trait loci detected by composite interval mapping for visually glossy vs. waxy phenotypes and amounts of hentriacontanone-16 (H16), octacosanol-1 (Oct1), and triacontanol-1 (Tri1) on foliage of F₂ progenies from the B9885 × B8667 onion family.

Trait	Chr	Pos (cM)	SNP ^z	1.5 LOD interval ^z	Var (%)	LOD	Thresh	Add ^y	Dom ^y
Visual phenotype	8	46.2	i20235_630	i41653_558–i28633_2705	65.4	23.8	9.2	0.41	0.40
H16	8	41.1	i19082_1721	i41653_558–i20235_630	45.6	15.8	9.3	0.57	0.33
Oct1	8	46.2	i20235_630	i19082_1721–i28633_2705	16.9	3.7	3.5	0.36	0.31
Tri1	8	46.2	i20235_630	i19082_1721–i28633_2705	23.6	5.5	3.6	0.30	0.19

^zi = isotig as described by Duangjit et al. (2013). Map positions of SNPs are listed in Supplemental Table 1.

^ySignificant additive (Add) and dominance (Dom) effects of marker alleles in which a positive value indicates that allele from waxy parent B8667 increased amounts.

Table 3. Chromosome (Chr) and position (Pos) of the most significant single nucleotide polymorphism (SNP), SNPs flanking the 1.5 logarithm of odds (LOD) confidence interval, percent of phenotypic variation (Var) explained, LOD with threshold (Thresh) values from permutation analysis, and allelic effects for quantitative trait loci significantly affecting amounts of hentriacontanone-16 (H16), octacosanol-1 (Oct1), and triacontanol-1 (Tri1) on foliage of F₂ progenies from the B9897 × B5351 family of onion.

Trait	Chr	Pos (cM)	SNP ^z	1.5 LOD interval ^z	Var (%)	LOD	Thresh	Add ^y	Dom ^y
H16	1	41	i31382_1702	i17679_1985–c00172_269	9.0	6.0	7.4	0.11	–0.20
H16	5	38	i31940_1090	i36154_963–i26178_547	11.4	7.6	7.4	–0.20	0.02
H16	8	53	i20266_1040	i37010_277–i38873_388	21.0	13.3	7.4	0.13	0.01
Oct1	1	0	i37443_625	i37443_625–i32558_1278	37.1	9.7	7.6	–0.14	–0.09
Tri1	1	0	i37443_625	i37443_625–i32558_1278	46.4	13.7	7.4	–0.08	–0.03

^zi = isotig and c = contig as described map positions of SNPs are listed in Supplemental Table 2.

^ySignificant additive (Add) and dominance (Dom) effects of marker alleles in which a positive value indicates that allele from waxy parent B5351 increased amounts.

hybrids that were self-pollinated. Visually scored F₂ progenies into two phenotypic classes (9 glossy vs. 81 nonglossy) did not fit models for one or two recessive loci ($P < 0.001$) as a result of a paucity of glossy plants. This observation is consistent with F₂ segregations reported by Jones et al. (1944) and from our B9885-by-B8667 families, in which too few glossy plants appeared. Genotyping of F₂ progenies from B9897 × B5351 resulted in 142 segregating SNPs, and mapping yielded eight linkage groups at LOD 8.0 with a total map length of 10.4 Morgans (Supplemental Table 2).

Significant QTL were revealed by CIM controlling amounts of H16 on chromosome 8 at 53 cM (LOD 13.3) and chromosome 5 at 38 cM (LOD 7.6), explaining 21% and 11% of the phenotypic variation, respectively (Table 3). The two QTL model accounted for 38% of the phenotypic variation at LOD 18.1. The region on chromosome 5 from B5351 showed an additive effect reducing H16 amounts, in agreement with Damon and Havey (2014). The region on chromosome 8 from the semiglossy parent (B5351) showed an additive effect increasing the amount of H16 (Table 3). Interactions between the QTL on chromosomes 5 and 8 were not significant. Amounts of H16 were affected by an epistatic interaction between regions on chromosomes 1 and 8 (Fig. 2). Significantly greater amounts of H16 were conditioned by homozygous regions on chromosome 1 from semiglossy B5351 and on chromosome 8 from glossy B9885. Chromosome 1 had no effect on amounts of H16 when the region from B9885 was present, regardless of the genotype in chromosome 8 (Fig. 2). There was no interaction between the QTL on chromosomes 1 and 5 ($P = 0.773$). The three QTL model (chromosomes 1, 5, and 8) for amounts of H16 explained 54% of the phenotypic variation at LOD 16.6.

A region on chromosome 1 at 0 cM was detected by CIM affecting amounts of Oct1 and Tri1, explaining 37% of the phenotypic variation at LOD 9.7 for Oct1 and 46% of the phenotypic variation at LOD 13.7 for Tri1 (Table 3). The region from semiglossy B5351 reduced amounts of Oct1 and Tri1. No significant QTL affecting amounts of Oct1 or Tri1 were detected on chromosome 8 as observed in the B9885-by-B8667 family.

Discussion

We studied the genetic basis of the glossy phenotype of onion using two inbred lines (B9885 and B9897) derived from ‘White Persian’ onion (Goldman et al., 2001; Molenaar, 1984). Although both B9885 and B9897 have glossy foliage, there were differences for major QTL affecting amounts of epicuticular waxes. In the segregating family from B9885 crossed with B8667, a region on chromosome 8 was significantly associated with the visual glossiness of the foliage and amounts of the ketone H16 and fatty alcohols Oct1 and Tri1 (Table 2). In the B9897 × B5351 family, the same region on chromosome 8 significantly affected amounts of H16, but not Oct1 and Tri1 (Table 3). The region on chromosome 8 in the B9897 × B5351 family interacted with one on chromosome 1 to increase the numbers of waxy progenies (Fig. 2). This interaction likely contributed to observations by Jones et al. (1944) and our research of too few glossy progenies in F₂ families segregating for the glossy phenotype from ‘White Persian’. The region on chromosome 1 from glossy B9897 may act as a dominant repressor of H16 accumulation. In the segregating family from B9897 × B5351, we identified a significant QTL on chromosome 5 affecting amounts of H16 (Table 2), which is in

Fig. 2. Epistatic interaction between genotypes on chromosome 1 (isotig31382_1702) vs. chromosome 8 (isotig20266_1040) affecting amounts of hentriacontanone-16 in the F_2 family from B9897 $\times$ B5351 of onion. A indicates allele from the female parent (B9897) and B is the allele from the male parent (B5351).

agreement with the work by Damon and Havey (2014), who detected this same region in the cross of waxy DehyA $\times$ B5351. The QTL on chromosome 5 showed no significant interaction with chromosome 1 for amounts of H16.

In the glossy B9885-by-waxy B8667 family, the region on chromosome 8 was significantly associated with amounts of Oct1 and Tri1 (Table 2). However, in the family from glossy B9897 by semiglossy B5351, amounts of Oct1 and Tri1 were significantly associated with a region on chromosome 1 (Table 3) and no effect was detected on chromosome 8. In previous research (Damon and Havey, 2014), we detected a region on chromosome 2 from semiglossy B5351 reducing amounts of Oct1 and Tri1. Because chromosomes 2 and 8 had no significant effect on amounts of Oct1 and Tri1 in the B9897 $\times$ B5351 family, it may be there was no variation between B9897 and B5351 for these QTL.

Jones et al. (1944) studied the genetics of the glossy phenotype from onion cultivars Australian Brown and White Persian. A glossy plant from 'Australian Brown' was self-pollinated for two generations and progenies were crossed with waxy plants from 'Australian Brown' to produce an F_2 family. Visually scored foliar phenotypes indicated that a single locus (*gl*) conditioned glossy foliage (Jones et al., 1944). Segregating families from crosses of glossy plants from 'White Persian' with waxy plants resulted in a paucity of glossy progenies. Jones and Mann (1963) reported that crosses between glossy plants from 'White Persian' and 'Australian Brown' produced waxy hybrids, and therefore different loci must condition the

glossy phenotype in these two populations. In our current research, genetic mapping of the glossy phenotype from 'White Persian' revealed a region on chromosome 8 associated with the visual foliar phenotype and amounts of H16. Because this region was detected in two segregating families, we propose the name *gl^{wp}* for a recessive locus on chromosome 8 from 'White Persian' (wp) that reduces the accumulation of epicuticular waxes on onion foliage.

Our research demonstrates that major QTL on chromosomes 5 and 8 affect amounts of H16, the main epicuticular wax on onion foliage (Tables 2 and 3) (Damon and Havey, 2014). Amounts of the two most predominant fatty alcohols (Oct1 and Tri1) are highly correlated and controlled by QTL on chromosomes 1 (Table 3), 2 (Damon and Havey, 2014), and 8 (Table 2). Lower amounts of H16 relative to the other epicuticular waxes are significantly associated with reduced damage by onion thrips (Damon et al., 2014; Munaiz et al., 2019). The glossy phenotype of onion has been reported as susceptible to powdery mildew (Mohan and Molenaar, 2005), which limits its usefulness for commercial production. However, selection of onion for lower amounts of H16 conditioned by QTL chromosomes 5 or 8 and greater amounts of the other wax components, such as fatty alcohols on chromosomes 1 and 2, may produce onion populations that suffer less damage by onion thrips and accumulate enough total epicuticular wax to be commercially acceptable.

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Supplemental Table 1. Observed segregations, goodness-of-fit probabilities (P), and chromosome (Chrom) and centiMorgan (cM) positions for single nucleotide polymorphisms (SNPs) from the F_2 family from B9885 $\times$ B8667 of onion.

Locus ^z	Chrom	cM	Observed			P value
			B9885	Het	B8667	
i30179_1191	1	0.0	28	46	19	0.416
i36152_724	1	7.8	25	49	19	0.594
i32522_1235*	1	10.0	23	53	17	0.274
i30361_1487*	1	17.8	20	54	19	0.295
i33035_121*	1	18.4	20	53	19	0.341
i14997_1521	1	29.1	23	49	21	0.837
i33254_1237	1	29.1	23	49	21	0.837
i33254_614	1	29.1	23	49	21	0.837
i33340_203	1	31.9	22	48	23	0.943
i33340_1003*	1	31.9	22	48	23	0.943
i33340_887	1	31.9	22	48	23	0.943
i35034_431	1	34.1	23	46	24	0.984
i31711_966	1	35.7	23	45	25	0.913
i39994_602*	1	36.8	23	45	25	0.913
i32264_1259	1	38.4	23	46	24	0.984
i30097_102	1	42.4	23	47	23	0.995
i30097_1638	1	42.4	23	45	18	0.681
i27517_440*	1	45.1	24	50	19	0.587
i42358_331	1	45.1	24	50	19	0.587
i21900_461*	1	48.5	26	46	21	0.760
i29455_1102*	1	49.0	26	47	20	0.675
i29455_1536	1	49.0	26	47	20	0.675
i29462_1710*	1	49.0	26	47	20	0.675
i34700_979	1	49.0	26	47	20	0.675
i32775_1170*	1	49.0	26	47	20	0.675
i12342_956	1	49.5	25	48	20	0.728
i31577_1144	1	52.9	26	48	19	0.563
i31577_1041	1	52.9	26	48	19	0.563
i30515_574	1	54.5	26	49	18	0.439
i37083_872*	1	56.2	27	48	18	0.399
i31432_1654*	1	56.2	27	48	18	0.399
i26043_1262	1	56.7	26	49	18	0.439
i26043_1328	1	56.7	26	49	18	0.439
i31038_1437	1	56.7	26	49	18	0.439
i36219_573	1	56.7	26	49	18	0.439
i37206_878	1	56.7	26	49	18	0.439
i32512_171*	1	56.7	26	49	18	0.439
i33053_287*	1	56.7	26	49	18	0.439
i40160_557	1	57.2	26	50	17	0.322
i26783_856	1	57.2	26	50	17	0.322
i40160_437	1	57.2	26	50	17	0.322
i31077_592	1	63.5	24	49	20	0.736
i31077_970	1	63.5	24	49	20	0.736
i31077_501	1	63.5	24	49	20	0.736
i30950_933*	1	65.7	23	51	19	0.545
i38354_763	1	66.8	21	53	19	0.386
i31134_1070	1	66.8	21	53	19	0.386
i39326_587	1	69.6	23	52	18	0.399
i36532_814	1	72.9	28	46	19	0.416
i28615_1377*	1	75.1	27	48	18	0.399
i33710_856	1	78.5	26	46	21	0.760
i33710_284	1	78.5	26	46	21	0.760
i32235_511	1	79.0	26	45	22	0.802
i32235_925	1	79.0	26	45	22	0.802
i30917_789	1	82.9	28	44	21	0.516
i43680_368*	1	90.5	26	49	18	0.439

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Supplemental Table 1. Continued.

Locus ^z	Chrom	cM	Observed			P value
			B9885	Het	B8667	
i27259_617	1	94.4	25	50	18	0.454
i27259_657	1	94.4	25	50	18	0.454
i29102_2197*	1	95.0	24	51	18	0.439
i18286_1848	1	97.2	21	53	19	0.386
i27655_417	1	97.2	21	53	19	0.386
c00534_1708	1	110.7	24	48	20	0.770
i32287_1090*	1	110.7	24	49	20	0.736
i35260_950	1	110.7	24	49	20	0.736
c00534_1431	1	110.7	24	49	20	0.736
i12304_1152	1	110.7	24	49	20	0.736
i29256_270	1	110.7	24	49	20	0.736
i32674_652	1	110.7	24	49	20	0.736
c00534_1173*	1	110.7	24	49	20	0.736
i29256_196*	1	110.7	24	49	20	0.736
i30468_1110	1	118.1	21	52	20	0.516
i38094_540	1	121.0	20	53	20	0.403
i30789_1663	1	121.0	20	53	20	0.403
i20220_175*	1	125.5	21	53	19	0.386
i39789_529*	1	125.5	21	53	19	0.386
i34723_664*	1	130.1	22	47	24	0.953
i37397_482	1	135.9	21	45	27	0.647
i40509_271*	2	0.0	24	51	18	0.439
i27223_722*	2	0.0	24	51	18	0.439
i30750_1375*	2	0.0	24	51	18	0.439
i40509_373	2	0.0	24	51	18	0.439
i40509_454	2	0.0	24	51	18	0.439
i27567_733*	2	2.8	27	46	20	0.587
i35692_534	2	8.5	25	46	22	0.903
i16572_1239	2	12.4	27	43	23	0.647
i30197_935	2	12.4	27	43	23	0.647
i41662_224*	2	14.6	27	43	23	0.647
i42489_333	2	14.6	27	43	23	0.647
i40703_447*	2	14.6	27	43	23	0.647
i32690_448	2	19.1	23	47	23	0.995
i32690_809	2	19.1	23	47	23	0.995
i45110_199	2	19.1	23	47	23	0.995
i36256_344*	2	27.3	27	47	19	0.500
i28709_1940	2	34.3	22	49	22	0.874
i33051_1252*	2	40.7	21	48	24	0.865
i30946_1559	2	41.8	21	46	26	0.760
i30946_1170*	2	41.8	21	46	26	0.760
i32826_701*	2	44.0	20	50	23	0.698
i31070_860	2	58.6	25	46	22	0.903
i36775_363*	2	62.0	25	44	24	0.865
i39285_573	2	65.4	23	46	24	0.984
i34604_502	2	82.4	24	49	20	0.736
i36691_206	2	82.4	24	49	20	0.736
i26569_544	2	82.4	24	49	20	0.736
i40415_423	2	82.4	24	49	20	0.736
i40415_434	2	82.4	24	49	20	0.736
i34604_649	2	82.4	24	49	20	0.736
i19682_515*	2	84.6	23	51	18	0.443
i28284_2118*	2	87.4	23	50	20	0.698
i28284_1005*	2	87.4	23	50	20	0.698
i25587_1581	2	90.2	23	51	19	0.545
i33006_887	2	90.7	23	52	18	0.399

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Supplemental Table 1. Continued.

Locus ^z	Chrom	cM	Observed			P value
			B9885	Het	B8667	
i27483_359	2	92.4	21	53	19	0.386
i30965_1211	2	93.4	20	55	18	0.203
i30965_1461	2	93.4	20	55	18	0.203
i33705_468*	2	93.4	20	55	18	0.203
i30965_600	2	93.4	20	55	18	0.203
i32277_250	2	95.1	23	52	18	0.399
i32277_988	2	95.1	23	52	18	0.399
i30715_1591*	2	95.1	23	52	18	0.399
i32277_352	2	95.1	23	52	18	0.399
i32277_481	2	95.1	23	52	18	0.399
i32277_1124	2	95.1	23	52	18	0.399
i37633_393	2	96.7	24	51	18	0.439
i29186_1942	2	96.7	24	51	18	0.439
i37633_597	2	96.7	24	51	18	0.439
i34815_815	2	96.7	24	51	18	0.439
i20534_292	2	96.7	24	51	18	0.439
i28648_659	2	96.7	24	51	18	0.439
i33142_579*	2	96.7	24	51	18	0.439
i28648_2282	2	96.7	24	51	18	0.439
i30225_1454	2	96.7	24	51	18	0.439
i30225_524	2	96.7	24	51	18	0.439
i28648_1787	2	96.7	24	51	18	0.439
i33859_995	2	97.3	23	52	18	0.399
i33859_983	2	97.3	23	52	18	0.399
i33859_755	2	97.3	23	52	18	0.399
i35870_429	2	98.3	21	54	18	0.271
i35870_603	2	98.3	21	54	18	0.271
i30979_1313	2	98.3	21	54	18	0.271
i03354_1601*	2	98.3	21	54	18	0.271
i30979_525	2	98.3	21	54	18	0.271
i22791_480	2	112.8	27	42	24	0.587
i32865_1404*	2	116.1	29	40	24	0.308
i32695_459*	2	127.4	25	44	24	0.865
i32416_701	2	129.1	25	43	25	0.768
i25879_1232	2	135.6	25	40	28	0.366
i37220_578	2	137.2	22	43	28	0.522
i33538_1298*	2	138.3	23	43	27	0.647
i25059_110	2	138.3	23	43	27	0.647
i20252_1096*	2	138.3	23	43	27	0.647
i16559_476	3	0.0	20	52	21	0.516
i26975_1187*	3	3.4	22	46	25	0.903
i40425_584	3	5.6	20	46	27	0.587
i40425_522*	3	5.6	20	46	27	0.587
i29139_1610	3	6.1	21	45	27	0.647
i29139_382	3	6.1	21	45	27	0.647
i29139_1381	3	6.1	21	45	27	0.647
i29139_508	3	6.1	21	45	27	0.647
i29139_510	3	6.1	21	45	27	0.647
i29139_1024*	3	6.1	21	45	27	0.647
i32712_698	3	11.8	20	47	26	0.675
i32712_689	3	11.8	20	47	26	0.675
i32712_566*	3	11.8	20	47	26	0.675
i30297_1692*	3	12.4	20	48	25	0.728
i32564_718	3	15.1	19	47	27	0.500
i28424_1060*	3	15.7	18	48	27	0.399
i31416_1679*	3	19.6	19	49	25	0.594

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Supplemental Table 1. Continued.

Locus ^z	Chrom	cM	Observed			P value
			B9885	Het	B8667	
i33256_667*	3	19.6	19	49	25	0.594
i30944_607*	3	20.1	19	48	26	0.563
i28789_2782	3	21.2	20	48	25	0.728
i25371_2488*	3	22.3	19	48	26	0.563
i29424_1948	3	25.6	22	46	25	0.903
i34791_1013*	3	27.3	21	47	25	0.837
i34791_497	3	27.3	21	47	25	0.837
i31015_812*	3	28.4	20	49	24	0.736
i32375_378	3	31.2	21	48	24	0.865
i32581_207	3	40.7	23	46	24	0.984
i35740_662	3	54.4	15	54	24	0.125
i34505_441	3	54.4	15	54	24	0.125
i20004_416*	3	65.8	15	59	19	0.029
i40743_349*	3	66.9	16	57	20	0.079
i29422_1797	3	66.9	16	57	20	0.079
i30817_727*	3	69.4	17	55	21	0.178
i33909_665	3	73.6	17	55	21	0.178
i35663_432	3	73.6	17	55	21	0.178
i35352_760*	3	73.6	17	55	21	0.178
i29736_2072	3	76.4	16	56	21	0.110
i21577_861*	3	79.7	18	56	19	0.142
i42494_328*	3	79.7	18	56	19	0.142
i36487_789*	3	79.7	18	56	19	0.142
i16345_1175	3	85.4	23	50	20	0.698
i34054_350*	3	85.4	23	50	20	0.698
i28422_1226*	3	85.4	22	47	20	0.831
i28422_191	3	85.4	23	50	20	0.698
i35180_158*	3	85.4	23	50	20	0.698
i28422_1511	3	85.4	23	50	20	0.698
i32798_1400	3	85.4	23	50	20	0.698
i43361_292*	3	85.4	23	50	20	0.698
i16345_1005*	3	85.4	23	50	20	0.698
i33431_426*	3	85.9	23	49	21	0.837
i29185_1803	3	87.6	22	48	23	0.943
i30896_1092	3	87.6	22	48	23	0.943
i29185_2039	3	87.6	22	48	23	0.943
i29185_1241*	3	87.6	22	48	23	0.943
i37687_135*	3	88.7	23	46	24	0.984
i30509_198	3	89.7	22	48	23	0.943
i30509_1016	3	89.7	22	48	23	0.943
i32768_708	3	90.3	22	49	22	0.874
i30363_1184*	3	90.3	22	49	22	0.874
i30363_1478	3	90.3	22	49	22	0.874
i25849_1265	3	90.3	22	49	22	0.874
i30363_1650	3	90.3	22	49	22	0.874
i43826_494*	3	90.3	22	49	22	0.874
i25849_1264	3	90.3	22	49	22	0.874
i19473_316	3	90.3	22	49	22	0.874
i27896_600*	3	90.3	22	49	22	0.874
i31999_483	3	96.0	25	45	23	0.913
i14858_165	3	97.1	26	45	22	0.802
i14858_255	3	97.1	26	45	22	0.802
i14858_364	3	97.1	26	45	22	0.802
i18449_434	3	99.3	29	41	23	0.354
i29570_2011*	3	100.4	28	41	24	0.439
i28885_779	3	103.1	27	42	24	0.587

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Supplemental Table 1. Continued.

Locus ^z	Chrom	cM	Observed			P value
			B9885	Het	B8667	
i28885_2053	3	103.7	28	41	24	0.439
i32926_115*	3	112.5	30	40	23	0.238
i27361_431	3	113.0	30	39	24	0.203
i27361_174	3	113.0	30	39	24	0.203
i20406_751*	3	113.6	31	38	24	0.125
i42169_329*	3	117.5	31	37	25	0.097
i32969_715*	3	118.0	30	37	25	0.131
i32949_1292*	3	119.1	29	40	24	0.308
i32949_1307	3	119.1	29	40	24	0.308
i32949_1359	3	119.1	29	40	24	0.308
i45375_278	3	120.2	28	40	25	0.366
i33627_701*	3	120.2	28	40	25	0.366
i31106_505	4	0.0	27	43	23	0.647
i33886_426*	4	13.7	19	49	23	0.641
i16136_978	4	14.9	20	50	23	0.698
i16136_983	4	14.9	20	50	23	0.698
i16136_752	4	14.9	20	50	23	0.698
i16136_1083*	4	14.9	20	50	23	0.698
i36793_769*	4	21.2	23	45	25	0.913
i37821_673*	4	21.2	23	45	25	0.913
i41409_219*	4	23.4	24	45	24	0.953
i28561_1513*	4	28.5	26	42	25	0.640
i38440_478*	4	43.5	25	42	26	0.640
i38440_560	4	43.5	25	42	26	0.640
i34748_190*	4	47.5	25	43	25	0.768
i33429_1166	4	49.6	25	43	25	0.768
i37658_754	4	49.6	25	43	25	0.768
i34321_920*	4	50.7	23	45	25	0.913
i21411_854	4	50.7	23	45	25	0.913
i21411_971	4	50.7	23	45	25	0.913
i35268_979	4	51.8	24	45	24	0.953
i35268_784	4	51.8	24	45	24	0.953
i35268_1082*	4	51.8	24	45	24	0.953
i44683_192*	4	56.9	21	50	22	0.760
i44683_264	4	56.9	21	50	22	0.760
i37385_830*	4	60.8	20	53	20	0.403
i34670_762	4	60.8	20	53	20	0.403
i19574_601*	4	64.7	18	55	20	0.203
i32123_1465*	4	69.3	17	58	18	0.058
i29778_1782	4	69.3	17	58	18	0.058
i29778_1617*	4	69.3	17	58	18	0.058
i29243_971	4	72.0	20	54	18	0.238
i27809_650	4	73.1	18	57	18	0.093
i28612_1966	4	73.1	18	57	18	0.093
i28527_2095	4	73.1	18	57	18	0.093
i28527_2500	4	73.1	18	57	18	0.093
i28527_985	4	73.1	18	57	18	0.093
i28319_131*	4	75.9	18	54	21	0.271
i28319_140	4	75.9	18	54	21	0.271
i32504_1288	4	75.9	18	54	21	0.271
i28878_757	4	79.2	20	52	21	0.516
i26252_1243	4	86.8	20	51	22	0.620
c00160_1270	4	89.0	20	49	24	0.736
i31547_938	4	95.4	25	44	24	0.865
i31547_1204	4	95.4	25	44	24	0.865
i31547_1238	4	95.4	25	44	24	0.865

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Supplemental Table 1. Continued.

Locus ^z	Chrom	cM	Observed			P value
			B9885	Het	B8667	
i13427_579	4	95.4	25	44	24	0.865
i31547_1040	4	95.4	25	44	24	0.865
i13427_483	4	95.4	25	44	24	0.865
i30382_1919	4	107.5	18	48	27	0.399
i30382_300	4	107.5	18	48	27	0.399
i19074_908	4	108.6	19	46	28	0.416
i19074_994	4	108.6	19	46	28	0.416
i19074_815*	4	108.6	18	45	26	0.484
i31588_1120*	4	116.7	21	46	26	0.760
i31588_436	4	116.7	21	46	26	0.760
i29719_588*	4	117.8	19	48	26	0.563
i15820_1734	4	120.0	17	50	26	0.322
i29163_274	4	127.0	16	58	19	0.053
i32380_191*	4	127.6	15	59	19	0.029
i32177_996	5	0.0	24	46	23	0.984
i33448_1077	5	1.1	24	48	21	0.865
i15960_904	5	1.1	24	48	21	0.865
i33448_101*	5	1.1	24	48	21	0.865
i31544_957	5	6.8	24	46	23	0.984
i31544_1008	5	6.8	24	46	23	0.984
i32307_1162*	5	11.4	24	44	25	0.865
i39052_406	5	13.1	23	45	25	0.913
i35396_340	5	20.6	21	45	27	0.647
i27321_153*	5	28.1	23	43	27	0.647
i39512_345	5	29.8	23	42	28	0.494
i38068_242*	5	36.8	18	47	27	0.406
i31940_1090	5	43.8	17	46	30	0.162
i31940_928	5	43.8	17	46	30	0.162
i31940_820	5	43.8	16	46	30	0.119
i34751_952	5	51.9	16	49	28	0.186
i31628_1166*	5	57.1	15	51	27	0.138
i31628_1504	5	57.1	15	51	27	0.138
i31628_1613	5	57.1	15	51	27	0.138
i33430_179	5	57.6	15	52	26	0.142
i35125_448	5	57.6	15	52	26	0.142
i35125_241*	5	57.6	15	52	26	0.142
i32271_985	5	58.1	15	53	25	0.138
i31483_1602	5	61.5	15	51	27	0.138
i38424_472	5	64.8	13	55	25	0.045
i29003_1631	5	65.4	13	56	24	0.039
i29933_398	5	65.4	13	56	24	0.039
i29003_1460*	5	65.4	13	56	24	0.039
i16297_258	5	66.4	14	54	25	0.081
i28862_1377	5	66.4	14	54	25	0.081
i34069_110*	5	68.1	14	55	24	0.072
i31406_1529*	5	68.1	14	55	24	0.072
i28803_1689*	5	69.2	14	55	24	0.072
i28803_2444	5	69.2	14	55	24	0.072
i28803_236	5	69.2	14	55	24	0.072
i37926_754	5	74.0	15	59	19	0.029
i28536_1567*	5	83.7	14	59	20	0.024
i31727_537	5	84.2	14	60	19	0.015
i26546_969*	5	94.7	17	58	18	0.058
i34130_984	5	98.2	18	56	19	0.142
i07967_1919	5	99.9	19	55	19	0.211
i31477_611*	5	113.7	23	48	22	0.943

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Supplemental Table 1. Continued.

Locus ^z	Chrom	cM	Observed			P value
			B9885	Het	B8667	
i40014_661*	5	117.6	23	47	23	0.995
i31278_1407*	5	118.7	23	47	23	0.995
i37209_549*	5	118.7	23	47	23	0.995
i43201_503	5	119.8	24	45	24	0.953
i44638_388*	6	0.0	21	53	19	0.386
i31615_250	6	2.8	23	54	16	0.176
i06563_373	6	2.8	23	54	16	0.176
i29123_1522	6	7.3	26	52	15	0.142
i18674_1408	6	7.3	26	52	15	0.142
i29123_815	6	7.3	26	52	15	0.142
i18674_1075	6	7.3	26	52	15	0.142
i29123_1503*	6	7.3	26	52	15	0.142
c00298_108*	6	9.0	27	51	15	0.138
i30657_998	6	15.9	27	47	19	0.500
i28330_2105*	6	15.9	27	47	19	0.500
i29848_1923	6	16.4	28	46	19	0.416
i31703_857	6	18.7	28	44	21	0.516
i20052_1261	6	19.8	28	46	19	0.416
i32903_767	6	29.8	28	45	20	0.479
i32903_633	6	29.8	28	45	20	0.479
i31280_1328*	6	32.0	28	43	22	0.522
i35768_1013*	6	32.0	28	43	22	0.522
i31777_1516*	6	32.0	28	43	22	0.522
i29154_1644*	6	38.3	25	44	24	0.865
i19852_1220	6	45.9	27	45	21	0.647
i34771_689*	6	49.3	27	47	19	0.500
i16577_681*	6	49.3	27	47	19	0.500
i30848_646*	6	55.6	28	43	21	0.483
i35291_456*	6	55.6	28	44	21	0.516
i33185_888	6	56.1	27	45	21	0.647
i30880_1388*	6	56.6	26	46	21	0.760
i30880_478	6	56.6	26	46	21	0.760
i30738_359*	6	56.6	26	46	21	0.760
i29864_187	6	61.8	26	47	20	0.675
i30463_249	6	70.6	22	50	21	0.760
i28768_2758	6	74.5	23	51	19	0.545
i16030_756	6	76.7	24	49	20	0.736
i30686_364	6	78.9	23	47	23	0.995
i30686_371	6	78.9	23	47	23	0.995
i30686_719	6	78.9	23	47	23	0.995
i29491_511	6	80.6	21	47	24	0.887
i21395_588*	6	80.6	21	48	24	0.865
i15712_1480*	6	82.8	19	50	24	0.587
i26300_278	6	93.6	18	46	29	0.271
i26300_327	6	93.6	18	46	29	0.271
i30204_129*	6	99.9	19	45	29	0.325
i31266_1507*	6	101.5	17	46	30	0.162
i31266_752	6	101.5	17	46	30	0.162
i31266_696	6	101.5	17	46	30	0.162
i31425_789*	6	101.5	17	46	30	0.162
i31266_1737	6	101.5	17	46	30	0.162
i28455_440	6	102.6	16	46	31	0.089
i28455_379	6	102.6	16	46	31	0.089
i28455_1613	6	102.6	16	46	31	0.089
i28455_1895	6	102.6	16	46	31	0.089
i37324_287	6	103.7	17	46	30	0.162

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Supplemental Table 1. Continued.

Locus ^z	Chrom	cM	Observed			P value
			B9885	Het	B8667	
i33663_1163*	6	103.7	17	46	30	0.162
i33663_1189	6	103.7	17	46	29	0.209
i37324_542	6	103.7	17	46	30	0.162
i33493_569	6	103.7	17	46	30	0.162
i33663_319	6	103.7	17	46	30	0.162
i41141_540*	6	105.3	17	43	33	0.049
i30426_1135	6	105.9	18	42	33	0.058
i30426_1109	6	105.9	18	42	33	0.058
i28625_2789	7	0.0	25	47	21	0.837
i42645_520	7	0.0	25	47	21	0.837
i38160_373	7	6.3	21	52	20	0.516
i34205_651	7	7.9	20	53	20	0.403
i31632_1362*	7	10.7	19	56	18	0.142
i31632_1677	7	10.7	19	56	18	0.142
i41937_218*	7	11.2	19	55	19	0.211
i37252_318*	7	12.9	19	56	18	0.142
i37252_697	7	12.9	19	50	15	0.180
i32924_1099	7	15.1	21	51	19	0.492
i30695_284	7	15.1	22	52	19	0.474
c00456_208	7	15.1	22	52	19	0.474
i39918_486	7	16.7	23	51	19	0.545
i44788_441	7	16.7	23	51	19	0.545
i39918_357*	7	16.7	23	51	19	0.545
i44788_107	7	17.3	24	49	19	0.627
i33385_293*	7	18.4	23	50	20	0.698
i33385_518	7	18.4	23	50	20	0.698
i30655_190*	7	18.4	23	50	20	0.698
i33385_137	7	18.4	23	50	20	0.698
i26063_757	7	24.6	26	43	24	0.736
i26063_406*	7	24.6	26	43	24	0.736
i26063_439	7	24.6	26	43	24	0.736
i35377_1073*	7	24.6	26	43	24	0.736
i30182_1370*	7	24.6	26	43	24	0.736
i40501_344*	7	24.6	26	43	24	0.736
i29530_152*	7	32.2	27	42	24	0.587
i29530_461	7	32.2	27	42	24	0.587
i31941_1372	7	36.7	29	42	22	0.382
i40020_600	7	36.7	29	42	22	0.382
i44847_391*	7	38.9	29	42	22	0.382
i30642_156	7	41.1	29	42	22	0.382
i33650_1205	7	45.0	30	43	20	0.262
i37263_469	7	45.0	30	43	20	0.262
i36575_914	7	55.7	35	39	19	0.019
i36575_764	7	55.7	35	39	19	0.019
i36575_674	7	55.7	35	39	19	0.019
i29157_1149	7	56.8	35	41	17	0.016
i31770_176	7	60.7	32	42	19	0.105
i30735_456	7	67.1	28	43	22	0.522
i30735_621	7	67.1	28	43	22	0.522
i32031_755	7	68.2	29	43	21	0.386
i28692_1127	7	70.4	28	43	22	0.522
i26488_927*	8	0.0	27	41	25	0.500
i29465_759*	8	4.6	22	47	24	0.953
i29264_2200*	8	4.6	22	47	24	0.953
i29044_2564	8	10.2	21	50	21	0.706
i28920_2554	8	16.7	21	46	26	0.760

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Supplemental Table 1. Continued.

Locus ^z	Chrom	cM	Observed			<i>P</i> value
			B9885	Het	B8667	
i28367_3064	8	17.8	21	46	26	0.760
i28367_2185*	8	17.8	21	46	26	0.760
i28367_2330	8	17.8	21	46	26	0.760
i28367_2980	8	17.8	21	46	26	0.760
i28367_3109	8	17.8	21	46	26	0.760
i29155_779	8	18.9	21	46	26	0.760
i33645_542*	8	19.9	19	48	26	0.563
i29442_1161	8	24.5	20	48	25	0.728
i28373_866	8	27.2	20	49	24	0.736
i28373_3204	8	27.2	20	49	24	0.736
i28412_1165	8	28.9	21	46	26	0.760
i28412_1177	8	28.9	21	46	26	0.760
i31042_660	8	28.9	21	46	26	0.760
i31042_827	8	28.9	21	46	26	0.760
i28412_1084	8	28.9	21	46	26	0.760
i33362_1094*	8	28.9	21	46	26	0.760
i33298_528*	8	29.4	21	47	25	0.837
i32346_843*	8	30.5	22	47	24	0.953
i32535_303	8	31.0	23	46	24	0.984
i28432_3294	8	31.0	23	46	24	0.984
i43294_202*	8	31.0	23	46	24	0.984
i28432_1209	8	31.0	23	46	24	0.984
i32358_978*	8	31.0	23	46	24	0.984
i32535_462	8	31.0	23	46	24	0.984
i28432_1302	8	31.0	23	46	24	0.984
i33589_1176*	8	32.7	22	49	22	0.874
i30323_747*	8	32.7	22	49	22	0.874
i41653_558*	8	37.2	23	47	23	0.995
i19082_1721	8	41.1	21	48	24	0.865
i20235_630*	8	46.2	20	47	26	0.675
i28633_2705*	8	54.5	18	47	28	0.339
i20905_594	8	55.0	18	46	29	0.271
i32520_283	8	58.4	19	44	30	0.238
i30907_420*	8	60.7	20	42	31	0.176
i30913_1644	8	60.7	20	42	31	0.176
i30913_630	8	60.7	20	42	31	0.176
i30913_822	8	60.7	20	42	31	0.176
i31862_140*	8	62.3	20	43	30	0.262
i30020_476*	8	64.6	21	45	27	0.647
i28584_524	8	77.1	23	42	28	0.494
i31315_598*	8	78.2	22	42	29	0.382
i32428_1284	8	79.3	23	39	30	0.202

^zi = isotig; c = contig as described by Duangjit et al. (2013). Loci followed by an asterisk indicate SNPs not previously mapped by Duangjit et al. (2013) or by Damon and Havey (2014)).

Het = heterozygote.

Supplemental Table 2. Observed segregations, goodness-of-fit probabilities (*P*), and chromosome (Chrom) and centiMorgan (cM) positions for single nucleotide polymorphisms (SNPs) segregating in the F₂ family from B9897 × B5351 of onion.

Locus ^z	Chrom	cM	Observed			<i>P</i> value
			B9897	Het	B5351	
i37443_625	1	0.0	21	47	19	0.721
i31626_574	1	1.2	26	49	22	0.844
i32558_1278	1	18.2	31	44	23	0.312
i29102_2197	1	29.2	32	43	21	0.168
c00676_1004	1	29.7	31	44	22	0.286
i43680_368	1	32.6	29	42	22	0.382
i37265_852	1	37.2	25	46	22	0.903
i17679_1985	1	37.8	28	48	22	0.679
i31382_1702	1	41.0	29	44	25	0.510
c00172_269	1	56.4	25	48	25	0.980
i36532_752	1	58.0	22	51	24	0.844
i26470_847	1	61.7	20	56	22	0.353
i29923_361	1	71.2	18	48	27	0.399
i29455_1102	1	81.3	23	45	30	0.438
i33340_1003	1	100.1	25	47	26	0.912
i25793_1638	1	102.2	26	46	25	0.870
i39310_315	1	121.8	25	44	29	0.510
i27567_733	2	0.0	17	56	20	0.130
i34018_317	2	4.0	15	55	23	0.106
i32786_424 z	2	5.1	17	57	23	0.156
i30197_935	2	9.5	15	55	23	0.106
i41662_224	2	10.0	17	56	24	0.189
i28947_218	2	27.7	14	55	23	0.071
i32826_701	2	29.4	13	56	23	0.038
i31728_1335	2	42.2	11	60	22	0.005
i35805_506	2	58.6	19	55	23	0.355
i34382_910	2	60.8	21	56	21	0.368
i39908_252	2	60.8	20	55	21	0.357
i17237_4883	2	79.7	24	44	27	0.703
i25181_1307	2	80.5	21	45	27	0.647
i32987_1114	2	81.8	20	45	28	0.479
i09466_645	2	81.8	20	45	27	0.574
i29186_1830	2	81.8	20	45	28	0.479
i12617_836	2	82.4	21	44	28	0.516
i34671_610	2	82.9	22	47	29	0.559
i30979_1091	2	85.0	22	47	29	0.559
i28971_120	2	88.7	20	52	25	0.600
i32865_1404	2	94.0	19	52	22	0.474
i37220_578	2	113.1	22	51	20	0.620
i32786_424	2	206.9	23	57	17	0.156
i41662_224 z	2	212.0	24	56	17	0.189
i32712_566	3	0.0	27	43	26	0.588
i34929_648	3	6.6	25	45	23	0.913
i31068_1490	3	13.7	27	46	25	0.799
i35014_773*	3	16.6	25	42	26	0.640
i13565_195	3	22.6	21	50	26	0.738
i32139_534	3	46.5	23	45	25	0.913
i35882_404*	3	49.8	20	47	26	0.675
i34107_268	3	50.9	20	49	24	0.736
i33166_330	3	52.5	22	49	25	0.892
i30958_160	3	53.5	20	49	24	0.736
i30558_762	3	53.5	22	51	25	0.841
i35352_760	3	63.7	17	53	27	0.235
i33431_426	3	79.0	13	52	31	0.025
i29185_1241	3	81.1	15	52	31	0.061
i30453_1365	3	84.8	14	50	28	0.084

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Supplemental Table 2. Continued.

Locus ^z	Chrom	cM	Observed			P value
			B9897	Het	B5351	
i35214_301	3	85.4	14	49	28	0.089
i32041_215	3	85.4	17	52	29	0.191
i26005_1583	3	87.5	17	52	28	0.223
i29570_2011	3	96.7	18	45	30	0.203
i33627_701	3	112.2	18	52	28	0.300
i28982_1927	4	0.0	26	50	22	0.832
i36605_668	4	8.4	22	50	20	0.676
i26442_1225	4	15.3	23	50	20	0.698
i19902_1078	4	16.4	23	50	20	0.698
i26182_1158	4	26.2	27	49	17	0.298
i39401_224	4	26.2	27	49	17	0.298
i37158_542	4	30.9	26	54	18	0.312
i31465_1093	4	36.1	24	54	20	0.510
i37005_693*	4	39.1	25	48	20	0.728
i32407_591	4	39.1	25	51	22	0.841
i34783_896	4	40.6	26	50	22	0.832
i30027_971	4	41.7	26	52	20	0.576
i38830_623	4	42.7	25	51	21	0.745
i37023_265	4	43.2	25	50	18	0.454
i33399_1211	4	43.2	25	50	18	0.454
i32829_1057	4	45.3	26	51	21	0.714
i44683_192	4	47.1	27	49	17	0.298
i35268_1082	4	50.6	26	53	18	0.341
i28561_1513	4	68.0	30	49	19	0.291
i36793_769	4	75.8	32	45	21	0.210
i37821_673	4	76.4	32	46	19	0.154
i45610_340	4	91.3	32	42	19	0.105
i31106_505	4	100.8	28	48	22	0.679
i17653_1910	5	0.0	21	45	32	0.210
i41553_294	5	10.0	20	48	30	0.353
i36154_963	5	14.8	21	46	25	0.840
i31940_1090	5	38.3	22	47	29	0.559
i26178_547	5	45.6	23	45	28	0.639
i35125_241	5	50.6	20	48	29	0.432
i32130_277	5	50.6	20	49	29	0.438
i29167_1843	5	50.6	20	46	29	0.407
i33680_1027	5	50.6	19	47	27	0.500
i31628_1166	5	50.6	20	49	29	0.438
i33430_179	5	50.6	19	47	27	0.500
i37670_180	5	54.1	20	45	28	0.479
i34198_886	5	55.8	19	47	26	0.574
i38424_376	5	56.3	18	49	26	0.439
i31406_1529	5	57.9	19	51	28	0.403
i34069_110	5	57.9	18	48	27	0.399
i28889_2528	5	68.0	19	52	27	0.433
i30732_1102	5	69.1	20	50	28	0.510
i33965_554	5	82.5	19	54	25	0.416
i35535_407	5	83.5	19	51	27	0.454
i30652_1141	6	0.0	13	55	25	0.045
i29293_1435	6	105.7	15	47	31	0.063
i31425_789	6	191.2	26	46	20	0.676
i29703_1814	6	192.3	27	47	19	0.500
i33111_603	6	192.3	27	47	19	0.500
i30420_839	6	197.0	26	51	16	0.221
i28276_1535	6	246.7	28	48	17	0.259
i16577_681	6	253.2	30	43	20	0.262

Continued next page

Supplemental Table 2. Continued.

Locus ^z	Chrom	cM	Observed			<i>P</i> value
			B9897	Het	B5351	
i40098_630	6	256.0	27	46	20	0.587
i19852_1220	6	256.5	27	45	21	0.647
i10023_2319	6	257.6	27	45	21	0.647
i35768_1013	6	267.5	27	46	19	0.499
i31280_1328	6	267.5	27	46	20	0.587
c00298_108	6	295.5	29	50	14	0.068
i29123_1503	6	297.8	28	49	15	0.131
i32739_152	6	298.4	29	49	15	0.106
i36782_698	6	298.4	29	49	14	0.071
i31632_1362	7	0.0	21	55	17	0.178
i25801_1760	7	7.8	28	48	17	0.259
i36042_298	7	7.8	28	51	19	0.403
i28716_113	7	7.8	28	48	17	0.259
i40501_344	7	16.0	30	47	21	0.403
i28365_1152	7	27.7	27	52	18	0.337
i28398_1732	7	27.7	28	52	18	0.300
i34666_385	7	33.3	27	48	23	0.832
i26488_927	8	0.0	17	48	33	0.072
i30151_1442	8	10.7	16	51	30	0.117
i28367_2185	8	10.7	15	52	30	0.076
i12913_342	8	25.5	15	55	28	0.086
i33298_528	8	27.1	14	56	28	0.050
i32346_843	8	27.6	13	57	28	0.027
i28432_1104	8	28.6	13	55	30	0.025
i43294_202	8	29.6	13	50	30	0.034
i37010_277	8	29.6	14	53	31	0.038
i20266_1040	8	53.5	18	52	28	0.300
i38873_388	8	56.2	20	51	27	0.559
i28911_1986	8	59.9	21	52	25	0.707
i31315_598	8	73.3	24	51	20	0.653

^zi = isotig; c = contig as described by Duangjit et al. (2013). SNP names ending with z were phase-unknown markers and were analyzed in both phases, keeping the one showing linkage with phase-known markers; those indicated by * were not previously mapped by Duangjit et al. (2013) or by Damon and Havey (2014).

Het = heterozygous.