**Supplementary Table S1. Genetic status of the known altered genes in UM cell lines.**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Cell line | Tumor | GNAQ mutation | GNA11 mutation | BAP1 status | SF3B1 status | EIF1AX status |
| 92.1 | Primary | Q209L | – | WT | WT | Heterozygous mutation in exons 1 and 2: c.17G/A; p.Gly6Asp |
| Mel270 | Primary | Q209P | – | WT | WT | WT |
| Omm1 | Subcutaneous metastasis | – | Q209L | WT | WT | WT |
| Omm2.3 | Liver metastasis | Q209P | – | WT | WT | WT |

**Supplementary Table S2. Detailed information for 45 patients with UM.**

|  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Pt# | S/Y | Location | LBD (mm) | Thickness (mm) | Therapy | Vimentin | HMB45 | S100 | Ki67 | TNM7 | NAE1 |
| 1 | M/60 | Choroid | 11.5 | 6.2 | Enucleation | + | + | + | 5% | T2 | N |
| 2 | F/45 | Choroid | 10.6 | 8.2 | Enucleation | + | + | + | 20% | T2 | L |
| 3 | F/51 | Choroid | 14.5 | 11.6 | Enucleation | + | + | + | 15% | T3 | N |
| 4 | M/64 | Choroid | 11.8 | 6.2 | Enucleation | + | + | + | 10% | T2 | L |
| 5 | M/61 | Choroid | 10.6 | 22.1 | Enucleation | + | + | + | 10% | T4 | H |
| 6 | M/44 | Choroid | 15.1 | 14.6 | Enucleation | + | + | + | 20% | T4 | L |
| 7 | F/51 | Choroid | 12.8 | 5 | Enucleation | + | + | + | 10% | T2 | N |
| 8 | F/57 | Choroid | 16.1 | 15.5 | Enucleation | + | + | + | <5% | T4 | H |
| 9 | F/46 | Choroid, sclera ingrowth | 11.49 | 8.34 | Enucleation | + | + | + | 15% | T2 | N |
| 10 | M/47 | Choroid | 10.2 | 9.2 | Enucleation | + | + | + | 10% | T3 | L |
| 11 | F/46 | Choroid | 9.0 | 15.9 | Enucleation | + | + | + | 25% | NA | L |
| 12 | M/37 | Choroid | 11.2 | 10.1 | Enucleation | + | + | + | 20% | T3 | H |
| 13 | F/60 | Choroid | 12.5 | 9.3 | Enucleation | + | + | + | 40% | T3 | L |
| 14 | M/42 | Choroid | 18.3 | 17.1 | Enucleation | + | + | + | 10% | T4 | H |
| 15 | M/28 | Choroid | 10.5 | 13.5 | Enucleation | + | + | + | 5% | T3 | L |
| 16 | F/57 | Choroid | 9.9 | 11.3 | Enucleation | + | + | + | 5% | T3 | N |
| 17 | F/35 | Choroid | 17 | 15 | Enucleation | + | + | + | 10% | T4 | H |
| 18 | M/45 | Choroid | 214.4 | 10.4 | Enucleation | + | + | + | 10% | NA | N |
| 19 | M/44 | Choroid | 11.6 | 4.1 | Enucleation | + | + | + | 10% | T2 | M |
| 20 | F/45 | Choroid | 13.3 | 11.8 | Enucleation | + | + | + | 10% | T3 | M |
| 21 | F/52 | Choroid/Ciliary body  | 17.4 | 13 | Enucleation | + | + | + | 10% | T4 | L |
| 22 | M/67 | Choroid | 11.4 | 14.3 | Enucleation | + | + | + | 11% | T3 | L |
| 23 | F/45 | Choroid | 11.5 | 10.1 | Enucleation | + | + | + | 10% | T3 | M |
| 24 | M/41 | Choroid | 14.2 | 8.2 | Enucleation | + | + | + | 20% | T3 | M |
| 25 | F/64 | Choroid | 8.5 | 11.1 | Enucleation | + | + | + | 10% | T3 | N |
| 26 | M/65 | Choroid | 21.2 | 12.1 | Enucleation | + | + | + | 30% | T4 | H |
| 27 | F/60 | Choroid | 11.8 | 11.4 | Enucleation | + | + | + | 10% | T3 | H |
| 28 | F/45 | Choroid | 14.1 | 9.4 | Enucleation | + | + | + | 20% | T3 | H |
| 29 | M/30 | Choroid | 11 | 16.4 | Enucleation | + | + | + | <5% | T4 | M |
| 30 | M/23 | Choroid, sclera ingrowth | 17.7 | 15.4 | Enucleation | + | + | + | <5% | T4 | H |
| 31 | F/48 | Choroid | 9.3 | 13 | Enucleation | + | + | + | <5% | T3 | H |
| 32 | M/64 | Choroid | 12 | 11 | Enucleation | + | + | + | <5% | T3 | N |
| 33 | M/63 | Choroid | 10.7 | 9.5 | Enucleation | + | + | + | 8% | T3 | L |
| 34 | F/50 | Choroid | 12.5 | 11.2 | Enucleation | + | + | + | 5% | T3 | L |
| 35 | M/77 | Choroid | 16.9 | 15.9 | Enucleation | + | + | + | 5% | T4 | H |
| 36 | F/60 | Choroid | 11.5 | 9.4 | Enucleation | + | + | + | <5% | T3 | M |
| 37 | M/27 | Choroid | 8.1 | 6.1 | Enucleation | + | + | + | <5% | T2 | L |
| 38 | F/38 | Choroid | 18.5 | 12.6 | Enucleation | + | + | + | <5% | T4 | M |
| 39 | M/38 | Choroid | 6.6 | 12.8 | Enucleation | + | + | + | <5% | NA | N |
| 40 | M/62 | Choroid/ Ciliary body | 16.3 | 9.7 | Enucleation | + | + | + | <5% | T3 | M |
| 41 | M/41 | Choroid/ Ciliary body | 11.8 | 11.3 | Enucleation | + | + | + | <5% | T3 | L |
| 42 | M/67 | Choroid | 10.5 | 19.3 | Enucleation | + | + | + | <5% | T4 | H |
| 43 | F/60 | Choroid/ Ciliary body | 16.5 | 10.7 | Enucleation | + | + | + | <5% | T3 | H |
| 44 | F/49 | Choroid, sclera ingrowth | 13.3 | 7.5 | Enucleation | + | + | + | <5% | T3 | M |
| 45 | M/49 | Choroid, sclera ingrowth | 14.1 | 13.4 | Enucleation | + | + | + | <5% | T3 | N |

Pt#: Patient number; S/Y: Sex/Year; LBD: Largest basal diameter; N: Negative; L: Low; M: Medium; H: High; TNM7: T, Primary tumor; N, Regional lymph node metastasis; M, Systemic metastasis. NA: not available.

**Supplementary Table S3. The association between NAE1 expression and clinicopathologic features in UM patients.**

|  |  |  |
| --- | --- | --- |
| **Variables** |  **NAE1** |  |
| **Cases** | **Negative** | **Low** | **Medium** | **High** | **p Value§** |
| Age (y) |  |  |  |  |  | 0.1327 |
|  ≤50>50  | 2520 | 47 | 85 | 71 | 67 |  |
| Gender Male Female | 2520 | 56 | 85 | 44 | 76 | 0.8818 |
| The largest basal diameter (mm) ≤15 >15 | 2817 | 92 | 103 | 53 | 49 | 0.0373 |
| Thickness (mm) ≤12 >12 | 2322 | 83 | 76 | 62 | 211 | 0.0461 |
| Ki67 expression level ≤10% >10% | 3411 | 92 | 85 | 71 | 103 | 0.5226 |
| TNM StageT1T2T3T4 | 072312 | 0360 | 0372 | 0152 | 0058 | 0.0374 |

§ Chi-square test.

**Supplementary Table S4. Limiting dilution analysis in NOD-SCID mice.**

|  |  |
| --- | --- |
| Cell number | Engrafted mice |
|  | Control | MLN4924 |
| 3×106 | 6/6 | 1/6 |
| 1×106 | 6/6 | 0/6 |
| 5×105 | 4/6 | 0/6 |
| 1×105 | 0/6 | 0/6 |
| Frequency | 1/433,059 | 1/26,071,238 |

**Supplementary Table S5. Primers for qRT-PCR analysis.**

|  |  |  |
| --- | --- | --- |
| Genes | Sense primer | Antisense primer |
| NAE1 | 5'-ATCATGTTGCCAAATTGCTG-3' | 5'-CCACTCGAAGAAATGCAGAA-3' |
| UBA3 | 5'-GAATGCACGCTGGAACTTTA-3' | 5'-GCCACTGCAACATCCTTACA-3' |
| UBC12 | 5'-GCGGATCCAGAAGGACATA-3' | 5'-GGACAGATGACCAGCTTGAA-3' |
| NEDD8 | 5'-GACCGGAAAGGAGATTGAGA-3' | 5'-GCTTGCCACTGTAGATGAGC-3' |
| BCL2L1VEGF-AVEGF-BVEGF-CVEGF-Db-FGF | 5'-ACAGCAGCAGTTTGGATGCC-3'5'-TCCTGGAGCGTGTACGTTGG-3'5'-ACGATGGCCTGGAGTGTGTG-3'5'-GTCCGGACTCGACCTCTCG-3'5'-GCAAAGAACTCAGTGCAGCC-3'5'-AGCCGGGAGCATCACCAC-3' | 5'-GGGTGATGTGGAGCTGGGAT-3'5'-TTCTCCGCTCTGAGCAAGGC-3'5'-AGGTCTGCATTCACACTGGCT-3'5'-CATCTACACTGGACACAGACCGT-3'5'-CGGAACACGTTCACACAAGG-3'5'-GTGGATGCGCAGGAAGAAGC-3' |
| GAPDH | 5'-GATCGAATTAAACCTTATCGTCGT-3' | 5'-AGCAGCAGAACTTCCACTCGGT-3' |