**Candidate genes affect the uptake of Mg2+ in *Brassica oleracea* near QTLs**

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Abstract

Magnesium is fundamental to the vitality of plants due to its roles as a common cofactor and as the central ion of chlorophyll. Without magnesium, a plant would not be able to photosynthesize or regularly distribute sugar. Therefore, sufficient magnesium concentrations are essential for plants to function and survive, and it is important to understand how plants acquire magnesium. We searched for genes involved in the uptake and storage of magnesium in the model plant organism, *Arabidopsis thaliana,* and broccoli, *Brassica oleracea*. We found *Arabidopsis* genes in the *B. oleracea* genome and to determine their distances from the magnesium QTLs in *B. oleracea*, which were provided by NCSU. We searched for more relevant genes within 1 Mbp of the QTL through exploring the *B. oleracea* genome on IGB. Candidate genes were associated with the uptake of magnesium and were within 1 Mbp of the QTL marker locations. Each candidate gene was assigned three SSR primers for further experimentation. Through BLAST, we found that of our nine BLASTed genes, CNGC10, which is an ion transport channel on the plasma membrane, was the closest to its QTL, at 1.9 Mbp away. Therefore, when we searched through IGB, we had no genes from BLAST to verify, as CNGC10 was not within 1 Mbp. We did not find many genes specific to magnesium uptake on IGB. Rather, we found multiple general cation exchangers and heavy metal or calcium transporters. Genes associated with calcium uptake were included due to magnesium’s inverse relationship with calcium. Due to the generality and the lack of information on the genes we found, it was difficult for us to effectively narrow down the gene list. Therefore, we labeled the 19 genes found through IGB and CNGC10, even though it is a little more than 1 Mbp away, as candidate genes, and provided 3 SSR primers for each gene. NCSU can use these SSR primers to experimentally determine the genes’ alleles and whether they are, in fact, linked to the QTL.

Introduction

Magnesium (Mg2+) is an essential component in many of plants’ most vital biological processes. For instance, it is a common cofactor for over 300 intracellular enzymes, including ATPases (Robinson, 2013). Due to its required function as a cofactor for ATPases, magnesium is an integral component in energy transfer and intracellular processes that require energy. Moreover, Mg2+ is the central ion in the porphyrin ring in chlorophyll. Therefore, without magnesium, the plant would not be able to photosynthesize. Magnesium accumulates the most, to concentrations of 30-50 mM, in the thylakoid lumen of the chloroplast (Bose *et al.,* 2010). Compared to its 0.5 mM concentration in the cytoplasm, Mg2+ is very concentrated in the thylakoid membrane. During photosynthesis, the magnesium ions move from the thylakoid lumen into the stroma, which usually has a Mg2+ concentration of 0.5-5 mM (Bose *et al.,* 2010). This transfer balances the negative change in charge due to photosynthesis and also activates RuBisCO, an enzyme essential for carbon fixation (Robinson, 2013). Thus, through its function as a cofactor and as a central component of photosynthesis, magnesium is essential to the vitality of plants.

Because the presence of magnesium is required for various biological processes, magnesium deficiency harms plants. Magnesium deficiency leads to chlorophyll breakdown, which can be first seen through the discoloration on the veins of mature leaves in many plant species (Hermans *et al*., 2010). Additionally, it causes an immobilization of sugar throughout the plant, possibly due to magnesium’s role as a cofactor for ATPases. This impairment causes sugar and starch to accumulate in the leaves (Hermans *et al.,* 2010). The breakdown of chlorophyll and the over-accumulation of sugar in leaves inhibits the plant’s ability to grow normally and to full capacity., Plants cannot function properly without a sufficient amount of magnesium.

Plants require a relatively high concentration of it for normal functioning. For instance, a study showed that, when compared to 28 other mineral elements, magnesium has the fifth largest “sufficient” concentration in plants, during which study sufficiency was determined by the concentration in tissue needed for the plant to reach 90% of its maximum yield (White *et al.,* 2010). The same study showed that magnesium has the third largest minimum toxic concentration. In other words, magnesium is only toxic to the plant at very high concentrations and its toxicity is, therefore, uncommon (White *et al.,* 2010).

Clearly, magnesium is a fundamental component of plants and is it, therefore, important to understand how plants take up and store Mg2+. This understanding requires research on what specific genes are involved in magnesium uptake in plants. Additionally, many of these genes could potentially be linked to quantitative trait loci (QTLs), which are segments of DNA that are associated with quantitative traits, such as magnesium allocation. The study of magnesium QTLs and their linked genes can allow us to understand how exactly plants acquire magnesium.

The purpose of our research is to find genes involved in the allocation and storage of magnesium in *Brassica oleracea*, the broccoli plant. If these genes are located near the magnesium QTLs that were given to us by Dr. Allan Brown from North Carolina State University (NCSU), than they are candidate genes that could be linked to the nearby QTLs. The ultimate goal of our research is to provide NCSU with the SSR (single sequence repeat) primers of our candidate genes. NCSU will then be able to use the SSRs to determine the specific alleles of the genes.

Methods

|  |  |
| --- | --- |
| **Chromosome** | **Location** |
| C06 | Bn-C6-p07628656 |
| C08 | Bn-C8-p40141039 |
| C05 | Bn\_A05\_22813805 |
| C07 | Bn-C7-p39973848 |
| C07 | Bn-C7-p35691416 |
| C04 | Bn-C4-p40219218 |

We searched PubMed for existing literature pertaining to genes that are known to be associated with the uptake and storage of magnesium in the model plant organism, *A. thaliana* (PubMed, 2014). After finding several relevant genes, we used the Basic Local Alignment Search Tool (BLAST) to compare the protein sequences of these genes to the *B. oleracea* genome, specifically using tBLASTn on the Genome Database for *Vaccinium*website (*Vaccinium* BLAST, 2014**)**. We referenced the resulting hits to the locations of the quantitative trait loci (QTL) markers associated with the uptake of magnesium in broccoli, which were provided by Dr. Allan Brown from North Carolina State University (Table 1). Hits were deemed significant if they were on one of the five chromosomes with magnesium QTLs, had e-values that were less than 10-6, and were within 10 million base pairs (Mbp) of the QTL marker on that chromosome. Because NCSU gave us the QTL marker for chromosome 5 in relation to the *Brassica rapa*genome (Bn\_A05\_22813805), genes that had hits on chromosome 5 were compared to the location of the closest significant QTL marker on *B. oleracea* (Bn\_C05\_44023210; Figure 1).

**Table 1.** QTLs for the storage and uptake of magnesium in *B. oleracea*

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**Figure 1** QTL map of chromosome 5 of *B. oleracea* genome. The magnesium QTL provided by NCSU is highlighted in yellow. QTLs in bold represent QTLs that have a strong correlation to their associated trait.

Using the Integrated Genome Browser (IGB), we explored the *B. oleracea* genome to search for more genes involved with the uptake and storage of magnesium in broccoli (IGB, 2014). Through searching IGB, we were able to meticulously consider every recorded gene within 1 Mbp of the given QTLs. If the gene name, or further research on the gene, suggested its involvement with magnesium, the gene was BLASTed through IGB, which redirected us to BLAST on the NCBI database (NCBI). Genes listed as “hypothetical proteins” or “unknown proteins” were not considered. The Arabidopsis Information Resource (TAIR) website and the NCBI database were used to provide more information about the genes found through both the primary research and IGB (TAIR, 2014; NCBI, 2014**)**.

After compiling a list of candidate genes that could support the location of the QTL marker for the uptake and storage of magnesium, we used the list of SSR primers that Dr. Allan Brown from NCSU provided to determine the primers for the candidate genes. We chose three primers with di- or trinucleotide repeats for each gene by prioritizing primers that were closest to the QTL location and had the highest number of repeats.

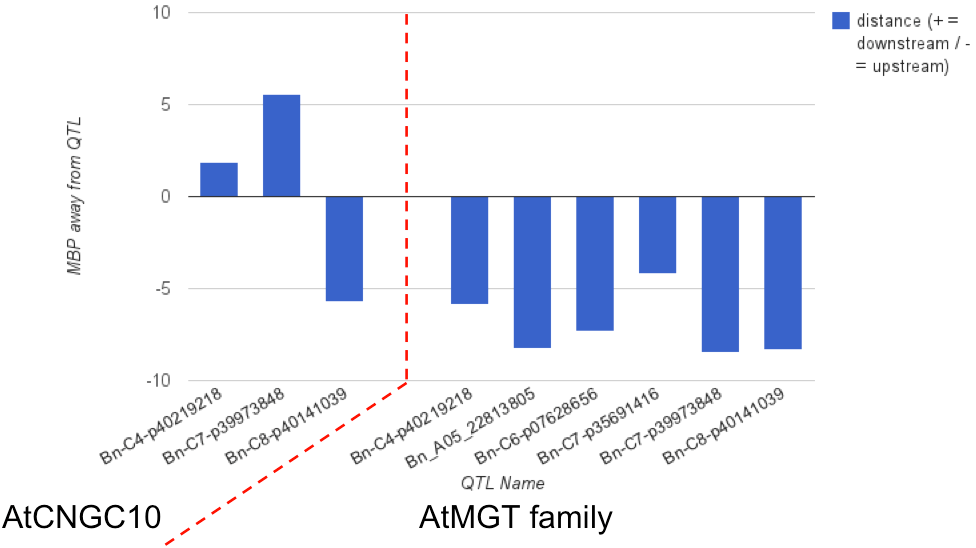
Results

Through exploring the research on magnesium transport and storage in *A. thaliana*, we found nine relevant genes to BLAST against the*B. oleracea* genome (Table 2). The biological process, cellular location, and function of the genes are shown in the table, and it also should be noted that all of these genes are expressed in many (more than 15) developmental stages and plant structures. These genes primarily consist of channels, such as AtCNGC10, and carrier proteins, such as AtMHX and the AtMGT genes, that transport magnesium within and between plant cells (Bose *et al.,* 2010**)**. Specifically, AtCNGC10 (cyclic nucleotide gated channels) mediates magnesium mobilization throughout the plant through its non-selective ion channels. Both AtCNGC10 and AtTPC1 have been shown to transport magnesium and calcium across the plasma membrane (Bose *et al.,* 2010; Robison, 2013). The AtMGT genes are a family of carrier proteins that vary in their affinity with magnesium and are sensitive to both aluminum and calcium concentrations (Bose *et al.,* 2010). This sensitivity is due to the competitive nature of the cations, as they use many of the same transporters.

**Table 2.** Genes involved in magnesium transport and uptake in *A. thaliana*. (**TAIR**)

|  |  |  |  |
| --- | --- | --- | --- |
| **Gene** | **Biological Process** | **Cellular Component** | **Molecular Function** |
| AtMHX | Cation/magnesium ion transport | Vacuolar membrane | Cation antiporter activity |
| AtTPC1 | Calcium ion transport | Plasma and vacuolar membranes | Calcium channel activity |
| AtCNGC10 | Ion transport | Plasma membrane | Ion channel activity |
| CorA-like family protein | Metal ion transport | Plasma membrane | Metal ion transmembrane transporter activity |
| AtMGT1, 5, 7a, 9, and 10 | Metal ion transport | Plasma membrane; mitochondrion; chloroplast | Magnesium ion transmembrane transporter activity |

Through BLASTing these genes, we found that two of the nine genes, the MGT family and CNGC10, had significant hits (Figure 2). Because each of the members of the MGT carrier protein family, AtMGT1, 5, 7a, 9, and 10, had significant hits on the same chromosomes, we grouped and labeled them the MGT family, instead of showing their results individually. This gene family had hits on every chromosome with a QTL marker for magnesium uptake. Additionally, CNGC10 had significant hits on multiple chromosomes, and had the closest hit of 1.9 Mbp away from the QTL on chromosome 4. Ultimately, the candidate genes, the MGT family and CNGC10, had significant hits for each QTL, which locations ranged from 1.9 to 8.3 Mbp from the QTL marker locations.



**Figure 2** Hits from BLASTing AtCNGC10 (first query) and AtMGT (second query) against the *B. oleracea* genome. The X-axis states which gene was searched, and what chromosome and QTL the gene was compared to.

Because our search on IGB was narrowed to 1 Mbp within the QTL marker location, we did not find any of our original candidate genes, including the MGT family and CNGC10. However, we did find many general cation exchangers and transporters, as well as calcium and heavy metal transporters (Figure 3).



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**Figure 3** Results from searching the *B. oleracea* genome on IGB for genes involved in the uptake and storage of magnesium within 1 Mbp of the QTL locations. The blue bars represent the five chromosomes that have QTLs for magnesium. The orange segment(s) on the chromosomes mark(s) the search areas, and these orange search areas are enlarged to the right of their respective chromosome, where the locations of the QTL markers and candidate genes are reported.

As Figure 3 shows, all of the chromosomes, except for chromosome 6, had relevant genes within 1 Mbp of the QTL locations. Specifically, on chromosome 4 we found a transporter that is hypothesized to be involved in magnesium transport, but we could not find concrete evidence on the function of the gene. We also found a general mitochondrial transporter, UCP5 Mitochondrial Dicarboxylate Carrier 1, which we considered relevant due to the fact that there are unknown mitochondrial transporters of magnesium (Bose *et al.,* 2010). Unlike the general transporters on chromosome 4, many of the genes on chromosome 5 and 7 were involved, specifically, in the transport of calcium, which is a heavy metal. These included the metallothionein-like protein, which binds heavy metals, the heavy metal transporters, the calcium transporting ATPase, and the calmodulin-like protein and caleosin-related family protein, which both bind calcium (NCBI, 2014). Chromosome 7 also had two V-type proton ATPases, which are involved in proton transport and could therefore affect cation proton exchangers, like the gene MHX and the other gene found on chromosome 7. Finally, chromosome 8 had many cation/proton or calcium exchangers. The descriptions of these three genes did not state what cations were specifically exchanged.

We then provided 3 SSR primers for the candidate genes, which included CNGC10 from our BLAST results, and all 19 of the genes found on IGB (Appendix A). The chosen SSR primers are within 100,000 base pairs of the QTL location, are larger than 100 base pairs, and have more than five di- or trinucleotide repeats.

Discussion

Although we expected some of the genes related to magnesium in *Arabidopsis* to appear and have the same function in *B. oleracea* and be positioned in close proximity to the provided QTLs, only one of our nine *Arabidopsis* genes was within 2 Mbp of a QTL. These far locations, for the majority of the genes, do not necessarily signify a difference of function in broccoli; rather, the distances suggest that these genes do not significantly impact the provided QTLs, and should, therefore, not be candidate genes.

MGT and CNGC10 were within 10 Mbp of the QTL location as found through BLAST, but neither was within 1 Mbp, our IGB search range. Therefore, we did not verify their presence on IGB and neither of them appeared in our IGB results. Because we had no previous genes to confirm through IGB and because we rarely found any specific magnesium transporters, we decided to be generous with the IGB genes we deemed relevant, and included general cation exchangers and calcium transporters. Although the calcium and heavy metal transporters do not directly affect the uptake and storage of magnesium, calcium and magnesium have an indirect relationship (Robinson 2013). Therefore, when the concentration of calcium is too high, which can occur if these calcium and heavy metal transporters are overexpressed, the plant can become magnesium deficient. Conversely, decreased expression of these calcium genes can lead to increased levels in magnesium (REF). Due to this inverse relation, we included the heavy metal transporters, metallothionein-like protein, calmodulin-like protein, and caleosin-related family protein.

Because of the lack of specificity of the genes that we found on IGB, such as the general cation transporters, we considered it necessary to include in our results as many of the broad, yet still relevant, genes as we could find. Additionally, we included CNGC10 as a candidate gene, even though it was over the 1 Mbp range, because it was the only gene specifically associated with magnesium transport. Of these candidates, we do not expect that the NCSU researchers will find every one linked to a QTL. Rather, I believe CNGC10, the cation exchangers on chromosome 5, and the heavy metal transporters will be the most likely successful candidates, due to their more direct relationship to magnesium. Nevertheless, we suggest all 20 as candidates that must be tested experimentally to determine if and which genes are linked to the QTLs.

We provided the SSR primers prompt this future experimentation. With the 3 SSR primers we chose for each of the twenty genes that we determined to be candidate genes, the researchers at NCSU can design oligonucleotides, amplify the SSRs through a polymerase chain reaction (PCR), and ultimately determine the genes’ specific alleles. This research will confirm whether the candidate genes we propose are linked to the QTLs.

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\*This journal article describes IGB its download information. Throughout this report, it is referenced just as IGB.

**Appendix 1**. SSR primers for chosen candidate genes.

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Gene Name** | **Chrom.** | **Gene Start** | **SSR ID** | **Size** | **Forward** | **Reverse** | **Motif** | **SSR Start** |
| Magnesium Transporter | C4 | 39,807,316 | 4,636 | 334 | TCCAACTATTATTTTCTCCA | CCGTTAAAACCTATTGTGTA | (TA)13 | 39,789,812 |
|  |  |  | 4,637 | 399 | GAACGTCAGTTGTCATAGTT | TCTGTTTTTATTTCAATGGT | (TA)10 | 39,798,676 |
|  |  |  | 4,638 | 378 | CCAATACATATTTACAAGGC | ATTAACCTAGGATTAGGGAA | (GA)7 | 39,818,088 |
| UCP5 Mitochondrial Dicarboxylate Carrier 1 | C4 | 40,692,677 | 4,757 | 380 | TCTAAATTATTTTGGCAATC | AATAGAACACCAAATCAAAA | (AG)6 | 40,680,595 |
|  |  |  | 4,758 | 382 | GTACAGATGATTTCCTTGAA | CTTATTCTTTTGCTGTTGTT | (AAG)5 | 40,696,771 |
|  |  |  | 4,759 | 254 | GTGAATAAGCTTGTGAATGT | AGTAAGTTTCACCAAAAACA | (AG)7 | 40,697,410 |
| CNGC10 | C4 | 42,081,858 | 4,958 | 337 | GTGAAGAAACAAACAAAGAG | ATAAAGAACGAATTACACGA | (AG)15 | 42,056,068 |
|  |  |  | 4,959 | 174 | AAAAACACTCGTTTATCTGA | TTATAATTACACACGCATTG | (AT)12 | 42,062,194 |
|  |  |  | 4,959 | 392 | TTCACCATTCCTATGTTTAC | AAATATTGTCTCATATTGCC | (TA)9 | 42,080,283 |
|  |  |  | 5,277 | 260 | TATCCTTTTTGAGAATTACG | TTTTCGAATTAACTAACCTG | (TC)14 | 43,808,462 |
| Calmodulin-like protein | C5 | 43,857,726 | 5,291 | 353 | TTTTTGTCGACTATCCATAC | ATACCTAGAGGTTTGAGGTC | (TTA)5 | 43,854,968 |
|  |  |  | 5,292 | 335 | ACAGACTCGTCAAAACTAAA | TATTGCTTCAATGTAGAGGT | (AG)6 | 43,875,620 |
|  |  |  | 5,289 | 244 | AGAGAAGATTGTGATTGCTA | TTGTCTTATCTTCATGGAAC | (AC)6 | 43,851,375 |
| Calcium transporting ATPase | C5 | 43,882,735 | 5,292 | 335 | ACAGACTCGTCAAAACTAAA | TATTGCTTCAATGTAGAGGT | (AG)6 | 43,875,620 |
|  |  |  | 5,295 | 399 | GAGGTCATGTATTCTACAGC | GGGTAAATAAATCAAAACAA | (TA)7 | 43,887,337 |
|  |  |  | 5,296 | 292 | TGCATTGTAAGTGTAGTGAA | GGGTAAATAAATCAAAACAA | (TG)6 | 43,894,566 |
| **Gene Name** | **Chrom.** | **Gene Start** | **SSR ID** | **Size** | **Forward** | **Reverse** | **Motif** | **SSR Start** |
| Heavy metal transport/detoxification superfamily protein | C5 | 44,678,689 | 5,382 | 391 | TTTCATACAAGGTTGTTTTT | CATCTTTTTCAGAGTCAGAG | (TA)6 | 44,678,112 |
|  |  |  | 5,383 | 346 | TTTAATAGAAAGAATTGCGT | ACATTCAAGGACACAATAAG | (TC)6 | 44,678,313 |
|  |  |  | 5,384 | 314 | GGAAAGAACAATAACAACAA | GTGAACTCATCATCAAACTC | (GAT)6 | 44,679,476 |
| V-type proton ATPase subunit d2 | C7 | 35,255,037 | 3,932 | 169 | TTCTCCTTCAATAACACATC | AAATTGTACAAGGAATTCAA | (AT)8 | 35,222,713 |
|  |  |  | 3,933 | 205 | AATGTCACAAATAATACGCT | AGATTTCGATTAGGTTTCTT | (GA)6 | 35,223,247 |
|  |  |  | 3,936 | 331 | GAAACCTCAACATAAGACAA | TTTAGTCTGTGGAACATCTC | (AGA)7 | 35,231,835 |
| Caleosin-related family protein | C7 | 35,570,359 | 3,983 | 293 | GAATACATTAACCGAAATGA | GTATAAAACACGGGATGATA | (TGA)5 | 35,572,640 |
|  |  |  | 3,984 | 367 | AAGCATGAAGTCATTAGAGA | TCATCATCATTTTCTTCTTC | (TG)8 | 35,574,108 |
|  |  |  | 3,985 | 256 | TAATAGAATATCGAGACGGA | TTGTTTGAAGATAAAACGAT | (AT)16 | 35,582,938 |
| Major facilitator superfamily protein | C7 | 35,825,037 | 4,026 | 360 | TACTCATCTCGTTAGACCAC | GTGGGATTCTTTTCTCTATT | (CT)7 | 35,814,654 |
|  |  |  | 4,028 | 346 | TAGTAAATGGTTGGGAATTA | TAACAGGTTTTGAAAGCTAC | (AAC)8 | 35,832,939 |
|  |  |  | 4,029 | 286 | CTCGTTTACGATTTGTATTT | AAGAACCTTCTTAGACCATC | (AT)14 | 35,833,599 |
| Heavy metal transport/detoxification superfamily protein | C7 | 36,474,786 | 4,120 | 282 | TATGAGCATGTTCTTACCTT | TGAGTTAAAGATTGGGATTA | (AG)8 | 36,474,867 |
|  |  |  | 4,121 | 207 | ACTAGATCTTCTTTTTCGGT | TTCTACTTTCGCCTAAAAC | (TC)16 | 36,478,622 |
|  |  |  | 4,124 | 324 | TTTCTGATTTAATTTTCAGC | GTCCACATTTAGTTTTGTTT | (AT)17 | 36,488,377 |
|  |  |  |  |  |  |  |  |  |
| **Gene Name** | **Chrom.** | **Gene Start** | **SSR ID** | **Size** | **Forward** | **Reverse** | **Motif** | **SSR Start** |
| Cation transport regulator-like protein | C7 | 36,606,567 | 4,137 | 330 | TGAAACATGCAATGTTAGTA | ATAAATCTATGAAGCGAAAA | (TA)6 | 36,602,291 |
|  |  |  | 4,139 | 383 | GGAGAAGAGAGAGGAGTAGA | GCAACAAAGAAAATAAATGT | (GA)8 | 36,615,709 |
|  |  |  | 4,140 | 369 | GATTTCAACTGTTTACTTGC | AGAACAGAGAAACAACACAC | (GT)7 | 36,623,055 |
| Vacuolar cation/proton exchanger | C7 | 39,433,988 | 4,554 | 332 | AAACAAACAAGTAAAGACCA | TGATATAGCGAAAGGTAGAG | (CT)6 | 39,418,808 |
|  |  |  | 4,555 | 293 | ATTGGTTGATAAATACGAAA | TTTATTGGGAAGAAGACATA | (TA)11 | 39,423,764 |
|  |  |  | 4,557 | 387 | CTTAGCGTAACTCTTAACCA | AGACATGCATAATCAAGAAC | (TC)17 | 39,426,991 |
| Cation proton exchanger | C7 | 39,509,003 | 4,567 | 238 | TCAATCTTCTTTTGTTTTGT | TATGTCTGCGTCTAGAGATT | (TC)9 | 39,500,473 |
|  |  |  | 4,568 | 391 | TTTTACCTCAGCTATTCTTG | ATTCAGTTATGATATGGTCG | (TCT)5 | 39,508,358 |
|  |  |  | 4,571 | 366 | TCTTTACGTTATTCCATTTC | GATCAAGGAAGTAAACATGA | (CT)9 | 39,521,603 |
| V-type proton ATPase 16 kDa proteolipid subunit c1/c3/c5 | C7 | 39,793,003 | 4,605 | 322 | CATTCACTCAATCTCAAACT | ACAAATGACTTGACACAGAT | (GA)7 | 39,775,858 |
|  |  |  | 4,606 | 198 | TTAAGTGTCAAATTTTTGGT | AATGTTTCGACTAAGCTATG | (AT)9 | 39,795,984 |
|  |  |  | 4,608 | 368 | CTAAGCGTAATTTTCAGTGT | ACAGAAAACAAAAATTTGAA | (TA)8 | 39,819,167 |
| Heavy metal transport/detoxification superfamily protein | C7 | 40,122,906 | 4,657 | 246 | ATACAGACAGCAAAAAGAGA | AAACAAAACAAGAATGAGAA | (TC)6 | 40,122,838 |
|  |  |  | 4,658 | 381 | CTTAAACGCATTTTACTGAT | TAAAATAGGATTGAAGACCA | (CT)9 | 40,129,784 |
|  |  |  | 4,659 | 365 | GAAAATTCTCACTGTTTTTG | GTTTTGATTTCTTGTTTCAG | (TA)7 | 40,132,933 |
|  |  |  |  |  |  |  |  |  |
| **Gene Name** | **Chrom.** | **Gene Start** | **SSR ID** | **Size** | **Forward** | **Reverse** | **Motif** | **SSR Start** |
| CAX11 Cation/Calcium Exchanger 5 | C8 | 39,717,422 | 5,014 | 356 | ATCTCTTTGAACTGGTTCTC | TTGAGTCTTTCATACTCACC | (GCT)7 | 39,713,408 |
|  |  |  | 5,015 | 230 | GTGATGAGGATGTAGAAGTG | ATCACAAACTCAGCTCTATG | (GGA)5 | 39,717,335 |
|  |  |  | 5,016 | 298 | ATTTATGTACATGACAACCC | TTACTAGAGATTTTAGCCCA | (TA)13 | 39,728,082 |
| CHX6A Cation/H+ Exchanger 6A/6B | C8 | 39,945,936 | 5,044 | 293 | TTCTTTCAATATCCCACTAA | ATAGTCGACGAACTTATCAA | (TC)6 | 39,931,532 |
|  |  |  | 5,045 | 321 | AACCAATACCAAGTACACAG | TCAAGAAATCTTAGAAGCTG | (TA)8 | 39,940,309 |
|  |  |  | 5,046 | 115 | GTAAGAGCCTTTCTTTTCTT | TTAATAGGAGAACTTGTGGA | (AT)7 | 39,942,444 |
| CHX14 Cation/H+ Antiporter 14 | C8 | 40,277,578 | 5,082 | 338 | AAAAGAAAGAGAAGGTGTTT | ATCAATAGAAAATGGATGTG | (CA)6 | 40,263,468 |
|  |  |  | 5,084 | 379 | ATTTCATAAAAGACATCGAA | CACATCTTTCTTCTCTATGC | (CT)7 | 40,294,106 |
|  |  |  | 5,085 | 221 | AAGTTTGAATCCTCTCTCTC | AAACATTTAAACAGAAGCTG | (TC)12 | 40,294,830 |