

Gene expression for HIV-associated dementia and HIV encephalitis in microdissected neurons I: preliminary analysis

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Abstract: We analyzed gene expression in neurons from 16 cases divided into four groups, ie, human immunodeficiency virus (HIV)-associated dementia (HAD)/HIV encephalitis (HAD/HIVE), HAD alone, HIVE alone, and HIV positive alone. We produced the neurons using laser capture microdissection from cryopreserved basal ganglia (specifically globus pallidus). Gene expression in pooled neurons from each case was analyzed on GE CodeLink Microarray chips with 55,000 gene fragments per chip. One-way analysis of variance showed significant changes in expression of 197 genes among the four groups ($P < 0.005$). The three groups, ie, HAD/HIVE, HAD alone, and HIVE alone, were compared with the HIV-positive group using Fisher's least significant difference test, and associated gene expression changes were assigned to each of the three comparisons. Identified genes were associated with 159 functional categories and many of the genes had more than one function. The functional groups included adhesion, amyloid, apoptosis, channel complex, cell cycle, chaperone, chromatin, cytokine, cytoskeleton, metabolism, mitochondria, multinetwork detection protein, sensory perception, receptor, ribosome, noncoding miRNA, signaling, synapse, transcription factor, homeobox, transport, multidrug resistance, and ubiquitin cycle. Several genes were associated with other neurodegenerative and developmental diseases, including Alzheimer's disease, Huntington's disease, and diGeorge syndrome. Thus, a wide range of dysregulated biochemical processes was reported in neuroanatomically precise neurons. This line of investigation is useful and provides specific information about gene expression dysfunction in NeuroAIDS.

Keywords: NeuroAIDS, human immunodeficiency virus, dementia, encephalitis, laser capture microdissection, globus pallidus, neuron, genes, expression

Introduction

The term "NeuroAIDS" has been used generally to describe the involvement of the central nervous system in human immunodeficiency virus (HIV)-related disease. The neurodegenerative processes in HIV infection result in neurocognitive decline, which range in severity from asymptomatic neurocognitive impairment to minor neurocognitive disorder and to HIV-associated dementia (HAD). There have been changes in the definitions during the last decade that evolved to the current term used for these conditions, ie, HIV-associated neurocognitive disorders (HAND). The precise pathogenesis of neurodegeneration associated with HIV infection is still unclear. However, neurons are the final targets of the neurodegenerative process, and additional cells are involved as well. The concomitant damaged substrate exhibits brain inflammation that is associated with HIV infection, ie, HIV encephalitis (HIVE), and can also involve macrophage/microglial infiltration and astrocytosis.¹⁻⁵ Although

the incidence of HAND has decreased due to combination antiretroviral therapy and other treatments, in recent years the prevalence of HAND has actually increased. Several factors are responsible for this trend, including side effects of combination antiretroviral therapy, increased longevity of patients, and viral mutations including drug resistance.⁵⁻¹⁰ The importance of psychiatric symptoms, including anxiety and depression, are relevant as well because they are components of the stressors to which the brain is subject.^{2,6,10,11} Tissues from the cases utilized here derive from the period prior to the advent of HAND classifications and reflect earlier work.

Several studies of gene expression in culture and in postmortem brain tissue relate to gene expression in patients who died with HIVE and HAND and have been reviewed.^{5,12-19} Most brain studies analyzed RNA purified from small aliquots of brain tissue dissected from postmortem specimens. One such study, for example, utilized the frontal cortex from five subjects infected with HIV-1 and four controls negative for HIV-1 using microarrays. These two groups were analyzed by K-means cluster analysis. Genes with perturbed expression were identified that included cell cycle, inducible nitric oxide, chemokine, splicing, synapse, ribosomal proteins, maltose binding protein, myelin proteolipid protein, N-methyl-D-aspartic acid receptor, myelin-associated glycoprotein, astrocytic protein, Notch 3, amyloid precursor protein, senescence, proteasome, ferritin, and signaling.²⁰ In related work, IFN- γ showed increased expression in brain tissue from patients who died with NeuroAIDS and drug abuse compared with controls, while other cytokines did not show elevation.²¹ In another study, gene expression in gray matter from the frontal lobe was analyzed using microarrays comparing cases with HIVE versus control cases without HIVE. This study indicated that HIV-1 infection in brain tissue associated with HIVE resulted in neurodegeneration and interfered with genes that regulate the cytoskeleton, synaptic-dendritic integrity and function, and signaling, and induced a neuroinflammatory response. Seventy-four genes were downregulated and 59 genes were upregulated. Downregulated genes had functions related to signaling (phosphatidylinositol-3-kinase, Ras-Raf-MEK1), transcription, cytoskeleton (MAP-1B, MAP-2, tubulin, adducin-2), the cell cycle (p35, p39, CDC-L2, CDC42, PAK1), synaptic plasticity, and synaptic transmission (ion channels, synaptogyrin, synapsin II). Upregulated genes had functions related to signaling modulation (MEK3, EphB1), cytoskeleton (myosin, adducin-3, radixin, and dystrobrevin), transcription (STAT1, OLIG2, Pax-6), neuroimmune response (immunoglobulin G,

major histocompatibility complex, β_2 -microglobulin) and antiviral response (interferon inducible).²²

Gene expression profiles related to astrocytes were shown to have many similarities across differing brain tissues (from patients with HIV-1 dementia and from macaques infected with simian immunodeficiency virus) and included several human and murine astrocyte cell culture systems. The use of astrocyte culture systems in the study of NeuroAIDS is supported because of the similarity of gene expression profiles in brain tissue and cultured cells and because astrocytes constitute a large percentage of cells in brain tissue. Several in vitro studies utilized HIV-1 and HIV-1 proteins, ie, Tat, envelope glycoprotein gp120, or negative regulatory factor. The correspondence of gene expression perturbed in these systems and in the brain includes cytokines, chemokines, and their receptors, and is also consistent with astrocyte activation.^{23,24}

Neuronal cell cultures are also model systems. For example, a neuronal culture model of the dysfunctional NeuroAIDS brain including drug abuse utilized eight treatment conditions ($2 \times 2 \times 2$), with and without each of cocaine, Tat, and envelope protein. Statistically significant perturbation of gene expression was demonstrated for 35 genes across all treatment conditions using one-way analysis of variance. Functions of these genes included signaling, immune-related functioning, and transcription control.²⁵

Human brain cortex middle frontal gyrus gene expression profiles were compared for cases of HAD or milder cognitive dysfunction versus HIV-negative cases. This work focused on neuronal dysfunction and possible relationships with subcortical dementia. Genes studied were ionic conductance carriers that control membrane excitation. Overexpressed genes included calcium-driven K^+ channel, leak type of K^+ channel, adenosine receptor, serotonin receptor, and the gamma aminobutyric acid receptor subunit. Underexpressed genes included two voltage-gated K^+ channels, a Na^+ channel subunit, a neuronal type of voltage-sensitive Ca^{2+} channel, a metabotropic glutamate receptor, and the N-methyl-D-aspartic acid receptor subunit. Although unfractionated tissue was used, the perturbed gene expression was considered to stem from neurons because changed expression of these genes changes did not occur in gyral white matter and were not associated with overall changes in glial markers. Moreover, these changes occurred with HAD, with and without HIVE, and were not associated with increased inflammatory gene expression.^{26,27} The Trojan horse model predicts that HIV-1-infected monocytes are a risk for brain penetration of

HIV-1 via monocyte trafficking into the brain.²⁸ Surface gene expression associated with such cells included CD14, CD68, CD14a, and HLA-DR.²⁹ Pulliam et al studied gene expression on CD14+ monocytes from HIV-infected cases. Cases with high virus load showed increased expression of sialoadhesin, CD16, CCR5, and MCP-1. However, proinflammatory cytokine gene expression (interleukin-1, interleukin-6, and tumor necrosis factor- α) was unchanged.³⁰

Microarray analysis in a monkey model using frontal lobe tissue from simian immunodeficiency virus-infected brains identified 98 genes with altered expression. Genes expressed were associated with promoting macrophage entry into the brain and associated toxic products. Those significantly upregulated included proteins in infiltrating macrophages, endothelial cells, and resident glia (eg, CD163, Glut5, and ISG15). Proteins found in cortical neurons included cyclin D3, tissue transglutaminase, α 1-antichymotrypsin, and STAT1.³¹

Laser capture microdissection has been used successfully in the study of several human brain diseases, including the HIV-1-infected brain, subacute sclerosing panencephalitis, Parkinson's disease, and Huntington's disease.^{32–39} In the current study, only cases with HAD and HIVE (as well as HIV-1-positive controls) were used. Thus, work in NeuroAIDS has progressed to the point where cell-specific studies will be able to elucidate additional information using novel approaches. We report on gene expression in specific neuroanatomically defined neurons.

Materials and methods

Brain tissue

As previously described,³⁹ autopsied cryopreserved brain tissue was obtained from the National Institutes of Health-sponsored National NeuroAIDS Tissue Consortium sites^{40,41} (Table 1). At each of the National NeuroAIDS Tissue Consortium sites, the diagnosis of HIV-1-positive individuals with and without HAD and HIVE was made based on premortem neurological and clinical neuropsychological examination of the patients and at postmortem by neuropathological examination. Each subject was given a diagnosis, using a standardized, algorithmic diagnostic worksheet to combine neurological, neuropsychological, functional, and laboratory information. Postmortem tissues were examined by board-certified neuropathologists to exclude subjects with opportunistic central nervous system infections, tumors, or other causes of dementia, such as Alzheimer's disease. Furthermore, most subjects were below the age at which a dementing neurodegenerative illness would be expected.^{7,8,11–13,42} Tissue was dissected from the globus pallidus and embedded in optimal cutting temperature compound. Sections 10 microns thick were cut using a cryostat at -23°C . The cryosections were mounted on laser capture microdissection slides (Microoptics of Florida, Palm Beach, FL). Prior to laser capture microdissection, the slides were cryopreserved at -80°C in sealed Bakelite slide boxes containing drierite.³⁹

Table 1 Patient demographics and diagnosis

| Subject number | HAD | HIVE | Gender | Race | Ethnicity | Hispanic | Risk | Duration HIV infection Y | Age at death Y |
|----------------|-----|------|--------|-------|-----------|----------|------------|--------------------------|----------------|
| 1 | + | + | M | Cauc | – | – | MSM | 17 | 47.30 |
| 2 | + | + | M | Cauc | + | – | MSM | 4 | 44.13 |
| 3 | + | + | M | Black | – | – | MSM | 10 | 43.53 |
| 4 | + | + | M | Cauc | – | – | MSM | 3 | 35.42 |
| 5 | + | – | F | Black | – | – | IDU | 7 | 64.96 |
| 6 | + | – | F | Cauc | – | – | BPR HS | 3 | 58.27 |
| 7 | + | – | M | Cauc | – | – | IDU | 13 | 62.48 |
| 8 | – | + | M | Cauc | + | – | HS IDU | 12 | 33.10 |
| 9 | – | + | M | Black | – | – | BPR HS MSM | 12 | 46.75 |
| 10 | – | – | M | Cauc | + | – | MSM | 10 | 50.14 |
| 11 | – | – | M | NaAl | – | – | MSM | 15 | 42.41 |
| 12 | – | – | M | Cauc | – | – | MSM | 15 | 46.16 |
| 13 | – | – | M | Cauc | + | – | MSM | 8 | 34.65 |
| 14 | – | – | M | Cauc | – | – | MSM | U | 54.35 |
| 15 | – | – | M | Cauc | – | – | U | 23 | 39 |
| 16 | – | – | M | Cauc | – | – | U | 12 | 64.69 |

Notes: All patients HIV-positive; all tissues from globus pallidus; +, present; –, absent.

Abbreviations: HAD, HIV-associated dementia; HIVE, HIV encephalitis; Cauc, Caucasian; Hisp, Hispanic; NaAl, native Alaskan; Y, years; IDU, injection drug abuser; U, unknown; HS, heterosexual; MSM, men who have sex with men; BPR, blood product recipient (blood transfusion); Y, years.

Laser capture microdissection

Slides for laser capture microdissection were lightly stained with Nissl (Arcturus Inc, Mountain View, CA) and dehydrated using an ethanol series followed by xylenes as previously described.³⁹ A Leica laser microdissection microscope (Leica Corporation, Bannockburn, IL) was used for laser capture microdissection using standardized settings and the laser beam precisely followed the neuron's outer membrane. Only neurons with nucleoli were microdissected. No other cells had nucleoli.³⁹

RNA purification

For each case and control tissue, batches of 200 microdissected single cell neurons were suspended in 20 μ L of extraction buffer (Picopure RNA extraction kit, Arcturus Inc) and RNA was extracted. The batches were pooled from multiple cryosections of each tissue. A CapSure-ExtractureSure assembly incubation block with cover (Arcturus Inc) was used to house the tubes. The block was incubated for 30 minutes at 42°C to extract the RNA. The RNA was cryofrozen on dry ice and stored under liquid nitrogen.³⁹

Gene expression analysis

Biotin-labeled cRNA was prepared by linear amplification of the poly (A)+ RNA population within the total RNA sample. Briefly, about 0.5 ng of total RNA (estimated by the number of cryosectioned cells used for RNA isolation) was amplified using a RiboAmp HS kit (Arcturus). After second-strand cDNA synthesis and purification of double-stranded cDNA, in vitro transcription was performed using T7 RNA polymerase in the presence of biotinylated uridine-5'-triphosphate. It must be noted as crucial in the method, that the quantity and quality of the cRNA were assayed by spectrophotometry followed by analysis on an Agilent Bioanalyzer (Agilent Technologies, Colorado Springs, CO). The quality of the cRNA is paramount to ensure nonbiased representation of labeled transcripts containing the complement of the probe sequences deposited on the array.³⁹

Ten micrograms of purified cRNA were fragmented to uniform size and applied to CodeLink Human Whole Genome Bioarrays (GE Healthcare, manufacturer instructions) in hybridization buffer. The specifications, use, and descriptions of the GenUS BIOSYSTEMS CodeLink human CHIPS were as described previously.^{43,44} CodeLink Human Whole Genome arrays comprise approximately 55,000 30-mer probes designed to probe conserved exons across the transcripts of targeted genes. These probes represent annotated, full length, and partial human gene sequences from major public databases.

All fragmented samples were visualized on the Agilent Bioanalyzer to verify complete fragmentation to about 0.1 kb size before array analysis. Arrays were hybridized at 37°C for 18 hours in a shaking incubator, washed in 0.75 \times tris sodium chloride EDTA (TNE) at 46°C for 1 hour, and stained for 30 minutes with Cy5-streptavidin dye conjugate. Arrays were then rinsed, dried, and scanned at 5 μ m resolution with a GenePix™ 4000B scanner (Axon Instruments, according to manufacturer instructions and software).

Statistical analysis

Data production

CodeLink Expression Analysis software (GE Healthcare) was used to process the scanned images from arrays (gridding and feature intensity) and the data generated for each feature on the array were analyzed using GeneSpring software (Agilent Technologies). All control genes and genes that did not pass the quality control metrics of the manufacturer were removed from further analysis.⁴⁴

To compare individual expression values across arrays, raw intensity data from each gene were normalized to the median intensity of the array. Only genes with values greater than background intensity in at least one treatment condition were used for further analysis. Using a ratio interpretation of the data and normalization of each gene to the median intensity across conditions, data were filtered by expression intensity for genes that did not vary by 50% across all samples within the experiment. These unchanging genes were also eliminated from further analysis. This set of present genes was filtered for genes that were within one standard deviation from the mean of replicates. The remaining qualified gene list was queried for genes in treated groups that had ratios >2.0 and <0.5 (two-fold changes) relative to controls. Gene identification based on the GE identifiers was further accomplished using standard websites.⁴³⁻⁴⁶

Statistical methods

The data from this two-way unbalanced cross-classification experiment were analyzed first using analysis of variance to find genes that were statistically significantly different among the four groups at $P \leq 0.005$. Following the analysis of variance, pairwise Student *t*-tests were performed using the mean square error from the analysis of variance to test the simple effects of (HAD⁺ HIVE⁺) versus HIV⁺ control, (HAD⁺ HIVE⁻) versus HIV⁺ control, and (HAD⁻ HIVE⁺) versus HIV⁺ control for each selected gene. These pairwise comparisons were used to find the simple effects giving

rise to the overall statistically significant difference among the four groups. Doing the pairwise comparisons this way is based on Fisher's least significant difference test, which is done only if the overall *F*-test is significant. Using this approach, the pairwise tests do not need to be adjusted for multiple comparisons because the experiment-wise error rate is controlled by the *F*-test.

Pathway analysis

Pathway figures and gene interactions were generated using Gene Network Central PRO.⁴⁷ Pathways were also analyzed using Ariadne Pathways Assist.⁴⁸

Results

Gene expression changes

Sixteen globus pallidus specimens were used as a single experiment (Table 1). The means and standard errors of 197 genes are shown in Table 2. Of these genes, 150 were identified from the GE CodeLink, NCBI, and GeneCards websites. Table 2 also shows the *P* values for overall and simple effects. Three gene expression comparisons made were HAD with HIVE, HAD alone, and HIVE alone, each versus HIV⁺ infected controls. Of the identified genes, HAD with HIVE versus HIV⁺ showed 27 genes upregulated and 30 genes downregulated. HAD alone versus HIV⁺ showed 108 genes upregulated and 22 downregulated. HIVE alone versus HIV⁺ showed 65 genes upregulated and 33 genes downregulated. In all three comparisons, three genes showed simultaneous upregulation and three genes showed simultaneous downregulation. In addition, comparing HAD/HIVE, HAD alone, and HIVE alone versus HIV⁺, the following gene expression shifts, respectively, were one up-up-down, up-down-up, down-up-up, up-down-down, two down-down-up, and three down-up-down (Table 2). The triply regulated genes were as follows: up-up-up, B3GALT1 (galactose transferase), FLJ14167 (potassium inwardly-rectifying channel), and an unidentified gene; up-up-down, NYD-SP26 (development), up-down-up, SLC44A5 (choline transporter-like protein 5), down-up-up, one gene unidentified; down-down-down, HoxD11/HoxD10 (transcription factor, homeobox-regulated development), TBC1D22A (GTPase activator); down-down-up, one gene unidentified, HNRPA1P5 (heterogeneous nuclear ribonucleoprotein A1 pseudogene 5); down-up-down, one gene unidentified, DNAJC3 (chaperone, interferon-induced, double-stranded RNA-activated protein kinase inhibitor), SLAMF6 (SLAM family member 6, CD2 surface receptor, membrane component); and up-down-down, SLC36A4 (amino acid transporter).

Gene expression groups

The identified genes and their functions are shown in Table 3. There are large numbers of functions and gene groups because many genes are in more than one group. The categories of these functions include adhesion cell, adhesion matrix, adhesion membrane, amyloid beta synthesis, amyloid beta precursor processing, apoptosis, apoptosis caspase activator, binding metal ion, binding nucleotide, binding GTP, binding heparin, binding phosphatidyl inositol, binding DNA, binding RNA, binding double-stranded RNA, biosynthesis, biosynthesis amino acid, channel complex Ca, cell cycle, cell differentiation, cell division, cell division arrest, channel potassium inward rectifier, chaperone, chaperone cochaperone, chromatin regulation assembly, chromatin regulation repair, collagen, cytokine, cytokine growth factor, cytoskeleton, microtubule, development nervous system, developmental protein, Alzheimer's disease, diGeorge syndrome, Huntington's disease, DNA polymerase, DNA repair, endoplasmic reticulum, endocytosis, esterase thio-acyl-CoA, exocytosis, factor viability, glutamate polyglutamylase, glutamyl transferase, glycan N-glycan processing, glycosylation N-linked, glycosylation O-linked, Golgi stack apparatus, Golgi clathrin coat, Golgi vesicle, G protein cycle, GTPase, heat shock, hydrogenase-like protein iron only, interferon induced pathway, lamin prelamin recognition factor, lamin prelamin binding protein, lamina nuclear, lipid biosynthesis, lipid phosphatidyl serine biosynthesis, lipid phospholipid biosynthesis, matrix cell, matrix extracellular, metabolism, mitochondrial electron transport, mitochondrial function, mitochondrial membrane, mitochondrial metalloproteinase protein, mitochondrial ribosomal protein, mitochondrial ribosome, motility cell, movement intracellular, mRNA transport, multinet network protein, multinet network detection protein or RNA, nucleopore, nucleopore mRNA transport, oligosaccharide biosynthesis, oligosaccharide hydrolase, oncogene, oxireductase, oxidase, peptidase, peptide cross-linking, perception sensory olfactory, perception sensory visual, proliferation cell, protease, protease endoprotease, protein biosynthesis, protein kinase, protein phosphatase, proteinase metallo, pseudogene, receptor AMPA, receptor cytokine, receptor cytokine ligand, receptor interacting protein, receptor NMDA, receptor glutamate, receptor glycophorin, receptor metabotropic, receptor nuclear interacting, receptor MHC class I, receptor MHC class I antigen presentation, ribosome, ribosome subunits, ribosome assembly, ribosome protein, ribosome protein synthesis, ribosome translation factor, ribosome translation initiation factor, RNA heterogeneous nucleoprotein, RNA noncoding, RNA miRNA, signaling ras

Table 2 Significant gene expression by one-way analysis of variance ($P < 0.005$)

| Identifier | Probe | HAD + HIVE | | HAD | | HIVE | | Control | | Between groups | | HAD + HIVE/ control | | HAD/ control | | HIVE/ control | |
|------------|----------|------------|------|-------|------|-------|------|---------|------|----------------|----|------------------------|---|------------------------|------------------------|------------------------|---|
| | | Mean | SE | Mean | SE | Mean | SE | Mean | SE | Mean | SE | P | P | P | P | P | P |
| 1244 | GE472453 | 0.84 | 0.15 | 0.58 | 0.21 | 1.97 | 0.06 | 0.50 | 0.12 | 0.000700 | | | | | | Up × 4 0.000084 | |
| 1420 | GE474010 | 0.84 | 0.11 | 0.16 | 0.08 | 2.51 | 0.92 | 0.45 | 0.19 | 0.001871 | | | | | | Up × 5 0.000411 | |
| 2021 | GE479725 | 0.47 | 0.08 | 0.49 | 0.28 | 1.55 | 0.27 | 0.35 | 0.07 | 0.001153 | | | | | | Up × 4 0.000135 | |
| 2172 | GE481051 | 0.38 | 0.22 | 0.47 | 0.03 | 1.95 | 0.16 | 0.67 | 0.15 | 0.001634 | | | | | | Up × 3 0.000805 | |
| 2573 | GE484741 | 0.33 | 0.11 | 2.06 | 0.40 | 1.09 | 0.32 | 0.69 | 0.17 | 0.001987 | | | | Up × 3 0.000946 | | | |
| 2642 | GE485413 | 1.96 | 0.59 | 4.86 | 0.43 | 1.68 | 0.38 | 3.62 | 0.26 | 0.001570 | | Down × 0.5 0.008019 | | | Down 0.5 0.013596 | | |
| 2830 | GE487382 | 1.13 | 0.40 | 4.42 | 0.45 | 0.69 | 0.19 | 1.38 | 0.37 | 0.000711 | | | | | | | |
| 3159 | GE490114 | 1.10 | 0.07 | 1.30 | 0.15 | -0.15 | 0.30 | 0.73 | 0.13 | 0.000793 | | | | | | Down × 0.2 0.002706 | |
| 3289 | GE491184 | 0.22 | 0.07 | 0.70 | 0.03 | 1.74 | 0.08 | 0.88 | 0.17 | 0.001857 | | Down × 0.3 0.008868 | | | Up × 2 0.007956 | | |
| 3577 | GE493533 | 1.37 | 0.26 | 4.09 | 0.63 | 1.01 | 0.08 | 1.26 | 0.25 | 0.000344 | | | | | | | |
| 4180 | GE498722 | 6.75 | 0.46 | 11.19 | 0.89 | 6.60 | 0.74 | 7.35 | 0.45 | 0.001320 | | | | Up × 3.2 0.000070 | | | |
| 4272 | GE499400 | 1.25 | 0.16 | 0.07 | 0.19 | -0.11 | 0.05 | 0.86 | 0.15 | 0.000980 | | | | Up × 1.5 0.000544 | | | |
| 4285 | GE499526 | 0.82 | 0.19 | 0.79 | 0.15 | 3.68 | 1.50 | 0.61 | 0.19 | 0.001679 | | | | Down × 0.1 0.006076 | | Down × 0.1 0.004390 | |
| 4361 | GE500216 | 0.67 | 0.44 | 0.94 | 0.59 | 7.47 | 3.64 | 0.78 | 0.17 | 0.001535 | | | | | | Up × 6 0.000226 | |
| 4711 | GE503208 | 0.26 | 0.24 | 0.41 | 0.06 | 1.64 | 0.22 | 0.95 | 0.09 | 0.000930 | | Down × 0.4 0.026397 | | | Down × 0.4 0.000262 | | |
| 5179 | GE507524 | 5.82 | 0.33 | 12.17 | 1.22 | 6.98 | 1.16 | 7.33 | 0.46 | 0.000326 | | | | Up × 1.7 0.000222 | | Up × 1.7 0.017107 | |
| 5674 | GE512134 | 1.09 | 0.25 | 0.51 | 0.20 | 2.33 | 0.01 | 0.52 | 0.12 | 0.000310 | | Up × 2 0.028641 | | | Up × 4.5 0.000049 | | |
| 6001 | GE515097 | 1.71 | 0.25 | 4.23 | 0.45 | 1.26 | 0.08 | 1.96 | 0.25 | 0.000385 | | | | | | | |
| 6005 | GE515151 | 1.65 | 0.35 | 4.45 | 0.95 | 0.59 | 0.14 | 1.79 | 0.22 | 0.001354 | | | | | | | |

| | | | | | | | | | | | |
|-------|----------|-------|------|------|------|------|------|------|------|----------|------------------------|
| 6006 | GE515161 | 0.98 | 0.17 | 3.29 | 0.57 | 0.07 | 0.10 | 0.92 | 0.33 | 0.001728 | Up × 3.6 0.000681 |
| 6064 | GE515618 | 0.86 | 0.04 | 0.33 | 0.18 | 0.93 | 0.24 | 1.46 | 0.15 | 0.001903 | Down × 0.2 0.000273 |
| 6121 | GE516084 | 1.01 | 0.26 | 0.82 | 0.23 | 2.59 | 0.13 | 0.27 | 0.09 | 0.000029 | Down × 0.6 0.012379 |
| 6204 | GE516830 | 1.52 | 0.49 | 4.18 | 0.52 | 1.49 | 0.33 | 1.63 | 0.15 | 0.000731 | Up × 3 0.043188 |
| 6530 | GE519581 | 0.80 | 0.08 | 0.11 | 0.10 | 1.25 | 0.01 | 0.76 | 0.12 | 0.001515 | Up × 2.6 0.000171 |
| 6580 | GE519998 | 1.72 | 0.37 | 3.63 | 0.31 | 0.43 | 0.04 | 1.66 | 0.19 | 0.000224 | Down × 0.1 0.002112 |
| 7181 | GE525253 | 0.79 | 0.27 | 3.78 | 0.57 | 0.67 | 0.61 | 1.16 | 0.32 | 0.001331 | Up × 2.2 0.000235 |
| 7348 | GE526744 | 1.17 | 0.11 | 1.41 | 0.24 | 0.44 | 0.08 | 0.49 | 0.11 | 0.001054 | Up × 3.3 0.000519 |
| 7400 | GE527127 | 2.26 | 0.48 | 4.77 | 0.35 | 1.59 | 0.47 | 2.65 | 0.25 | 0.001536 | Up × 3 0.000538 |
| 7585 | GE528706 | 2.73 | 0.43 | 4.51 | 0.40 | 1.03 | 0.13 | 3.48 | 0.30 | 0.001906 | Up × 1.8 0.001290 |
| 7999 | GE53107 | 0.80 | 0.23 | 0.97 | 0.02 | 5.69 | 2.15 | 1.18 | 0.29 | 0.000800 | Down × 0.3 0.001693 |
| 8018 | GE53116 | 1.69 | 0.25 | 5.07 | 0.76 | 0.95 | 0.15 | 1.49 | 0.45 | 0.001297 | Up × 4.8 0.000188 |
| 8334 | GE53271 | 1.19 | 0.16 | 2.30 | 0.11 | 1.34 | 0.08 | 1.31 | 0.13 | 0.001547 | Up × 3.4 0.000290 |
| 9079 | GE536414 | 0.92 | 0.17 | 2.87 | 0.18 | 1.67 | 0.00 | 1.74 | 0.18 | 0.000344 | Up × 1.8 0.000432 |
| 9181 | GE53692 | 0.90 | 0.37 | 2.63 | 0.62 | 5.06 | 0.56 | 1.81 | 0.37 | 0.001577 | Up × 1.6 0.001424 |
| 9536 | GE538621 | 1.56 | 0.05 | 1.08 | 0.05 | 0.50 | 0.24 | 0.36 | 0.15 | 0.000240 | Up × 2.8 0.000869 |
| 9814 | GE54005 | 1.32 | 0.15 | 2.45 | 0.40 | 0.98 | 0.23 | 0.82 | 0.15 | 0.001327 | Up × 3 0.004543 |
| 10760 | GE54509 | -0.17 | 0.13 | 0.78 | 0.15 | 0.54 | 0.13 | 1.05 | 0.17 | 0.001764 | Up × 3 0.000157 |
| 11409 | GE548504 | 0.12 | 0.15 | 1.00 | 0.21 | 2.08 | 0.06 | 0.82 | 0.17 | 0.000776 | Down × 0.2 0.000203 |
| 11522 | GE549123 | 0.69 | 0.10 | 0.16 | 0.19 | 1.58 | 0.60 | 0.21 | 0.05 | 0.000703 | Down × 0.1 0.014773 |
| 11545 | GE549241 | 0.82 | 0.24 | 0.10 | 0.07 | 1.78 | 0.14 | 0.92 | 0.10 | 0.000684 | Up × 8 0.000138 |
| | | | | | | | | | | | Up × 2 0.004963 |

(Continued)

Table 2 (Continued)

| Identifier | Probe | HAD + HIVE | | HAD | | HIVE | | Control | | Between groups | | HAD + HIVE/control | | HAD/control | | HIVE/control | |
|------------|----------|------------|------|------|------|-------|------|---------|------|----------------|------------|--------------------|------------|-------------|---|--------------|------------|
| | | Mean | SE | Mean | SE | Mean | SE | Mean | SE | P | P | P | P | P | P | | |
| 11877 | GE55094 | 1.46 | 0.34 | 3.82 | 0.49 | 1.27 | 0.26 | 1.08 | 0.25 | 0.000668 | | Up × 3.5 | 0.000087 | | | | |
| 12206 | GE55267 | 1.17 | 0.47 | 2.82 | 0.59 | 1.72 | 0.11 | 3.54 | 0.21 | 0.001579 | Down × 0.3 | 0.000267 | | | | | Down × 0.4 |
| 12366 | GE553476 | 2.88 | 0.99 | 9.99 | 1.81 | 1.84 | 1.15 | 3.96 | 0.61 | 0.001746 | | | Up × 2.5 | | | | 0.009922 |
| 12612 | GE554808 | 0.73 | 0.24 | 4.16 | 0.96 | 0.64 | 0.16 | 1.00 | 0.30 | 0.001054 | | | 0.001028 | | | | |
| 12915 | GE556336 | 0.84 | 0.27 | 3.90 | 0.73 | 0.78 | 0.07 | 0.93 | 0.33 | 0.001167 | | | Up × 4.2 | | | | |
| 13339 | GE558357 | 0.14 | 0.19 | 0.89 | 0.09 | 0.26 | 0.07 | 0.92 | 0.08 | 0.001035 | Down × 0.1 | | 0.000262 | | | | Down × 0.3 |
| 14400 | GE563896 | 0.83 | 0.16 | 1.03 | 0.11 | 1.13 | 0.38 | 0.07 | 0.13 | 0.001971 | 0.000340 | | | Up × 15 | | | 0.006495 |
| 14504 | GE564415 | 0.60 | 0.23 | 0.56 | 0.15 | 2.61 | 0.47 | 1.20 | 0.19 | 0.001793 | Up × 12 | | | 0.001706 | | | Up × 16 |
| 14612 | GE56503 | 1.15 | 0.09 | 3.73 | 1.00 | 0.86 | 0.34 | 0.46 | 0.14 | 0.000471 | 0.004071 | | | | | | 0.002432 |
| 14700 | GE565524 | 0.06 | 0.17 | 1.29 | 0.14 | 1.31 | 0.38 | 0.30 | 0.13 | 0.000847 | | | Up × 8 | | | | Up × 2.2 |
| 14740 | GE565731 | 0.79 | 0.26 | 0.98 | 0.18 | 2.58 | 0.42 | 0.58 | 0.12 | 0.000453 | | | 0.001557 | | | | 0.003328 |
| 14756 | GE56583 | 0.84 | 0.04 | 1.58 | 0.08 | 0.12 | 0.20 | 0.97 | 0.15 | 0.001455 | | | | | | | Up × 4.4 |
| 14820 | GE566190 | 0.52 | 0.12 | 2.26 | 0.36 | -0.28 | 0.32 | 0.92 | 0.23 | 0.000966 | | | Up × 1.6 | | | | 0.000049 |
| 15762 | GE57137 | 1.14 | 0.34 | 3.70 | 0.39 | 1.49 | 0.27 | 1.71 | 0.27 | 0.001871 | | | Down × 0.1 | | | | Down × 0.3 |
| 16114 | GE57325 | 0.72 | 0.07 | 0.45 | 0.38 | 2.74 | 0.98 | 0.61 | 0.13 | 0.001970 | | | 0.01588 | | | | 0.003885 |
| 16469 | GE57516 | 1.10 | 0.21 | 0.54 | 0.08 | -1.38 | 0.29 | 0.59 | 0.14 | 0.000036 | Up × 1.9 | | | Up × 2.5 | | | Down × 0.3 |
| 16547 | GE57557 | 1.31 | 0.36 | 3.88 | 0.77 | 1.37 | 0.44 | 1.01 | 0.17 | 0.000909 | 0.044226 | | | 0.002864 | | | 0.014591 |
| 16823 | GE576963 | 0.85 | 0.23 | 1.19 | 0.11 | -0.74 | 0.39 | 1.15 | 0.12 | 0.000238 | | | Up × 2.2 | | | | Up × 4.5 |
| 17180 | GE57883 | 0.83 | 0.24 | 1.27 | 0.30 | 2.06 | 0.08 | 1.92 | 0.09 | 0.001476 | Down × 0.4 | | 0.001043 | | | | 0.000363 |
| | | | | | | | | | | | 0.000391 | | | Up × 3.8 | | | Up × 2.3 |
| | | | | | | | | | | | | | | 0.000120 | | | 0.000018 |
| | | | | | | | | | | | | | | | | | Down × 0.6 |
| | | | | | | | | | | | | | | | | | 0.000033 |
| | | | | | | | | | | | | | | | | | 0.020768 |

| | | | | | | | | | | | |
|-------|----------|-------|------|-------|------|-------|------|-------|------|----------|------------------------|
| 17447 | GE58018 | 0.94 | 0.30 | 1.00 | 0.15 | 4.28 | 1.64 | 1.60 | 0.07 | 0.001327 | Up × 2.7 0.000814 |
| 17879 | GE582514 | 0.82 | 0.06 | 0.71 | 0.09 | 1.52 | 0.10 | 0.73 | 0.06 | 0.000148 | Up × 2.1 0.000021 |
| 17884 | GE58255 | 1.40 | 0.17 | 2.52 | 0.27 | 0.73 | 0.10 | 1.81 | 0.12 | 0.000540 | Down × 0.4 0.002016 |
| 17947 | GE58287 | 0.87 | 0.15 | 0.87 | 0.06 | 2.10 | 0.25 | 0.79 | 0.14 | 0.001773 | Up × 2.7 0.000241 |
| 17974 | GE583033 | 1.74 | 0.39 | 4.91 | 1.00 | 1.79 | 0.03 | 1.65 | 0.22 | 0.001220 | Up × 1.4 0.010039 |
| 18252 | GE58460 | 3.79 | 0.69 | 14.65 | 1.81 | 2.42 | 0.17 | 4.73 | 1.48 | 0.001689 | Up × 3 0.000208 |
| 18384 | GE585314 | 9.70 | 4.79 | 21.25 | 3.36 | 4.38 | 2.41 | 1.02 | 0.34 | 0.001385 | Up × 3.1 0.000624 |
| 18390 | GE58535 | 2.51 | 0.55 | 6.87 | 0.91 | 2.37 | 0.79 | 2.81 | 0.35 | 0.000747 | Up 20.8 0.000173 |
| 18598 | GE58654 | 0.53 | 0.14 | 1.50 | 0.11 | 0.78 | 0.07 | 0.88 | 0.10 | 0.001930 | Up × 2.4 0.000210 |
| 18634 | GE586724 | 6.33 | 0.88 | 10.61 | 0.70 | 3.35 | 1.32 | 5.86 | 0.48 | 0.000747 | Up × 1.7 0.003111 |
| 18765 | GE587496 | 4.55 | 0.54 | 7.64 | 0.60 | 1.30 | 1.39 | 4.99 | 0.48 | 0.001309 | Up × 1.8 0.000491 |
| 19105 | GE58946 | 0.30 | 0.09 | 1.39 | 0.08 | 1.30 | 0.64 | 0.43 | 0.10 | 0.001714 | Up × 1.53 0.010656 |
| 19877 | GE593831 | 9.97 | 0.86 | 17.17 | 2.23 | 8.78 | 0.70 | 10.32 | 0.63 | 0.001914 | Down × 0.3 0.003449 |
| 20353 | GE596515 | 0.66 | 0.14 | 0.98 | 0.33 | 3.76 | 1.57 | 0.83 | 0.13 | 0.001767 | Up × 3 0.007821 |
| 20752 | GE59877 | 1.11 | 0.10 | 0.94 | 0.20 | 0.60 | 0.16 | 0.14 | 0.11 | 0.000576 | Up × 4.5 0.000333 |
| 20798 | GE599024 | 1.46 | 0.12 | 2.46 | 0.17 | 1.20 | 0.20 | 0.79 | 0.21 | 0.001002 | Up × 6.7 0.001325 |
| 22253 | GE609375 | 0.88 | 0.12 | 2.18 | 0.52 | 0.19 | 0.04 | 0.63 | 0.09 | 0.000591 | Up × 3.1 0.00110 |
| 22901 | GE613705 | 34.79 | 3.54 | 59.02 | 4.98 | 26.20 | 1.77 | 33.46 | 2.98 | 0.001113 | Up × 3.5 0.000168 |
| 22971 | GE61413 | 0.50 | 0.20 | -0.08 | 0.10 | 1.06 | 0.07 | 0.92 | 0.11 | 0.001731 | Up × 1.8 0.000351 |
| 23200 | GE61539 | 0.66 | 0.18 | 1.84 | 0.15 | -0.01 | 0.01 | 0.51 | 0.20 | 0.001552 | Down × 0.1 0.000398 |
| 23526 | GE617302 | 4.95 | 0.46 | 11.10 | 1.36 | 4.38 | 0.78 | 6.26 | 0.60 | 0.000934 | Up × 3.6 0.000614 |
| | | | | | | | | | | 0.000818 | Up × 1.8 0.000818 |

(Continued)

Table 2 (Continued)

| Identifier | Probe | HAD + HIVE | | HAD | | HIVE | | Control | | Between groups | HAD + HIVE/control | | HAD/control | | HIVE/control | |
|------------|----------|------------|------|-------|------|------|------|---------|------|----------------|------------------------|----------------------|-------------|------------------------|--------------|-------------------------|
| | | Mean | SE | Mean | SE | Mean | SE | Mean | SE | | P | P | P | P | P | P |
| 24052 | GE620526 | 1.17 | 0.14 | 4.25 | 1.23 | 1.19 | 0.31 | 1.08 | 0.13 | 0.001876 | | Up × 4 0.000327 | | | | |
| 24282 | GE62190 | 1.06 | 0.26 | 2.46 | 0.35 | 0.47 | 0.11 | 0.86 | 0.14 | 0.000841 | | Up × 2.9 0.000221 | | | | |
| 24756 | GE624691 | 2.22 | 0.31 | 3.35 | 0.72 | 0.13 | 0.43 | 2.68 | 0.23 | 0.001680 | | | | | | Down × 0.26 0.000538 |
| 24982 | GE626074 | 1.28 | 0.18 | 0.50 | 0.15 | 1.91 | 0.29 | 0.74 | 0.09 | 0.000561 | Up × 1.7 0.012227 | | | | | Up × 2.6 0.000323 |
| 25091 | GE62673 | 0.36 | 0.12 | 0.38 | 0.20 | 1.76 | 0.20 | 0.62 | 0.07 | 0.000109 | | | | | | Up × 2.8 0.000068 |
| 25103 | GE62681 | 1.62 | 0.23 | 0.23 | 0.23 | 0.39 | 0.08 | 0.69 | 0.11 | 0.000997 | Up × 2.3 0.001279 | | | | | |
| 25830 | GE631063 | 1.56 | 0.26 | 4.38 | 0.39 | 1.51 | 0.68 | 2.01 | 0.30 | 0.001123 | | | | Up × 2.2 0.000536 | | |
| 26034 | GE63224 | 0.90 | 0.09 | 0.36 | 0.19 | 0.10 | 0.13 | 0.68 | 0.05 | 0.001590 | | | | Down × 0.5 0.034863 | | Down × 0.1 0.002654 |
| 26491 | GE636205 | 1.65 | 0.81 | 7.62 | 1.57 | 2.67 | 1.97 | 0.60 | 0.15 | 0.000358 | | | | Up × 12.7 0.000040 | | |
| 26776 | GE644246 | 0.68 | 0.14 | 0.59 | 0.07 | 4.58 | 2.02 | 0.86 | 0.18 | 0.001082 | | | | | | Up × 5.3 0.000243 |
| 26936 | GE648477 | 0.72 | 0.28 | 3.55 | 0.64 | 0.63 | 0.49 | 1.79 | 0.27 | 0.001761 | Down × 0.4 0.044409 | | | | | |
| 27283 | GE655391 | 0.88 | 0.25 | 0.62 | 0.21 | 1.99 | 0.02 | 0.65 | 0.06 | 0.001297 | | | | Up × 2 0.005878 | | |
| 27415 | GE657626 | 1.45 | 1.00 | 10.05 | 1.64 | 3.32 | 2.11 | 1.46 | 0.16 | 0.000092 | | | | Up × 6.9 0.000015 | | Up × 3.1 0.000188 |
| 27568 | GE660354 | 2.84 | 2.15 | 16.96 | 5.10 | 3.04 | 3.68 | 0.83 | 0.26 | 0.001685 | | | | Up × 20.4 0.000217 | | |
| 28284 | GE674173 | 0.49 | 0.15 | 0.31 | 0.21 | 2.58 | 0.29 | 0.71 | 0.13 | 0.000054 | | | | | | Up × 3.6 0.000024 |
| 28369 | GE675994 | 1.20 | 0.44 | 4.05 | 0.49 | 0.99 | 0.32 | 3.97 | 0.48 | 0.001972 | Down × 0.3 0.001275 | | | | | Down × 0.2 0.004302 |
| 28502 | GE678706 | 0.66 | 0.26 | 3.32 | 0.59 | 0.27 | 0.30 | 0.92 | 0.15 | 0.000126 | | | | Up × 3.6 0.000058 | | |
| 28508 | GE678803 | 1.13 | 0.52 | 1.20 | 0.35 | 4.44 | 0.20 | 1.07 | 0.14 | 0.000161 | | | | | | Up × 4.1 0.000023 |
| 28991 | GE687963 | 8.69 | 0.83 | 17.80 | 2.65 | 7.22 | 0.58 | 8.82 | 0.76 | 0.000816 | | | | | | |
| 29168 | GE691505 | 0.82 | 0.09 | 0.45 | 0.18 | 1.59 | 0.04 | 0.45 | 0.12 | 0.001285 | | | | | | Up × 3.5 0.000211 |

| | | | | | | | | | | | |
|-------|----------|-------|------|--------|-------|-------|------|-------|------|----------|------------------------|
| 29917 | GE705764 | 0.41 | 0.15 | 0.85 | 0.41 | 4.34 | 2.10 | 0.62 | 0.14 | 0.002083 | Up × 7 0.000386 |
| 30069 | GE708617 | 0.73 | 0.15 | 0.58 | 0.14 | 2.12 | 0.07 | 0.49 | 0.14 | 0.000308 | Up × 4.3 0.000035 |
| 30107 | GE709371 | 1.49 | 0.28 | 0.42 | 0.20 | 1.67 | 0.39 | 0.41 | 0.12 | 0.001897 | Up × 4.1 0.002802 |
| 30167 | GE710687 | 0.75 | 0.24 | 3.37 | 0.61 | 0.57 | 0.47 | 1.23 | 0.26 | 0.001563 | Up × 2.7 0.000954 |
| 31250 | GE726916 | 0.75 | 0.27 | 0.46 | 0.22 | -0.51 | 0.32 | 1.31 | 0.09 | 0.000457 | Down × 0.4 0.000065 |
| 31353 | GE728396 | 1.92 | 0.21 | 4.77 | 0.95 | 1.03 | 0.03 | 1.31 | 0.30 | 0.000741 | Up × 3.6 0.001461 |
| 31404 | GE729136 | 0.91 | 0.06 | 2.52 | 0.27 | 1.38 | 0.14 | 1.10 | 0.21 | 0.001999 | Down × 0.6 0.036268 |
| 31632 | GE732434 | -0.14 | 0.26 | 1.28 | 0.07 | 1.01 | 0.26 | 0.90 | 0.11 | 0.000750 | Up × 2.3 0.000533 |
| 32204 | GE740641 | 0.66 | 0.16 | 0.44 | 0.09 | 1.69 | 0.25 | 0.83 | 0.10 | 0.001980 | Down × 0.2 0.000510 |
| 32318 | GE742294 | 37.00 | 3.06 | 112.77 | 24.65 | 28.60 | 4.16 | 40.01 | 5.04 | 0.000741 | Up × 2 0.001889 |
| 32836 | GE749435 | 0.82 | 0.19 | 1.53 | 0.13 | 0.42 | 0.11 | 0.61 | 0.08 | 0.001171 | Up × 2.8 0.000208 |
| 33031 | GE752199 | 1.23 | 0.11 | 3.73 | 0.15 | 1.34 | 0.08 | 1.87 | 0.34 | 0.001822 | Up × 2.5 0.000262 |
| 33193 | GE754378 | 0.48 | 0.10 | 0.82 | 0.22 | 1.78 | 0.01 | 0.64 | 0.12 | 0.001454 | Up × 2 0.001571 |
| 33293 | GE755614 | 1.15 | 0.28 | 2.68 | 0.38 | 0.56 | 0.40 | 1.00 | 0.11 | 0.000748 | Up × 2.8 0.000362 |
| 33722 | GE762426 | 1.07 | 0.37 | 4.09 | 0.34 | 1.24 | 0.17 | 1.69 | 0.36 | 0.001841 | Up × 2.7 0.000210 |
| 33921 | GE765425 | 0.57 | 0.23 | 0.97 | 0.37 | 2.62 | 0.25 | 0.85 | 0.11 | 0.000699 | Up × 2.4 0.001030 |
| 34083 | GE767593 | 1.27 | 0.15 | 2.83 | 0.43 | 0.43 | 0.14 | 1.02 | 0.23 | 0.001286 | Up × 3.1 0.000190 |
| 34195 | GE769111 | 0.48 | 0.16 | 0.75 | 0.21 | 2.09 | 0.05 | 0.91 | 0.15 | 0.001954 | Up × 2.8 0.000425 |
| 34221 | GE769398 | 4.54 | 0.37 | 8.74 | 1.20 | 5.05 | 0.30 | 4.71 | 0.37 | 0.001547 | Up × 1.9 0.000312 |
| 34239 | GE769588 | 1.06 | 0.17 | 0.21 | 0.12 | -0.30 | 0.22 | 0.54 | 0.09 | 0.000504 | Up × 2 0.010348 |

(Continued)

Table 2 (Continued)

| Identifier | Probe | HAD + HIVE | | HAD | | HIVE | | Control | | Between groups | | HAD + HIVE/control | | HAD/control | | HIVE/control | |
|------------|----------|------------|------|------|------|-------|------|---------|------|----------------|------------|--------------------|------------|-------------|------------|--------------|----------|
| | | Mean | SE | Mean | SE | Mean | SE | Mean | SE | Mean | SE | P | P | P | P | P | P |
| 34398 | GE772048 | 0.34 | 0.38 | 1.86 | 0.42 | 2.34 | 0.35 | 0.61 | 0.11 | 0.001556 | | Up × 3 | 0.005964 | Up × 3.8 | 0.001863 | | |
| 35620 | GE78986 | 0.06 | 0.26 | 0.75 | 0.16 | 1.38 | 0.10 | 0.96 | 0.08 | 0.001613 | Down × 0.1 | | | | | | |
| 35666 | GE790167 | 0.98 | 0.13 | 0.44 | 0.09 | 2.45 | 0.64 | 0.76 | 0.10 | 0.000205 | | | | Up × 3.2 | 0.000054 | | |
| 35761 | GE79076 | 0.51 | 0.40 | 3.53 | 0.21 | 1.38 | 0.16 | 1.68 | 0.26 | 0.000473 | Down × 0.3 | 0.013393 | Up × 2.1 | | | | |
| 36075 | GE79260 | 0.72 | 0.12 | 0.41 | 0.15 | 2.04 | 0.57 | 0.80 | 0.11 | 0.001484 | | | 0.001299 | | | Up × 2.6 | 0.000754 |
| 36101 | GE79273 | 1.17 | 0.15 | 0.28 | 0.28 | 0.81 | 0.11 | 0.39 | 0.05 | 0.002042 | Up × 3 | 0.000610 | | | | Up × 23.9 | 0.000045 |
| 36278 | GE79382 | 1.41 | 0.25 | 2.75 | 1.40 | 8.59 | 3.08 | 0.36 | 0.18 | 0.000408 | | | | | | | |
| 36337 | GE79415 | 1.38 | 0.17 | 0.53 | 0.35 | -0.25 | 0.33 | 0.26 | 0.11 | 0.001504 | Up × 5.3 | 0.000716 | | | | | |
| 36363 | GE794289 | 0.52 | 0.05 | 0.32 | 0.09 | 1.69 | 0.02 | 0.67 | 0.11 | 0.000122 | | | Down × 0.5 | 0.042183 | Up × 2.5 | 0.000090 | |
| 36885 | GE797280 | 0.47 | 0.25 | 2.39 | 0.29 | 1.27 | 0.11 | 0.76 | 0.14 | 0.000298 | | | Up × 3.1 | 0.000105 | | | |
| 36949 | GE79764 | 0.72 | 0.10 | 2.07 | 0.29 | 0.75 | 0.42 | 0.52 | 0.11 | 0.000296 | | | Up × 4 | 0.000035 | | | |
| 36990 | GE79788 | 7.69 | 0.28 | 6.21 | 0.51 | 16.04 | 3.72 | 8.32 | 0.76 | 0.001578 | | | | | Up × 1.9 | 0.000688 | |
| 37273 | GE799491 | 1.14 | 0.19 | 0.93 | 0.08 | 0.98 | 0.39 | 0.28 | 0.07 | 0.002032 | Up × 4 | 0.000510 | Up × 3.3 | 0.007183 | Up × 3.5 | 0.011246 | |
| 38007 | GE80398 | 3.38 | 0.82 | 7.92 | 1.28 | 2.12 | 1.96 | 1.81 | 0.36 | 0.001087 | | | Up × 4.4 | 0.000136 | | | |
| 38049 | GE804261 | 1.07 | 0.08 | 0.55 | 0.06 | 0.11 | 0.09 | 0.59 | 0.07 | 0.000143 | Up × 1.8 | 0.000503 | | | Down × 0.2 | 0.003676 | |
| 38070 | GE804386 | 1.77 | 0.26 | 5.95 | 1.20 | 2.43 | 0.15 | 2.16 | 0.25 | 0.000569 | | | Up × 2.8 | 0.000142 | | | |
| 38729 | GE808417 | 1.26 | 0.13 | 0.32 | 0.08 | 0.37 | 0.26 | 0.78 | 0.11 | 0.001945 | Up × 1.6 | 0.013220 | Down × 0.4 | 0.024128 | | | |
| 38739 | GE80847 | 2.88 | 0.87 | 7.00 | 0.44 | 4.61 | 0.74 | 3.87 | 0.21 | 0.001721 | | | Up × 1.8 | 0.001012 | | | |
| 38813 | GE80890 | 0.76 | 0.09 | 0.47 | 0.07 | 3.06 | 1.16 | 0.78 | 0.09 | 0.000529 | | | | | Up × 4 | 0.000130 | |

| | | | | | | | | | | | | |
|-------|----------|------|------|-------|------|-------|------|-------|------|----------|------------------------|------------------------|
| 38882 | GE809301 | 0.82 | 0.21 | 1.45 | 0.18 | 0.35 | 0.12 | 1.52 | 0.10 | 0.001306 | Down × 0.5 0.003543 | Down × 0.2 0.000511 |
| 39376 | GE812224 | 1.13 | 0.27 | 10.48 | 1.77 | 0.47 | 0.19 | 1.28 | 0.57 | 0.000016 | Up × 8.2 0.000004 | Up × 3.8 0.004217 |
| 39516 | GE813126 | 0.67 | 0.12 | -0.32 | 0.42 | 1.46 | 0.40 | 0.38 | 0.07 | 0.001864 | Down × 0.8 0.019764 | Down × 0.1 0.003760 |
| 39690 | GE81418 | 1.16 | 0.21 | 0.17 | 0.07 | -0.11 | 0.26 | 0.85 | 0.12 | 0.001704 | Down × 0.2 0.012339 | Up × 2.6 0.000045 |
| 39743 | GE81449 | 0.84 | 0.15 | 0.48 | 0.20 | 2.03 | 0.11 | 0.77 | 0.08 | 0.000133 | Down × 0.1 0.022083 | Up × 2.8 0.000234 |
| 40333 | GE81822 | 0.78 | 0.19 | 0.54 | 0.08 | 2.56 | 0.52 | 0.91 | 0.14 | 0.000622 | Down × 0.2 0.001056 | Up × 3.1 0.000409 |
| 40551 | GE819522 | 0.71 | 0.26 | 0.03 | 0.14 | 2.24 | 0.42 | 0.73 | 0.11 | 0.000413 | Down × 0.1 0.022083 | Up × 2 0.001571 |
| 40642 | GE820114 | 1.23 | 0.12 | 0.13 | 0.16 | 0.58 | 0.12 | 0.95 | 0.12 | 0.001288 | Down × 0.2 0.000081 | Down × 0.4 0.001387 |
| 40690 | GE820397 | 0.26 | 0.14 | 0.86 | 0.12 | 1.86 | 0.33 | 0.91 | 0.11 | 0.000378 | Down × 0.4 0.002870 | Down × 0.6 0.026178 |
| 41169 | GE82307 | 8.51 | 0.56 | 15.50 | 1.93 | 4.70 | 0.07 | 13.41 | 1.16 | 0.001551 | Up × 10.8 0.000232 | Up × 3.5 0.000531 |
| 41665 | GE82602 | 0.19 | 0.08 | 0.41 | 0.12 | 0.52 | 0.17 | 0.94 | 0.08 | 0.000503 | Up × 3.2 0.001088 | Down × 0.4 0.001387 |
| 41868 | GE82723 | 1.83 | 0.66 | 15.01 | 5.12 | 1.58 | 0.40 | 1.39 | 0.53 | 0.001319 | Down × 0.4 0.002870 | Down × 0.6 0.026178 |
| 41968 | GE82785 | 0.20 | 0.22 | 0.35 | 0.06 | 2.72 | 0.63 | 0.78 | 0.21 | 0.000683 | Up × 10.8 0.000232 | Up × 3.5 0.000531 |
| 42072 | GE82842 | 0.26 | 0.35 | 3.59 | 0.76 | 0.95 | 0.96 | 1.12 | 0.20 | 0.001575 | Up × 3.2 0.001088 | Down × 0.4 0.001387 |
| 42550 | GE831160 | 0.75 | 0.11 | 2.35 | 0.33 | 0.77 | 0.30 | 1.08 | 0.13 | 0.000489 | Up × 2.2 0.000326 | Down × 0.4 0.001387 |
| 42718 | GE832143 | 1.19 | 0.07 | 1.01 | 0.06 | 0.39 | 0.13 | 0.42 | 0.13 | 0.001362 | Up × 2.4 0.000326 | Down × 0.4 0.001387 |
| 42726 | GE83218 | 0.98 | 0.06 | 2.36 | 0.51 | 0.57 | 0.05 | 0.54 | 0.15 | 0.000749 | Up × 2.4 0.000326 | Down × 0.4 0.001387 |
| 42766 | GE832421 | 1.09 | 0.37 | 6.45 | 0.42 | 0.50 | 0.05 | 1.55 | 0.32 | 0.000002 | Up × 2.4 0.000326 | Down × 0.4 0.001387 |
| 42791 | GE83256 | 3.96 | 1.01 | 16.40 | 3.16 | 2.46 | 0.19 | 6.93 | 1.35 | 0.001969 | Up × 2.4 0.000326 | Down × 0.4 0.001387 |
| 43138 | GE83463 | 1.77 | 0.23 | 3.83 | 0.34 | 1.91 | 0.03 | 1.99 | 0.25 | 0.001924 | Up × 1.9 0.000561 | Down × 0.2 0.011517 |
| 43379 | GE83611 | 3.40 | 0.44 | 4.97 | 0.57 | 0.40 | 0.52 | 2.31 | 0.26 | 0.000231 | Up × 1.5 0.049468 | Down × 0.2 0.011517 |

(Continued)

Table 2 (Continued)

| Identifier | Probe | HAD + HIVE | | HAD | | HIVE | | Control | | Between groups | | HAD + HIVE/control | | HAD/control | | HIVE/control | |
|------------|----------|------------|------|-------|------|-------|------|---------|------|----------------|----------|--------------------|------------|-------------|----------|--------------|--|
| | | Mean | SE | Mean | SE | Mean | SE | Mean | SE | P | P | P | P | P | P | | |
| 43489 | GE83679 | 0.37 | 0.11 | 1.46 | 0.09 | 0.57 | 0.07 | 1.00 | 0.07 | 0.000029 | 0.000029 | Down × 0.4 | Up × 1.5 | Down × 0.6 | 0.003600 | 0.013388 | |
| 43664 | GE837848 | 1.16 | 0.20 | -0.13 | 0.25 | 1.34 | 0.06 | 0.69 | 0.13 | 0.001488 | 0.001488 | 0.000145 | Down × 0.2 | Up × 1.9 | 0.005883 | 0.039977 | |
| 44056 | GE84011 | 1.31 | 0.21 | 0.29 | 0.10 | 1.07 | 0.21 | 0.43 | 0.10 | 0.001195 | 0.001195 | Up × 3 | Up × 1.8 | Up × 2.5 | 0.000529 | 0.020259 | |
| 44077 | GE84023 | 2.02 | 0.48 | 7.74 | 0.83 | 2.62 | 0.42 | 4.22 | 0.36 | 0.000066 | 0.000066 | Down × 0.5 | Up × 1.8 | 0.000324 | 0.000324 | 0.000324 | |
| 44304 | GE84156 | 1.37 | 0.23 | 3.52 | 0.25 | 1.20 | 0.18 | 1.79 | 0.28 | 0.001824 | 0.001824 | 0.005099 | Up × 2 | Down × 0.5 | 0.001229 | 0.001229 | |
| 44579 | GE843174 | 0.91 | 0.16 | 2.40 | 0.19 | 0.31 | 0.08 | 1.07 | 0.22 | 0.001392 | 0.001392 | 0.001287 | Up × 2.2 | Down × 0.1 | 0.001287 | 0.047180 | |
| 44682 | GE84381 | 1.30 | 0.38 | 3.28 | 0.58 | 0.12 | 0.30 | 1.35 | 0.21 | 0.001817 | 0.001817 | 0.000790 | Up × 2.4 | Down × 0.5 | 0.001712 | 0.000908 | |
| 44855 | GE84488 | 0.79 | 0.05 | 1.05 | 0.20 | 0.64 | 0.03 | 1.35 | 0.07 | 0.001428 | 0.001428 | Down × 0.6 | Up × 1.4 | Down × 0.3 | 0.000912 | 0.000912 | |
| 45018 | GE84584 | 1.03 | 0.26 | 2.72 | 0.20 | 0.50 | 0.40 | 1.88 | 0.18 | 0.000773 | 0.000773 | Down × 0.5 | Up × 1.4 | Down × 0.3 | 0.027500 | 0.003757 | |
| 45247 | GE847267 | 1.35 | 0.12 | 0.19 | 0.01 | 1.27 | 0.02 | 0.72 | 0.10 | 0.000099 | 0.000099 | Up × 1.9 | Down × 0.3 | Up × 1.8 | 0.015272 | 0.015272 | |
| 45905 | GE85117 | 1.56 | 0.22 | 3.84 | 0.53 | 0.44 | 0.19 | 1.37 | 0.29 | 0.000584 | 0.000584 | 0.000790 | Up × 2.8 | Up × 1.8 | 0.005713 | 0.010591 | |
| 46183 | GE852630 | 0.42 | 0.12 | 0.51 | 0.12 | 0.00 | 0.06 | 1.04 | 0.12 | 0.001524 | 0.001524 | Down × 0.4 | Down × 0.5 | 0.0000469 | 0.000237 | 0.000237 | |
| 46298 | GE853311 | 1.67 | 0.14 | 3.81 | 0.87 | 0.92 | 0.65 | 1.09 | 0.21 | 0.001881 | 0.001881 | 0.003474 | Up × 3.5 | 0.0000000 | 0.015075 | 0.000000 | |
| 47318 | GE859187 | 1.47 | 0.51 | 1.02 | 0.15 | 10.30 | 2.42 | 0.71 | 0.20 | 0.000003 | 0.000003 | 0.000299 | Up × 14.5 | Up × 14.5 | 0.000299 | 0.000299 | |
| 47492 | GE86023 | 0.52 | 0.09 | 0.47 | 0.14 | 1.47 | 0.10 | 0.46 | 0.10 | 0.001037 | 0.001037 | 0.000000 | Up × 3.2 | 0.000000 | 0.000000 | 0.000000 | |
| 47510 | GE86033 | 0.54 | 0.13 | 3.29 | 0.26 | 0.24 | 0.13 | 1.85 | 0.37 | 0.001128 | 0.001128 | Down × 0.3 | Up × 1.8 | Down × 0.1 | 0.015119 | 0.018845 | |
| 47839 | GE86226 | 2.35 | 0.46 | 4.43 | 0.10 | 1.40 | 0.49 | 2.75 | 0.16 | 0.000599 | 0.000599 | 0.015394 | Up × 1.6 | Down × 0.5 | 0.015394 | 0.015394 | |
| 47990 | GE863123 | 1.62 | 0.19 | 0.58 | 0.23 | 0.31 | 0.08 | 0.64 | 0.11 | 0.000872 | 0.000872 | Up × 2.5 | 0.001416 | 0.014709 | 0.001416 | 0.014709 | |
| 48005 | GE86324 | 0.85 | 0.32 | 3.46 | 0.86 | 0.48 | 0.02 | 0.84 | 0.20 | 0.001703 | 0.001703 | 0.000392 | Up × 4.1 | 0.000413 | 0.000392 | 0.000413 | |

| | | | | | | | | | | | | |
|-------|----------|-------|------|-------|------|-------|-------|-------|------|----------|------------------------|------------------------|
| 48087 | GE863731 | 1.51 | 0.21 | 0.07 | 0.22 | 0.06 | 0.13 | 0.87 | 0.12 | 0.000481 | Up × 1.7 0.012794 | Down × 0.1 0.014301 |
| 48127 | GE86393 | 0.59 | 0.17 | 2.27 | 0.18 | 1.03 | 0.17 | 0.67 | 0.18 | 0.000430 | Up × 3.4 0.000085 | Down × 0.1 0.006535 |
| 48128 | GE86394 | 2.43 | 0.35 | 8.01 | 1.16 | 3.88 | 0.30 | 3.35 | 0.64 | 0.002059 | Up × 2.4 0.000712 | Down × 0.1 0.014301 |
| 48161 | GE86416 | 1.43 | 0.07 | 0.45 | 0.04 | 1.04 | 0.25 | 0.50 | 0.13 | 0.000774 | Up × 2.9 0.000190 | Up × 2.1 0.034611 |
| 48356 | GE865354 | 28.90 | 1.02 | 27.32 | 5.38 | 89.57 | 23.60 | 28.03 | 2.13 | 0.000104 | Up × 3.2 0.000017 | Up × 3.2 0.000017 |
| 48491 | GE86614 | 0.78 | 0.10 | 0.61 | 0.21 | 1.52 | 0.03 | 0.45 | 0.09 | 0.001534 | Up × 3.4 0.000178 | Up × 3.4 0.000178 |
| 49316 | GE871079 | 0.52 | 0.13 | 1.09 | 0.20 | 1.32 | 0.26 | 0.27 | 0.08 | 0.000743 | Up × 4 0.000907 | Up × 4 0.000907 |
| 49481 | GE87211 | 1.47 | 0.24 | 1.83 | 0.18 | -0.19 | 0.06 | 1.08 | 0.19 | 0.002047 | Up × 1.7 0.031097 | Down × 0.2 0.003962 |
| 49512 | GE87230 | 1.06 | 0.23 | 2.90 | 0.41 | 1.34 | 0.05 | 0.87 | 0.16 | 0.000390 | Up × 3.3 0.000049 | Down × 0.2 0.003962 |
| 49941 | GE87458 | 0.74 | 0.31 | 3.16 | 0.81 | 0.29 | 0.04 | 0.89 | 0.17 | 0.001867 | Up × 3.6 0.000681 | Down × 0.2 0.003962 |
| 51072 | GE880744 | 6.13 | 0.78 | 12.28 | 1.17 | 5.23 | 0.08 | 6.53 | 0.56 | 0.000468 | Up × 1.9 0.000167 | Down × 0.2 0.003962 |
| 51170 | GE88133 | 0.77 | 0.18 | 8.17 | 0.18 | 0.91 | 0.66 | 1.11 | 0.21 | 0.000000 | Up × 7.4 0.000000 | Down × 0.3 0.003714 |
| 51284 | GE88203 | 0.08 | 0.20 | 0.75 | 0.11 | 0.30 | 0.09 | 1.01 | 0.06 | 0.000342 | Down × 0.1 0.000057 | Down × 0.3 0.003714 |
| 51549 | GE88364 | 5.64 | 0.04 | 15.48 | 3.73 | 5.01 | 0.14 | 5.35 | 0.58 | 0.001288 | Up × 2.9 0.000244 | Down × 0.3 0.003714 |
| 52061 | GE88659 | 0.57 | 0.03 | 2.89 | 0.68 | 0.59 | 0.34 | 0.23 | 0.21 | 0.000509 | Up × 12.6 0.000062 | Down × 0.3 0.003714 |
| 52255 | GE887730 | 2.62 | 0.37 | 5.14 | 0.65 | 1.16 | 0.25 | 2.40 | 0.37 | 0.002039 | Up × 2.1 0.000990 | Down × 0.3 0.003714 |
| 52268 | GE88782 | 0.15 | 0.36 | 11.58 | 4.37 | 0.89 | 0.08 | 0.85 | 0.15 | 0.001374 | Up × 13.6 0.000321 | Down × 0.3 0.003714 |
| 52992 | GE894844 | 0.99 | 0.05 | 0.27 | 0.04 | 0.45 | 0.13 | 1.09 | 0.12 | 0.001294 | Down × 0.2 0.000376 | Down × 0.4 0.006590 |
| 53339 | GE898157 | 0.31 | 0.24 | 0.47 | 0.18 | 2.12 | 0.43 | 1.32 | 0.16 | 0.001107 | Down × 0.2 0.003007 | Down × 0.4 0.006590 |
| 53778 | GE902064 | 1.41 | 0.21 | 0.66 | 0.09 | 1.92 | 0.40 | 0.52 | 0.11 | 0.000618 | Up × 2.7 0.001362 | Up × 1.6 0.040358 |
| 54138 | GE905236 | 1.11 | 0.26 | 3.16 | 0.15 | 1.25 | 0.13 | 1.20 | 0.28 | 0.001997 | Up × 2.6 0.000432 | Up × 3 0.000268 |

Abbreviations: HAD, HIV-associated dementia; HIVE, HIV encephalitis; SE, standard error.

Table 3 Select expressed genes and functions⁴⁴⁻⁴⁶

| Identifier | Probe | Alias | Functions and comments |
|------------|----------|---------------|---|
| 1244 | GE472453 | 2NbHMSP | Immune activation-like gene in multiple sclerosis. |
| 1420 | GE474010 | GRIN2A | Mg ion binding. Ion transport. Plasma membrane integral. |
| 2021 | GE479725 | HECW2 | E3 ubiquitin-protein ligase that mediates ubiquitination of TP73. Acts to stabilize TP73 and enhance activation of transcription by TP73. |
| 2172 | GE481051 | – | – |
| 2573 | GE484741 | ANKRD11 | Member of a family of ankyrin repeat-containing cofactors that interacts with p160 nuclear receptor coactivators and inhibits ligand-dependent transcriptional activation. |
| 2642 | GE485413 | SYPL2 | Transporter activity. Synaptic vesicle integral to membrane. Synaptophysin-like 2. |
| 2830 | GE487382 | – | – |
| 3159 | GE490114 | – | – |
| 3289 | GE491184 | – | – |
| 3577 | GE493533 | – | – |
| 4180 | GE498722 | – | – |
| 4272 | GE499400 | GNAQ | Nucleotide GTP binding GTPase. Signal transducer. Protein ribosylation. Signal transduction G protein coupled receptor signaling pathway. Plasma membrane. Cytoplasm heterotrimeric G protein complex. |
| 4285 | GE499526 | – | – |
| 4361 | GE500216 | – | – |
| 4711 | GE503208 | – | – |
| 5179 | GE507524 | – | – |
| 5674 | GE512134 | NR4A1 | Nuclear transcription factor. Translocation from nucleus to mitochondria induces apoptosis. |
| 6001 | GE515097 | – | – |
| 6005 | GE515151 | PML | Nuclear transcription factor. Protein ubiquitination ligase complex. Zn ion binding. Promyelocytic leukemia. |
| 6006 | GE515161 | MKLN1 | Cell motility. Cell matrix adhesion. Signal transduction. Cytoplasmic. |
| 6064 | GE515618 | TAF4B | Nuclear initiation transcription factor. TFIID complex. |
| 6121 | GE516084 | – | – |
| 6204 | GE516830 | – | – |
| 6530 | GE519581 | TMTC2 | Transmembrane and tetratricopeptide repeat containing 2. Multipass membrane protein. |
| 6580 | GE519998 | – | – |
| 7181 | GE525253 | FNDC5 | Fibronectin type 3 domain-containing 5. |
| 7348 | GE526744 | APOB | Receptor binding lipid transporter. Heparin binding. Signal transduction. ER microsomal. |
| 7400 | GE527127 | – | – |
| 7585 | GE528706 | – | – |
| 7999 | GE53107 | BACH1 | Transcription regulation. Nuclear factor. BTB and CNC homology 1. Basic leucine transcription factor 1 variant 1. |
| 8018 | GE53116 | TLK2 | Nuclear. ATP binding. Serine/threonine kinase. Transferase. Chromatin regulation assembly/disassembly. Response to DNA damage stimulus. Tousled-like kinase 2. |
| 8334 | GE53271 | TRAK2/ALS2CR3 | Receptor binding. Intracellular transporter. Neurotransmitter transport. Cytoplasm. Plasma membrane. Amyotrophic lateral sclerosis 2 juvenile. Chromosome candidate region3. |
| 9079 | GE536414 | ZDHHC5 | Metal ion binding. Membrane integral. Zn finger DHHC-type containing 5. |
| 9181 | GE53692 | B4GALT7 | Galactosyl transferase. Mn ion binding. Xylosyl-protein. Carbohydrate metabolism. Proteoglycan metabolism. Protein modification. Golgi stack. Membrane integral. Xylosyl protein beta 1,4-galactosyl transferase polypeptide 7. Galactosyl transferase 1. |
| 9536 | GE538621 | HIST1H2BC | Nucleosome assembly. DNA binding. Chromosome organization and biogenesis. Histone cluster 1, H2bc. |
| 9814 | GE54005 | CEACAM7 | Plasma membrane integral. Carcinoembryonic antigen-related cell adhesion molecule 7. |
| 10760 | GE54509 | SEC6L1 | Exocytosis protein transport. SEC6-like 1. |
| 11409 | GE548504 | LOC387856 | Hypothetical protein. Similar to expressed sequence AI836003 (GenBank). |
| 11522 | GE549123 | PRR15 | Hypothetical protein. LOC222171. Proline-rich 15 (PRR15). |
| 11545 | GE549241 | NPIP | Nuclear pore complex interacting protein. |
| 11877 | GE55094 | NUDCD1 | HR85 islet cDNA similar 2. |
| 12206 | GE55267 | ADAM28 | Metalloendopeptidase. Zn ion binding. Proteolysis. Spermatogenesis. Membrane integral. Disintegrin and metalloproteinase domain 28 variant 1. |
| 12366 | GE553476 | IAPP | Islet amyloid polypeptide. Like related beta-amyloid associated with Alzheimer's disease, can induce apoptotic cell death. |
| 12612 | GE554808 | LOC283488 | Proline-rich protein. |

(Continued)

Table 3 (Continued)

| Identifier | Probe | Alias | Functions and comments |
|------------|----------|------------|---|
| 12915 | GE556336 | MYO9A | Myosin, actin-based motor molecule, ATPase activity. Unconventional myosins, intracellular movement. Regulates Rho activity in neurons. Regulation of neuronal morphology and function. |
| 13339 | GE558357 | ADAM23 | Metalloendopeptidase. Integrin binding. Proteolysis. Cell adhesion. Central nervous system development. Plasma membrane integral. |
| 14400 | GE563896 | B3GALT1 | UDP-Gal:betaGlcNAc beta 1,3-galactosyltransferase, polypeptide 1. Member of the beta-1,3-galactosyltransferase gene family. Encodes type II membrane-bound glycoproteins with diverse enzymatic functions using different donor substrates (UDP-galactose and UDP-N-acetylglucosamine) and different acceptor sugars (N-acetylglucosamine, galactose, N-acetylgalactosamine). RP11-367C11.1 Stratagene fetal retina. |
| 14504 | GE564415 | – | – |
| 14612 | GE56503 | HIP2 | Huntington interaction protein 2. Ubiquitin-protein ligase-like activating enzyme. Ubiquitin cycle. |
| 14700 | GE565524 | DUSP15 | Protein tyrosine-threonine-serine phosphatase. Hydrolase. |
| 14740 | GE565731 | – | – |
| 14756 | GE56583 | FBLIM1 | Zn ion binding. Adhesion. Cell shape. Cytoskeleton. Filamin binding LIM protein 1. |
| 14820 | GE566190 | – | – |
| 15762 | GE57137 | KIF14 | ATP binding. Microtubule motor and movement. Microtubule-associated complex. Kinasin 14 family member. |
| 16114 | GE57325 | NELL1 | Structure. Ca ion binding. Cell adhesion. Nervous system development. |
| 16469 | GE57516 | TNNI1 | Actin and tropomyosin binding. Regulation of strial muscle contraction. Muscle development. Troponin complex. Slow twitch skeletal troponin I. |
| 16547 | GE57557 | CDK5 | ATP binding. Cyclin-dependent protein kinase 5. Cell cycle. Cell proliferation. Cell division. |
| 16823 | GE576963 | SESNI | Response to DNA damage stimulus. Cell cycle and proliferation arrest. Nucleus. |
| 17180 | GE57883 | SELL | Sugar binding. Cell adhesion and motility. Plasma membrane integral. Selectin-L. Lymphocyte adhesion molecule 1. |
| 17447 | GE58018 | TFAM/ATP88 | Transcription factor. Regulation from RNAP-I promoter. Nucleotide binding. Mg ion binding. Phospholipid translocating ATPase. DNA-dep-DNA replication. Mitochondrion membrane integral. Transcription factor A. |
| 17879 | GE582514 | – | – |
| 17884 | GE58255 | GCKR | Enzyme inhibitor. Glucokinase regulator. |
| 17947 | GE58287 | MGAT2 | Alpha-1,6-mannosyl-glycoprotein2-beta-N-acetyl glucosaminyl transferase. N-linked glycosylation. Oligosaccharide biosynthesis. Membrane integral. Golgi stack. |
| 17974 | GE583033 | – | – |
| 18252 | GE58460 | BSMAP | Transmembrane protein 59-like brain-specific membrane-anchored protein. Modulates the O-glycosylation and complex N-glycosylation steps occurring during the Golgi maturation of amyloid precursor protein. Inhibits amyloid precursor protein transport to the cell surface and further shedding. C19Orf4. |
| 18384 | GE585314 | CENTG2 | ArfGAP with GTPase domain, ankyrin repeat, and PH domain 1. GTPase-activating protein for ARF1 and, to a lesser extent, ARF5. ADP ribosylation factor. Directly and specifically regulates adapter protein 3-dependent trafficking of proteins in the endosomal-lysosomal system. GAP activity stimulated by phosphatidylinositol 3,4,5-trisphosphate (PIP3) and, to a lesser extent, by phosphatidylinositol 4,5-bisphosphate (PIP2). Phosphatidic acid potentiates PIP2 stimulation. C16Orf5. |
| 18390 | GE58535 | – | – |
| 18598 | GE58654 | APH1A | Plasma membrane integral protein ectodomain proteolysis. NOTCH receptor processing. Endoplasmic reticulum, Golgi stack. Anterior pharynx defective 1 homolog A. |
| 18634 | GE586724 | – | – |
| 18765 | GE587496 | BLK | ATP binding. Protein tyrosine kinase. Protein kinase cascade. |
| 19105 | GE58946 | CASP3 | Cysteine-type peptidase, caspase, apoptosis induction. |
| 19877 | GE593831 | MANIA2 | Mannosyl-oligosaccharide-1,2-alpha-mannosidase. Ca ion binding. Hydrolase. Acts on glycosyl bonds. Carbohydrate metabolism. N-glycan processing. Membrane integral Golgi stack. |
| 20353 | GE596515 | PTPRK | Integral transmembrane receptor tyrosine phosphatase. Hydrolase. |
| 20752 | GE59877 | PTPN6 | Protein tyrosine phosphatase. Hydrolase. Apoptosis. G protein coupled receptor protein signaling pathway. Intracellular. Cytoskeleton. Membrane. |
| 20798 | GE599024 | PDZRN3 | Ubiquitin-protein ligase. Zn ion binding. Protein ubiquitination complex. |
| 22253 | GE609375 | ZCSL3 | Heat shock protein binding. Metal ion binding. Unfolded protein binding. Protein folding. |
| 22901 | GE613705 | – | – |
| 22971 | GE61413 | POLDIP2 | Nucleus. Polymerase DNA directed delta-interacting protein 2. |
| 23200 | GE61539 | SMCR7L | RP5-1104E15.5. |

(Continued)

Table 3 (Continued)

| Identifier | Probe | Alias | Functions and comments |
|------------|----------|-----------|--|
| 23526 | GE617302 | RSAFDI | tRNA-γW synthesizing protein I homolog. Wybutosine is a hypermodified guanosine with a tricyclic base at the 3-prime position adjacent to the anticodon of phenylalanine tRNA that stabilizes codon-anticodon interactions during decoding on the ribosome. Wybutosine biosynthesis pathway. |
| 24052 | GE620526 | FMO5 | Mono-oxygenase. Demethyl-aniline mono-oxygenase (N-oxide forming). Electron transport. Endoplasmic reticulum, microsomal. Membrane integral. |
| 24282 | GE62190 | GNG3/GNG7 | Signal transduction. Regulation of G protein coupled receptor protein signaling pathway. Heterotrimeric G protein complex. Guanine nucleotide binding protein gamma-7. |
| 24756 | GE624691 | – | – |
| 24982 | GE626074 | ZA52P | Gastric protein uncharacterized. |
| 25091 | GE62673 | AASS | Lysine ketoglutarate reductase. Oxidoreductase. Saccharopine dehydrogenase. Electron transport. Lysine catabolism. Protein tetramerization. Mitochondrial. Aminoadipate semialdehyde synthase. |
| 25103 | GE62681 | ROMI | Cell adhesion. Sensory and visual perception. Plasma membrane integral. |
| 25830 | GE631063 | – | – |
| 26034 | GE63224 | ETFA | Electron carrier and transport. Mitochondrial matrix. |
| 26491 | GE636205 | HDHDI A | Haloacid dehalogenase-like hydrolase domain containing I. |
| 26776 | GE644246 | SIPA1I1 | Signal-induced proliferation-associated I-like protein I. Interacts with DLG4, PDLIM5, PDLIM7, PROSAPI1, actin cytoskeleton, HPV E6. Cytoplasm, cytoskeleton. Cell junction, postsynaptic density at cell membrane, dendritic spines hippocampal neurons, synaptosome. |
| 26936 | GE648477 | SOX5 | SRY-related HMG box (SOX) transcription regulation factor family. DNA dependent from RNAP2 promoter. Nuclear. |
| 27283 | GE655391 | CDC73 | Cell division cycle 73, Paf1/RNA polymerase II complex component. Tumor suppressor in transcriptional and post-transcriptional control pathways. Component of PAF protein complex, which associates with the RNA polymerase II subunit POLR2A and a histone methyltransferase complex. Facilitates association of 3' mRNA processing factors with actively transcribed chromatin. Cell cycle progression through the regulation of cyclin D1/PRADI expression. |
| 27415 | GE657626 | PTDSS1 | Transferase. Phosphatidyl serine biosynthesis. Phospholipid biosynthesis. Membrane integral. |
| 27568 | GE660354 | C14ORF119 | C14ORF119 |
| 28284 | GE674173 | GPR161 | Rhodopsin-like receptor. Signal transduction. G protein coupled receptor protein signaling pathway. Membrane integral. |
| 28369 | GE675994 | MRPL51 | Mitochondrial ribosomal protein L51. Encoded by nuclear genes. Mitochondrial ribosomes (mitoribosomes) consist of a small 28S subunit and a large 39S subunit. They have an estimated 75% protein to rRNA composition compared with prokaryotic ribosomes, where this ratio is reversed. No 5S rRNA. |
| 28502 | GE678706 | TLL5 | Tubulin tyrosine ligase-like protein family. Interacts with two glucocorticoid receptor coactivators, transcriptional intermediary factor 2, and steroid receptor coactivator 1. Coregulator of glucocorticoid receptor-mediated gene induction and repression. Alpha tubulin polyglutamylase. Involved in the side chain initiation step of the polyglutamylation reaction not elongation step. |
| 28508 | GE678803 | SPG7 | Paraplegin. Spastic paraplegia 7 (pure and complicated autosomal recessive). Cell matrix adhesion regulator. This gene encodes a nuclear-encoded mitochondrial metalloprotease protein that is a member of the ATPases associated with a variety of cellular activities protein family. Members of this protein family share an ATPase domain and have roles in diverse cellular processes including membrane trafficking, intracellular motility, organelle biogenesis, protein folding, and proteolysis. Mitochondrion membrane, multipass membrane protein. |
| 28991 | GE687963 | USP8 | Cysteine-type endopeptidase. Ubiquitin thiol esterase. Ubiquitin-dependent protein catabolism. Ubiquitin cycle. Cell proliferation. |
| 29168 | GE691505 | – | – |
| 29917 | GE705764 | NALP1 | Nod-like receptor family, pyrin domain containing I. Death effector filament-forming CED-4-like apoptosis protein. ATP binding. Caspase recruitment domain protein 7. Caspase activator. Enzyme binding. Apoptosis induction and regulation. Defense response to pathogen. Intracellular. |
| 30069 | GE708617 | – | – |
| 30107 | GE709371 | HRB | DNA, RNA, metal ion binding. mRNA export, nuclear pore. Regulation of GTPase. |
| 30167 | GE710687 | CCDC7 | Coiled-coil domain-containing 7. |

(Continued)

Table 3 (Continued)

| Identifier | Probe | Alias | Functions and comments |
|------------|----------|-----------------|--|
| 31250 | GE726916 | TBC1D22A | GTPase activator. |
| 31353 | GE728396 | – | – |
| 31404 | GE729136 | GGA1 | Protein transporter and complex assembly. Intracellular Golgi stack protein transport. Membrane. Clathrin coat of transGolgi network vesicle. |
| 31632 | GE732434 | TARSL2 | Threonyl-tRNA synthetase-like protein 2, ligase. |
| 32204 | GE740641 | – | – |
| 32318 | GE742294 | DMTF1 | Cyclin D binding MYB-like transcription factor 1. Contains a cyclin D-binding domain, three central MYB-like repeats, and two flanking acidic transactivation domains at the N-terminus and C-terminus. Induced by oncogenic Ras signaling pathway and functions as a tumor suppressor by activating the transcription of ARF-p53 pathway to arrest cell growth or induce apoptosis. Activates transcription of aminopeptidase N and plays role in hematopoietic cell differentiation. Transcription regulated by binding D-cyclins. Transcriptional activator activates CDKN2A/ARF locus in response to Ras-Raf signaling, thereby promoting TP53/p53-dependent growth arrest. Binds to the consensus sequence 5'-CCCG[GT]ATGT-3'. Isoform 1 may cooperate with MYB to activate transcription of the ANPEP gene. Isoform 2 may antagonize transcriptional activation by isoform 1. |
| 32836 | GE749435 | MSI2 | Nucleotide and RNA binding. |
| 33031 | GE752199 | – | – |
| 33193 | GE754378 | ELP4 | – |
| 33293 | GE755614 | GRM3 | Metabotropic glutamate, gamma aminobutyric acid B-like receptor. Signal transduction. G protein coupled receptor signaling pathway. Negative regulation of adenylyclase. Plasma membrane integral. |
| 33722 | GE762426 | – | – |
| 33921 | GE765425 | GRIA3 | Glutamate receptor, ionotropic, AMPA 3. AMPA-selective glutamate receptor 3. Excitatory. AMPA is alpha-amino-3-hydroxy-5-methyl-4-isoxazole propionate. AMPA receptors mediate fast excitatory synaptic transmission in the central nervous system and play a key role in hippocampal synaptic long-term potentiation and depression. |
| 34083 | GE767593 | CACNB2 | Voltage-gated Ca channel complex. Ca ion binding and transport. Neuromuscular junction development. Membrane fraction. |
| 34195 | GE769111 | GPRI33 or GPI33 | – |
| 34221 | GE769398 | MSI2 | Nucleotide and RNA binding. |
| 34239 | GE769588 | PTF1A | – |
| 34398 | GE772048 | WDFY1 | Phosphatidyl inositol and Zn ion binding. Nuclear. Early endosome. Cytosol. |
| 35620 | GE78986 | JUB | Component of cellular adhesive complexes. Contributes to cell fate determination and regulates cell proliferation and differentiation. Involved in the regulation of actin cytoskeleton dynamics and cell migration. Contributes to linking of epithelial cell junctions through adhesive receptors to actin cytoskeleton. Signal transduction from cell adhesion sites to the nucleus. Regulates kinase activity of AURKA/Aurora-A for mitotic commitment. Component of interleukin-1 signaling pathway modulating interleukin-1-induced nuclear factor kappa-B activation by influencing the assembly and activity of the PRKCZ/SQSTM1/TRAF6 multiprotein signaling complex. Transcription complex formation on DNA. Interacts with AURKA/Aurora-A during mitosis and both proteins are phosphorylated in a complex. Interacts with CTNNA1/alpha-catenin and with F-actin. Interacts with LATS2 during mitosis and regulates organization of the spindle apparatus through recruitment of gamma tubulin to the centrosome. Interacts with GRB2 and PIP5 K1 A. Forms a complex with SQSTM1, PRKCZ, and TRAF6. Interacts with SLC1 A2. Located in the cytoplasm, cytoskeleton, cell membrane, cell junction, nucleus, and centrosome. Shuttles between cytoplasm and the nucleus. Localizes on centrosomes during G2-M phase. Preferentially colocalizes with cadherin-adhesive complexes at sites of cell-cell contact. LIM region interacts with CTNNA1. The preLIM region binds directly actin filaments. LIM-2 and LIM-3 domains mediate the interaction with the N-terminal region of AURKA. The association between LATS2 and JUB required the second LIM domain of JUB. Belongs to the Zyxin/Ajuba family. Contains three LIM zinc-binding domains. |
| 35666 | GE790167 | – | – |
| 35761 | GE79076 | ULBP3 | Major histocompatibility complex class I receptor complex. Antigen presentation. Natural killer activation. Membrane. ULI6-binding protein 3. |
| 36075 | GE79260 | RAP1A | Small GTPase-mediated signal transduction. GTP binding. Intracellular protein transport. Cell cycle. Negative regulation of cell cycle progression. Membrane. Ras oncogene family (RAP1A). |

(Continued)

Table 3 (Continued)

| Identifier | Probe | Alias | Functions and comments |
|------------|----------|----------------|---|
| 36101 | GE79273 | USF1 | DNA-dependent specific RNA polymerase 2 promoter transcription factor and regulator. Nuclear. Upstream transcription factor 1. Secretoglobin, family 1 A member 1 (uteroglobin). |
| 36278 | GE79382 | RPL7A | Structural constituent of ribosome. Protein biosynthesis. Ribosome biogenesis and assembly. |
| 36337 | GE79415 | PSP | Hypothetical protein MGC17299. |
| 36363 | GE794289 | – | – |
| 36885 | GE797280 | – | – |
| 36949 | GE79764 | SFRS11/PLEKHA5 | RNA binding. Phosphatidyl inositol binding. Nuclear mRNA splicing factor via spliceosome. Arginine/Serine-rich 11. Plekstrin homology domain containing family A member 5 mRNA. |
| 36990 | GE79788 | SAMD13 | – |
| 37273 | GE799491 | FLJ14167 | KCNJN1. Potassium inwardly rectifying channel, subfamily J, member 12. Inward rectifier potassium channel Kir2.2v. IRK-2. ATP-sensitive inward rectifier potassium channel 1. Potassium inwardly-rectifying channel, subfamily J, inhibitor 1. Kir2.2v. Establishing action potential waveform and excitability of neurons. Voltage dependence regulated by concentration of extracellular potassium. Inwardly rectifying potassium channel blocked by divalent cations. Inward rectifier potassium channels allow potassium to flow into the cell rather than out of it. As external potassium is raised, the voltage range of the channel opening shifts to more positive voltages. Inward rectification is due to blockage of outward current by internal magnesium. Can be blocked by extracellular barium and cesium. The inward rectifier potassium channel family (also known as 2-TM channels) include the strong inward rectifier channels (KIR2.x), the G protein-activated inward rectifier channels (KIR3.x) and the ATP-sensitive channels (KIR6.x, which combine with sulfonylurea receptors). Structurally, the pore-forming subunit of KIR channels is the alpha subunit. It contains a single pore domain between two membrane-spanning regions. Four alpha subunits combine to form a tetramer, with the pore domain of each subunit contributing to the structure of the central pore. Heteromeric channels can also be formed within subfamilies, eg, KIR3.2 with KIR3.3. |
| 38007 | GE80398 | AKR1C1/AKR1C2 | Aldo-keto reductase family 1, member C2. Electron transporter. Bile acid transporter. Oxidoreductase. 20-alpha-hydroxy-steroid dehydrogenase. Trans-1,2-dehydrobenzene-1,2-diol dehydrogenase. Xenobiotic and lipid metabolism. Transport. Digestion. Steroid metabolism. Dehydrodiol dehydrogenase 2. Bile acid binding protein. 3-alpha-hydroxysteroid dehydrogenase type 3 (AKR1C2) transcript variant 1 mRNA. Canalicular bile acid transport. Cytoplasm. AKR1C1 mRNA. |
| 38049 | GE804261 | LOC285626 | Hypothetical protein. |
| 38070 | GE804386 | RP9 | Metal ion binding. Sensory and visual perception. RNA splicing. Nuclear. Retinitis pigmentosa. Autosomal dominant. |
| 38729 | GE808417 | TXNL6 | Thioredoxin-like protein 6. Nucleoredoxin-like protein. Rod-derived cone viability factor. |
| 38739 | GE80847 | TGM7 | Gamma glutamyl transferase. Ca ion binding. Acyl transferase. Peptide cross-linking. Transglutaminase 7. |
| 38813 | GE80890 | COL5A1 | Extracellular matrix structural constituent. Heparin binding. Phosphate transport. Cell adhesion. Collagen type V alpha 1. Cytoplasm. |
| 38882 | GE809301 | – | – |
| 39376 | GE812224 | MGC39606 | Hypothetical protein. Nonprotein coding RNA 86. Cytogenetic band Xq26.3. |
| 39516 | GE813126 | – | – |
| 39690 | GE81418 | SULT2B1 | Alcohol steroid sulfotransferase. Lipid and steroid metabolism. Cytoplasm. Sulfotransferase family, cytosolic, 2B, member 1. |
| 39743 | GE81449 | GDF15 | Cytokine. Growth factor. Signal transduction. Transforming growth factor beta-receptor signaling pathway. Cell-cell signaling. Extracellular space. Growth differentiation factor 15. |
| 40333 | GE81822 | PYCR2 | Pyrraline-5-carboxylate reductase family member 2. Oxidoreductase. Electron transport. Proline biosynthesis. |
| 40551 | GE819522 | – | – |
| 40642 | GE820114 | KIAA1370 | Hypothetical protein. LOC5620. |
| 40690 | GE820397 | – | – |
| 41169 | GE82307 | IQCC | IQ motif-containing C. |
| 41665 | GE82602 | HOXD11/HOXD10 | Transcription factor related to RNAP II. Development. Nuclear. Homeobox D11/10. Development. |
| 41868 | GE82723 | DGCR8 | Double-stranded RNA binding. DiGeorge syndrome, critical region gene 8. |
| 41968 | GE82785 | C7ORF26 | Chromosome 7 Orf 26. |

(Continued)

Table 3 (Continued)

| Identifier | Probe | Alias | Functions and comments |
|------------|----------|--------------|--|
| 42072 | GE82842 | NOX5 | NADPH oxidase, EF hand Ca binding domain 5. |
| 42550 | GE831160 | – | – |
| 42718 | GE832143 | MSI2 | RNA binding. |
| 42726 | GE83218 | APBA2BP | Amyloid beta (A4) precursor protein binding family A member 2 binding protein. Transcript variants 1 and 2. Ca ion binding. Oxidoreductase. Protein secretion. Antibiotic biosynthesis. Protein metabolism. Regulation of amyloid precursor protein biosynthesis. Golgi cisternae. Nuclear. Cytoplasm. Endoplasmic reticulum membrane. |
| 42766 | GE832421 | ACOT6 | Acyl-CoA thioesterase 6. |
| 42791 | GE83256 | NRIP2 | Nuclear receptor interacting protein 2. |
| 43138 | GE83463 | ATP2B4 | Hypothetical protein. MGC5457, mRNA. |
| 43379 | GE83611 | NYD-SP26 | Testis development protein. |
| 43489 | GE83679 | SLAMF6 | SLAM family member 6. CD2 surface receptor. Membrane integral. |
| 43664 | GE837848 | KCMK12 | Voltage-gated K ion channel transport. Membrane integral. |
| 44056 | GE84011 | – | – |
| 44077 | GE84023 | TGM2 | Protein-glutamine. Gamma glutamyl transferase. Ca ion binding. GTP binding. Acyl transferase. G protein coupled receptor. Signaling pathway. Peptide cross-linking. Positive regulation of cell adhesion. Extracellular matrix. Cytosol. Membrane. |
| 44304 | GE84156 | – | – |
| 44579 | GE843174 | – | – |
| 44682 | GE84381 | – | – |
| 44855 | GE84488 | – | – |
| 45018 | GE84584 | – | – |
| 45247 | GE847267 | SLC44A5 | Solute carrier family 44, member 5; choline transporter-like protein 5. |
| 45905 | GE85117 | NARF | Nuclear prelamin A recognition factor. Similarity to iron-only hydrogenase-like protein 2. Prenyl-dependent prelamin A binding protein. Prenylation and farnesylation at carboxyl terminal end for membrane attachment and protein interactions. On cysteine residue of carboxyl-terminal CaaX motif. Component of a prelamin A endoprotease complex. Cysteine residue is removed from prelamin A when it is endoproteolytically processed into mature lamin A. Co-localizes with the nuclear lamina. |
| 46183 | GE852630 | – | – |
| 46298 | GE853311 | KIAA0922 | Transmembrane protein 131-like isoform-1. |
| 47318 | GE859187 | – | – |
| 47492 | GE86023 | EIF4A2 | DNA and RNA binding. Translation initiation factor. Protein biosynthesis. Regulation of translational initiation. Eukaryotic translation initiation factor 4F complex. |
| 47510 | GE86033 | DNAJC3 | DnaJ (Hsp40) homolog, subfamily C, member 3. Interferon-induced, double-stranded RNA-activated protein kinase inhibitor. Tetratricopeptide repeat family of proteins. Highly conserved J domain found in DNAJ chaperone family members. Involved in the unfolded protein response during endoplasmic reticulum stress. Co-chaperone of HSPA8/HSC70, stimulates its ATPase activity. Inhibits both autophosphorylation of EIF2 AK2/PKR and the ability of EIF2 AK2 to catalyze phosphorylation of the EIF2 A. Inhibits EIF2 AK3/PERK activity. Structural constituent of ribosome. Protein biosynthesis. Small ribosomal subunit protein S23. |
| 47839 | GE86226 | RPS23 | – |
| 47990 | GE863123 | – | – |
| 48005 | GE86324 | TMED1 | Transmembrane emp-24 domain-containing 1. |
| 48087 | GE863731 | SLC36A4 | Solute carrier family 36 (proton/amino acid symporter), member 4. |
| 48127 | GE86393 | ATP2A2 | ATP, Mg, and Ca ion binding. Calcium transport ATPase. Hydrolase acts on acid anhydrides. Transmembrane transporter. Cation transport. Cell adhesion. Metabolism. Epidermis development. Membrane fraction. Microsome. Plasma membrane integral. Sarcoplasmic reticulum. |
| 48128 | GE86394 | CSNK1E | Nucleotide binding. Protein serine/threonine kinase. Casein kinase I. Protein Tyrosine kinase. DNA repair. Signal transduction. Casein kinase I epsilon. |
| 48161 | GE86416 | FDFT1 | Mg ion binding. Farnesyl diphosphate farnesyl transferase. Oxidoreductase. Cholesterol biosynthesis. Isoprenoid biosynthesis. Membrane integral. |
| 48356 | GE865354 | MGC39606 | Nonprotein coding RNA 86. Xq26.3 chromosome band location. NCRNA00086. |
| 48491 | GE86614 | OR2T35/OR2T2 | Olfactory receptor. Signal transduction. G protein coupled receptor. Sensory olfactory perception. Membrane integral. Olfactory receptor, family 2, subfamily T, members 35 and 2. |
| 49316 | GE871079 | KIAA1026 | Kazrin isoform A. |
| 49481 | GE87211 | – | – |

(Continued)

Table 3 (Continued)

| Identifier | Probe | Alias | Functions and comments |
|------------|----------|-----------|--|
| 49512 | GE87230 | LRFN5 | Leucine-rich repeat and fibronectin type 3 domain-containing 5. |
| 49941 | GE87458 | FKSG24 | Hypothetical protein. MGC12972 (FKSG24). |
| 51072 | GE880744 | KIAA1754 | Inositol 1,4,5-triphosphate receptor interacting protein. ITPRIP. Danger. |
| 51170 | GE88133 | SL336 A4 | Solute carrier family 36 (proton/amino acid symporter), member 4. |
| 51284 | GE88203 | SLC9 A9 | Sodium:hydrogen antiporter. Solute: hydrogen. Sodium ion binding. Sodium ion transport. Regulation of pH. Membrane integral. Solute carrier family 9 (sodium/hydrogen exchanger) isoform 9. |
| 51549 | GE88364 | MAPK11 | ATP binding. Protein serine/threonine kinase. MAP kinase. MP kinase and transferase. Response to stress. Signal transduction. Protein kinase cascade. Antimicrobial humoral response. Mitogen-activated protein kinase 11. |
| 52061 | GE88659 | ZNRF2 | Zinc and ring finger 2. |
| 52255 | GE887730 | – | – |
| 52268 | GE88782 | C10ORF118 | CTCL tumor antigen HD-CL-01/L14-2. |
| 52992 | GE894844 | GYP A | Glycophorin A sialoglycoprotein of the human erythrocyte membrane. Receptor for influenza virus and hepatitis A virus. Affects function of SLC4A1. |
| 53339 | GE898157 | HNRPAIP5 | Heterogeneous nuclear ribonucleoprotein A1 pseudogene 5. |
| 53778 | GE902064 | – | – |
| 54138 | GE905236 | ABCB11 | ATP-binding cassette, subfamily B (MDR/TAP), member 11. Membrane-associated protein. Member of the superfamily of ATP-binding cassette transporters that transport various molecules across extracellular and intracellular membranes. MDR/TAP subfamily involved in multidrug resistance. |

pathway, signal transduction, signaling transforming growth factor- β , signaling immediate early, spliceosome, splicing factor, splicing factor RNA, synaptic function, trafficking endolysosomal system, trafficking protein, transcription factor, transcription factor upstream, transcription factor antagonist, transcription promoter, transcription regulation at RNAP-1 promoter, transcription regulation at RNAP-2, transcription, homeobox (development), binding nucleotide, transferase acyl, transferase farnesyl, transferase steroid sulfo, transport antiporter (sodium-hydrogen), transport cation, transport carrier solute, transport intracellular, transport lipid, transport membrane associated, transport metal ion, transport multidrug resistance, transport neurotransmitter, transport phosphate, transport phospholipid, transport mRNA, transport protein, transport symporter amino acid, tRNA ligase, tRNA nucleotide modification, tRNA synthase, tubulin, tumor antigen, ubiquitin protein catabolism, ubiquitin cycle, ubiquitin pathway, and zinc finger.

Pathways

Figure 1 illustrates typical pathways and connections among seven select genes. The seven genes are *APOB*, *NECAB3* (*APBA2BP*), *GRIA3*, *IAPP*, *HOXD10*, *UBE2K*, and *NELL1*. Gene functions are shown in Table 3. The seven genes and their interconnected related pathways are: *APOB*, *IAPP*, and *NECAB3* (*Apba2BP*), the beta-amyloid pathway; *HOXD10* and *UBE2K*, the ubiquitination pathway; *GRIA3*, other glutamate receptors; and *NELL1*, signaling and amyloid

production. These seven genes are interconnected via genes (inserted by the GenePro program) in overlapping pathways that broadly include signaling, transcription, amyloid, and ubiquitination pathways. Similarly, interconnections and pathways may be produced for the other 143 genes in Table 3, that are too numerous and complex to show in one figure.

Discussion

Of the 197 genes that showed significant expression changes in HAD/HIVE, HAD alone, HIVE alone, versus HIV⁺, 150 genes were identified. These genes were members of 159 groups and functions. It is beyond the scope of this article to analyze the genes in detail and the ramifications of the disease state within which gene expression varied significantly. The groups and functions, within which the genes fall, overlap many of the cellular processes in neurons. Although several of these cellular processes may not be considered neuron-specific, they are most likely expressed as part of the stress and attempt-at-recovery processes that the neurons exhibit in HAD/HIVE, HAD, and HIVE, compared with the control HIV⁺.

Broadly, the categories (with some descriptors) include adhesion (intercellular interactions), amyloid (implicated in damage to cognition in Alzheimer's disease), apoptosis (neuronal dysfunction and cell death, also certainly associated with the end state of loss of cognition), binding (of various metal and biochemical ions, crucial in cellular processes), channel complexes (components of ion transport within cells and the plasma membrane), cell cycle (attempts

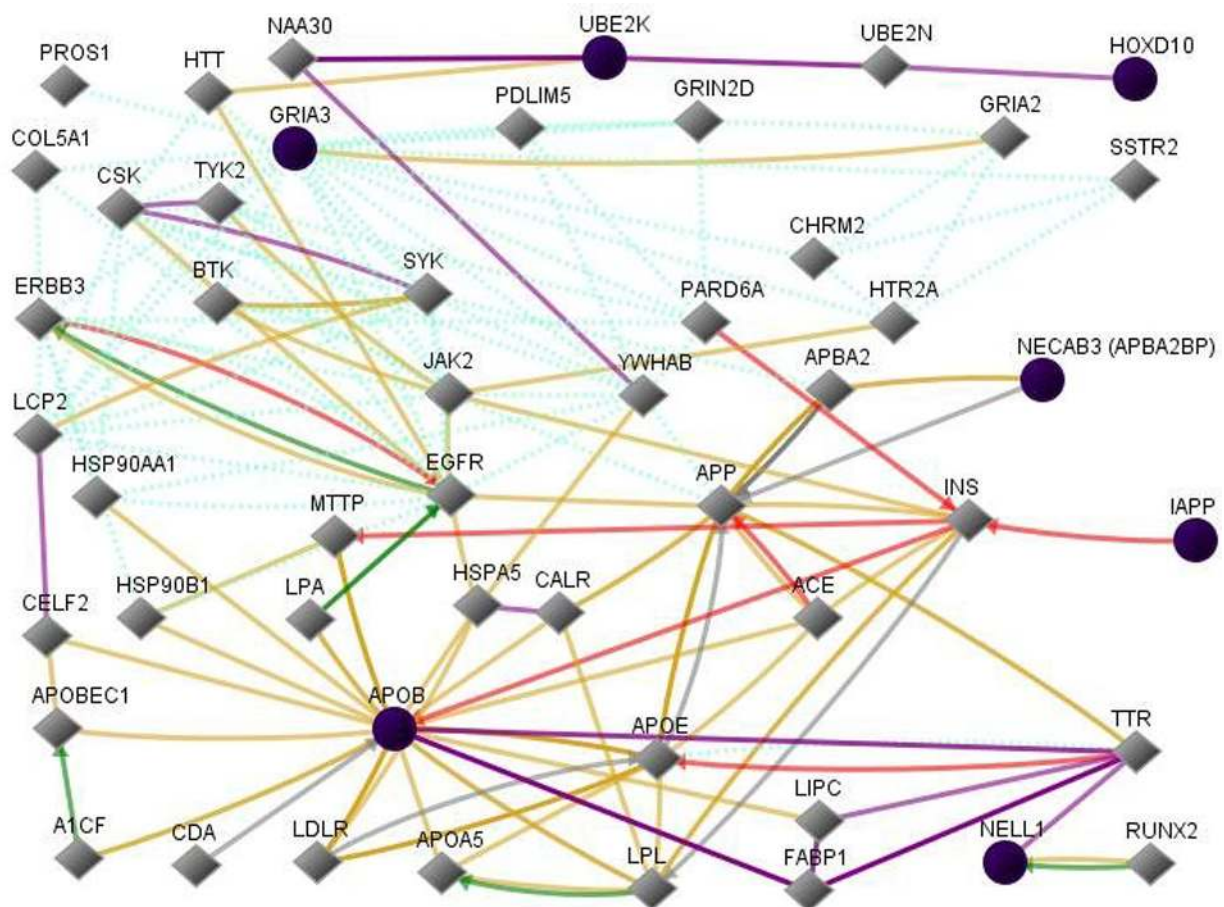


Figure 1 Pathway connections among *APOB*, *NECAB3 (APBA2BP)*, *GRIA3*, *IAPP*, *HOXD10*, *UBE2K*, and *NELL1*. The seven genes are indicated by solid circles. Diamonds indicate neighboring genes inserted by the GenePro program. Arrowheads indicate directional effects. The colors indicate the following: red, downregulation of function and transcription; green, upregulation of function and transcription; gray, regulation exists but direction unknown as yet; beige, gene products directly interact; dotted light blue, predicted protein–protein interaction; and purple, correlated expression detected by microarray experiments.⁴⁶ The seven genes are selected as representative of pathways including signaling, transcription, amyloid, and ubiquitination.

by the cell to expel noxious molecules through shutdown or traversing the cell division cycle), chaperone (assisting proteins to attain and maintain functional conformations), chromatin (central in transcription, genome maintenance and repair, and epigenetics), cytokines (inflammation), cytoskeleton, filaments, and matrix (scaffolding and intracellular transport), diGeorge syndrome (genes involved in brain development), and Huntington's disease (trinucleotide repeats that result in gene dysfunction), metabolism (breakdown of biochemical and cell components), mitochondria (energy production for the cell and also proteins needed for mitochondrion function and survival), multiregion detection protein or RNA (proteins or RNAs that are involved in multiple different molecular pathways and networks), sensory perception (in this study, visual-related and olfactory-related protein expression was perturbed), receptor (binding that is required prior to an effect being exerted, signaling, by proteins and solutes), ribosome and tRNA (key elements

in protein synthesis), noncoding miRNA (a novel realm in the control of gene expression), signaling (intracellular and extracellular molecular pathways), splicing (transcription), synapse (crucial in neuron function), transcription factor (proteins involved in initiation and process of transcription), transport (intracellular and intercellular movement of proteins and ionic and nonionic solutes), multidrug resistance (a process by which cells become resistant to drugs by shutting down their transport), and ubiquitin cycle (protein turnover). In addition, it should be noted that the ubiquitin pathway marks proteins for metabolism and degradation, whereas chaperones assist proteins to attain their optimal functional states.^{26,49} We hypothesize the existence of multi-network detection proteins and RNAs. Such proteins and RNAs would be involved in multiple unrelated molecular pathways and networks. This is consequently different from proteins that are involved in multiple, but related, pathways or homeobox transcription genes of development. For

example, MYO9A may be a multinetwork detection protein because it interacts with myosin filaments and actin-based motor molecules involved in intracellular movement, has ATPase activity, and regulates rho activity, integrin binding, proteolysis, cell adhesion, and central nervous system development.^{45,46}

Potentially devastating effects for neuronal function and survival could result from gene expression changes in beta-amyloid-like protein and amyloid beta-A4 precursor protein binding family A member 2 binding protein. The effects of the former may be due to its amyloid-like properties and the effects of the latter, changes that may occur in amyloid precursor protein metabolism and signaling, due to changes in the receptor protein expression. In addition, changes in apolipoprotein B expression could be associated with dementia in NeuroAIDS as it is in Alzheimer's disease.⁵⁰ Severe changes in gene expression are anticipated, due to the stress that results from chronic HIV-1 infection of the brain. Accordingly, expression of glycophorin A is an example of such severe changes that can possibly occur in the neuron in NeuroAIDS. Glycophorin is a well known component of red blood cell membranes. The RNA that is purified in our procedures is free of all proteins and the detection method used is purely nucleic acid. Moreover, even if glycophorin mRNA were present in mature circulating red blood cells, red blood cells would not be present in our neuronal preparations, because we excise neurons from 10 micron thick sections (ie, smaller than the diameter of these neurons), the neurons are clearly identified with Nissl stain, and are the only cells with nucleoli in these sections. In addition, there were no endothelial cells associated with the neurons because of precision of excision by the laser beam. Likewise, red blood cells would be even further away from the excised neurons and well outside the laser excision perimeter. This greatly reduces the possibility of purifying and amplifying mRNA for glycophorin from red blood cells or any other potentially contaminating cells in our preparations. The glycophorin or glycophorin-like RNA that we detected, in all likelihood, is derived from anomalous glycophorin gene expression in the neurons we analyzed. Also, this is most likely due to the stress undergone by these neurons in their chronic state of disease.

This study is an initial step towards identifying specific genes in neuroanatomically specific neurons that may be involved in neurodegenerative processes that result from HIV-1 infection of the brain. Moreover, a wide range of biochemical processes in the health and maintenance of the cell are dysregulated. Some genes are novel, including for

multinetwork detection proteins. This line of investigation is useful and will provide further specific information about dysfunction of gene expression in HAND.

Conclusion

Novel directions in the analysis and categorization of the transcriptome in disease and health are under development for HAND. For example, systems biological approaches are being developed to elucidate transcriptome organization patterns that are highly correlated across samples and that identify groups of genes or modules.⁵¹ In addition, future prospective studies should be designed to answer additional questions, for example, related to virus load, symptomatology, as well as comparisons across the different stages in the evolution of diagnostic criteria for NeuroAIDS. It will also be of use to validate the data with additional patient cohorts.

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Disclosure

The authors report no conflicts of interest in this work.

References

1. Petito CK, Kerza-Kwiatecki AP, Gendelman HE, et al. Neuronal injury in HIV infection. *J Neurovirol.* 1999;5:327–341.
2. Kaul M. HIV-1 associated dementia: Update on pathological mechanisms and therapeutic approaches. *Curr Opin Neurol.* 2009;22:315–320.

3. Minagar A, Shapshak P. *HIV-associated Dementia: Clinical Features and Pathogenesis in NeuroAIDS*. Hauppauge, NY: Nova Science Publishing; 2006.
4. Minagar A, Commins D, Alexander JS, et al. NeuroAIDS: Characteristics and diagnosis of the neurological complications of AIDS. *Mol Diagn Ther*. 2008;12:25–43.
5. Shapshak P, Kanguane P, Fujimura RK, et al. Editorial NeuroAIDS Review. *AIDS*. 2011;25:123–141.
6. Antinori A, Arendt G, Becker JT, et al. Updated research nosology for HIV associated neurocognitive disorders. *Neurology*. 2007;69:1789–1799.
7. Cysique LA, Maruff P, Brew BJ. Prevalence and pattern of neuropsychological impairment in human immunodeficiency virus-infected/acquired immunodeficiency syndrome (HIV/AIDS) patients across pre- and post-highly active antiretroviral therapy eras: A combined study of two cohorts. *J Neurovirol*. 2004;10:350–357.
8. McArthur JC. HIV dementia: An evolving disease. *J Neuroimmunol*. 2004;157:3–10.
9. Tozzi V, Balestra P, Lorenzini P, et al. Prevalence and risk factors for human immunodeficiency virus-associated neurocognitive impairment, 1996 to 2002: Results from an urban observational cohort. *J Neurovirol*. 2005;11:265–273.
10. Kopniski KL, Jing Bao, Lin YW. Neurobiology of HIV, psychiatric and substance abuse comorbidity research: Workshop report. *Brain Behav Immun*. 2007;21:428–441.
11. Budka H. The neuropathology of HIV-associated brain disease. In: Gendelman HE, Grant I, Everall I, Lipton SA, Switzer S, editors. *The Neurology of AIDS*. New York, NY: Oxford University Press; 2005.
12. Arendt G, von Giesen HJ. Human immunodeficiency virus dementia: Evidence of a subcortical process from studies of fine finger movements. *J Neurovirol*. 2002;8 Suppl 2:27–32.
13. Cherner M, Masliah E, Ellis RJ, et al. Neurocognitive dysfunction predicts postmortem findings of HIV encephalitis. *Neurology*. 2002;59:1563–1567.
14. Persidsky Y, Gendelman HE. Mononuclear phagocyte immunity and the neuropathogenesis of HIV-1 infection. *J Leukoc Biol*. 2003;74:691–701.
15. Sperber K, Shao L. Neurologic consequences of HIV infection in the era of HAART. *AIDS Patient Care STDS*. 2003;17:509–518.
16. Mattson MP, Haughey NJ, Nath A. Cell death in HIV dementia. *Cell Death Differ*. 2005;12 Suppl 1:893–904.
17. Persidsky Y, Poluektova L. Immune privilege and HIV-1 persistence in the CNS. *Immunol Rev*. 2006;213:180–194.
18. Minagar A, Shapshak P, Duran EM, et al. Gene expression in HIV-associated dementia, Alzheimer's disease, multiple sclerosis, and schizophrenia. *J Neurol Sci*. 2004;224:3–17.
19. Shapshak P, Minagar A, Duran EM, et al. Gene expression in HIV associated dementia. In: Minagar A, Alexander JS, editors. *Inflammatory Disorders of the Nervous System, Clinical Aspects, Pathogenesis, and Management*. Totowa, NJ: Humana Press; 2005.
20. Shapshak P, Duncan R, Torres-Munoz JE, Duran EM, Minagar A, Petitto CK. Analytic approaches to differential gene expression in AIDS vs control brains. *Front Biosci*. 2004;9:2935–2946.
21. Shapshak P, Duncan R, Minagar A, et al. Elevated expression of IFN-gamma in the HIV-1 infected brain. *Front Biosci*. 2004;9:1073–1081.
22. Masliah E, Roberts ES, Langford D, et al. Patterns of gene dysregulation in the frontal cortex of patients with HIV encephalitis. *J Neuroimmunol*. 2004;157:163–175.
23. Borjabad A, Brooks AI, Volsky DJ. Gene expression profiles of HIV-1-infected glia and brain: Toward better understanding of the role of astrocytes in HIV-1-associated neurocognitive disorders. *J Neuroimmune Pharmacol*. 2010;5:44–62.
24. Galey D, Becker K, Haughey N, et al. Differential transcriptional regulation by human immunodeficiency virus type 1 and gp120 in human astrocytes. *J Neurovirol*. 2003;9:358–371.
25. Shapshak P, Duncan R, Nath A, et al. Gene chromosomal organization and expression in cultured human neurons exposed to cocaine and HIV-1 proteins gp120 and tat: Drug abuse and NeuroAIDS. *Front Biosci*. 2006;11:1774–1793.
26. Gelman BB, Schuenke KW. Brain aging in AIDS: Increased ubiquitin-protein conjugate and correlation with decreased synaptic protein but not A β -stained diffuse plaque. *J Neurovirol*. 2004;10:98–108.
27. Gelman BB, Soukup VM, Schuenke KW, et al. Acquired neuronal channelopathies in HIV-associated dementia. *J Neuroimmunol*. 2004;157:111–119.
28. Gartner S. Mechanisms of HIV entry into the CNS. In: Minagar A, Shapshak P, editors. *Neuro-AIDS*. New York, NY: Nova Scientific Publishing Inc; 2006.
29. Shapshak P, Stewart RV, Rodriguez de la Vega P, et al. Brain macrophage surface marker expression with HIV-1 infection and drug abuse: A preliminary study. *J NeuroAIDS*. 2003;2:37–50.
30. Pulliam L, Bing Suna B, Hans Rempela H. Invasive chronic inflammatory monocyte phenotype in subjects with high HIV-1 viral load. *J Neuroimmunol*. 2004;157:93–98.
31. Roberts ES, Zandonatti MA, Watry DD, et al. Induction of pathogenic sets of genes in macrophages and neurons in NeuroAIDS. *Am J Pathol*. 2003;162:2041–2057.
32. Torres-Munoz J, Stockton P, Tacronte N, Roberts B, Maronpot RR, Petitto CK. Detection of HIV-1 gene sequences in hippocampal neurons isolated from postmortem AIDS brains by laser capture microdissection. *J Neuropathol Exp Neurol*. 2001;60:885–892.
33. Trillo-Pazos G, Diamanturos A, Rislove L, et al. Detection of HIV-1 DNA in microglia/macrophages, astrocytes and neurons isolated from brain tissue with HIV-1 encephalitis by laser capture microdissection. *Brain Pathol*. 2003;13:144–154.
34. Thompson KA, Churchill MJ, Gorry PR, et al. Astrocyte specific viral strains in HIV dementia. *Ann Neurol*. 2004;56:6873–6877.
35. Standaert DG. Applications of laser capture microdissection in the study of neurodegenerative disease. *Arch Neurol*. 2005;62:203–205.
36. Burgoon MP, Keays KM, Owens GP, et al. Laser-capture microdissection of plasma cells from subacute sclerosing panencephalitis brain reveals intrathecal disease-relevant antibodies. *Proc Natl Acad Sci U S A*. 2005;102:7245–7450.
37. Churchill MJ, Gorry PR, Cowley D, et al. Use of laser capture microdissection to detect integrated HIV-1 DNA in macrophages and astrocytes from autopsy brain tissues. *J Neurovirol*. 2006;12:146–152.
38. Lu L, Neff F, Fischer DA, et al. Regional vulnerability of mesencephalic dopaminergic neurons prone to degenerate in Parkinson's disease: A post-mortem study in human control subjects. *Neurobiol Dis*. 2006;23:409–421.
39. Duran EM, Shapshak P, Worley J, et al. Presenilin-1 detection in brain neurons and Foxp3 in peripheral blood mononuclear cells: Normalizer gene selection for real time reverse transcriptase PCR using the $\Delta\Delta Ct$ method. *Front Biosci*. 2005;10:2955–2965.
40. Morgello S, Gelman BB, Grant E, Singer E, Vinters H, Kozlowski P. The National NeuroAIDS Tissue Consortium. *Neuropathol Appl Neurobiol*. 2001;27:326–335.
41. National NeuroAIDS Tissue Consortium. Available from: <http://spitfire.emmes.com/study/hbb/> and <http://www.nntc.org/>. Accessed May 9, 2011.
42. No authors listed. Nomenclature and research case definitions for neurologic manifestations of human immunodeficiency virus-type 1 (HIV-1) infection. Report of a Working Group of the American Academy of Neurology AIDS Task Force. *Neurology*. 1991;41:778–785.
43. CodeLink GenUS Biosystems chip. Available from: <http://www.genusbiosystems.com/>. Accessed May 9, 2011.
44. CodeLink chip information. Available from: <http://dl.dropbox.com/u/2206738/Codelink%20Human%20022311.xls.zip>. Accessed May 9, 2011.
45. National Center for Biotechnology Information. Available from: <http://www.ncbi.nlm.nih.gov/>. Accessed May 9, 2011.

46. Gene Cards. Available from: <http://www.genecards.org/>. Accessed May 9, 2011.
47. GenePro SA Biosciences. Available from: <http://gncpro.sabiosciences.com/gncpro/gncpro.php>. Accessed May 9, 2011.
48. Ariadne Pathways Assist. Available from: <http://www.ariadnegenomics.com/>. Accessed May 9, 2011.
49. Douglas NR, Reissmann S, Zhang J, et al. Dual action of ATP hydrolysis couples lid closure to substrate release into the group II chaperonin chamber. *Cell*. 2011;144:240–252.
50. Xu J, Ikezu T. The comorbidity of HIV-associated neurocognitive disorders and Alzheimer's disease: A foreseeable medical challenge in post-HAART era. *J Neuroimmune Pharmacol*. 2009;4:200–212.
51. Oldham MC, Konopka G, Iwamoto K, et al. Functional organization of the transcriptome in human brain. *Nat Neurosci*. 2008;11:1271–1282.

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