Cognitive-Behavioral Stress Management Reverses Anxiety-Related Leukocyte Transcriptional Dynamics

Supplemental Information

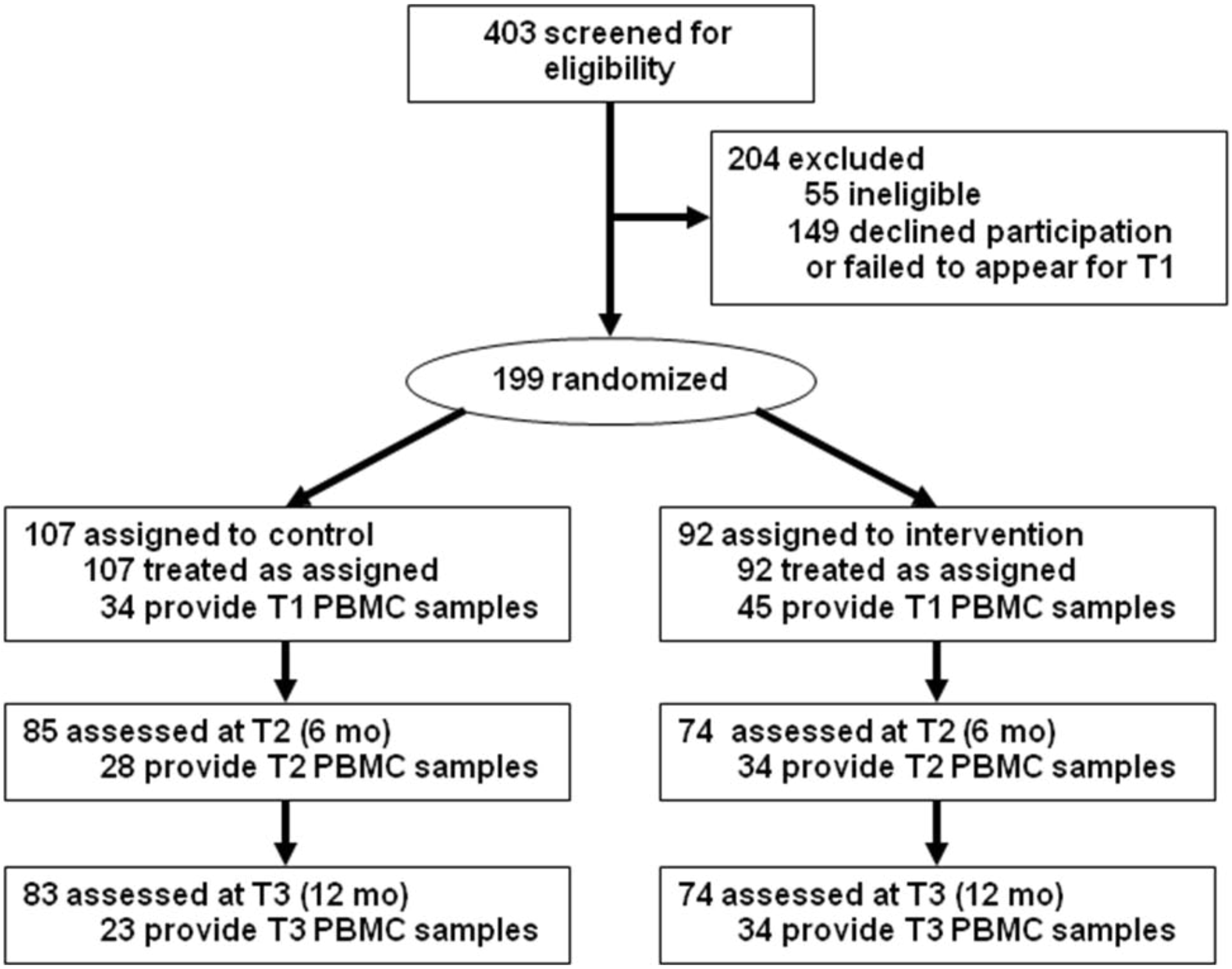

Figure S1. CONSORT Diagram. PBMC, peripheral blood mononuclear cells; T, time.

Detailed Molecular Methods

Gene Expression Profiling

Total RNA was extracted from 5-10 million PBMC, tested for suitable mass (Nanodrop ND1000; Thermo Scientific, Rockford, IL) and integrity (Bioanalyzer 2100; Agilent, Santa Clara, CA), and subject to genome-wide transcriptional profiling using Illumina Human HT-12 v3 Expression BeadChips (Illumina Inc., San Diego, CA) in the University of California Los Angeles Social Genomics Core Laboratory and the Southern California Genotyping Consortium Core Laboratory, as previously described (1, 2). Data on expression of 27,455 named human genes were quantile normalized (3) using Illumina Genome Studio software and deposited as NCBI Gene Expression Omnibus series GSE24079. Relationships between baseline affective state (ABS composite score) and expression of each analyzed transcript were assessed by analysis of covariance controlling for age, race (white vs. non-), and tumor stage, estrogen receptor (ER), and progesterone receptor (PR) status. Genes showing > 50% differential expression across the general range of ABS composite scores (i.e., ± 2 SD relative to the mean value) were identified as differentially expressed. Effects of cognitive-behavioral stress management (CBSM) on expression of each analyzed transcript were assessed in a 2 (Group: CBSM vs. Control) x 3 (Time: baseline, 6-, and 12-month follow-up) factorial design treating time as a repeated measure and controlling for individual differences in age, race, tumor stage, ER status, PR status, treatment with chemotherapy, and treatment with radiation. All analyses were conducted on an intent-to-treat basis with parameters estimated in the context of mixed effect linear models including all cases for which baseline and at least one follow-up observation were available (4). Genes showing > 50% difference across groups in the magnitude of change over time (contrast: average of 6- and 12-month follow-ups – baseline) were identified as differentially expressed (corresponding to a false discovery rate ≤ 5%) (5).

Bioinformatic Analysis

Functional characteristics of differentially expressed genes were identified by GOstat Gene Ontology analysis (5% false discovery rate) (http://gostat.wehi.edu.au) (6) and annotations from GeneCards (http://www.genecards.org/index.shtml) and EntrezGene (http://www.ncbi.nlm.nih.gov/gene). Activity of specific transcription control pathways hypothesized to mediate transcriptional responses to adversity was inferred from TELiS bioinformatic analysis of transcription factor-binding motifs (TFBMs) in the promoters of differentially expressed genes (http://www.telis.ucla.edu) (7). TELiS analyses averaged results over 9 combinations of 3 TFBM scan stringencies (mat_sim .80, .90, .95) and 3 core promoter lengths (-300, -600, and -1000 to +200 bp relative to the RefSeq-designated transcription start site) with statistical significance of differential prevalence ratios (CBSM upregulated genes / Control upregulated genes) tested relative to the null hypothesis ratio of 1.0 by single-sample *t* test (7). GATA-family TFBMs were assessed by the TRANSFAC V$GATA1_01 and V$GATA2_01 position-specific weight matrices, NF-κB/Rel-family motifs by V$CREL_01 and V$NFKB_Q6, Interferon Response Factors (IRFs) by V$IRF1_01 and V$ISRE_01, and the glucocorticoid receptor (GR) by V$GR_Q6 (8). Analyses of GR signaling controlled for concurrent serum cortisol concentrations as previously described (1) to assess GR signal transduction efficiency above and beyond the effects of CBSM in altering glucocorticoid ligand availability (9). Ancillary analyses of gene expression also controlled for flow cytometry-determined prevalence of B lymphocytes, CD4+ and CD8+ T lymphocytes, and natural killer (NK) cells assessed as previously described (9). Differential prevalence of those lymphocyte subsets was examined in 2 (Group: CBSM vs. Control) x 3 (Time: baseline, 6-, and 12-month follow-up) mixed effect linear model analysis as described above. Transcript origin analysis was employed as previously described (10) to identify the specific leukocyte subsets predominately mediating CBSM effects on the overall PBMC pool transcriptome (using single-sample *t* tests to assess over-representation of cell type-diagnostic transcripts within the list of differentially expressed genes).

Confirmatory Reverse Transcriptase-Polymerase Chain Reaction (RT-PCR)

Twelve transcripts identified as differentially expressed by microarray analysis at either 6-month follow-up, 12-month follow-up, or on average across both follow-ups were re-verified using quantitative RT-PCR with TaqMan gene expression assays (Applied Biosystems Inc., Foster City, CA), a one-step enzyme system (Quantitect RT-PCR; Qiagen, Valencia, CA), and the manufacturer's specified thermal cycling protocol on a iCycler real-time PCR instrument (BioRad Inc., Hercules, CA). Data were analyzed by standard threshold cycle analysis after normalization to parallel-assayed *ACTB* mRNA concentrations as previously described (2), using mixed effect linear model analysis to appropriately account for repeated measurements at 6- and 12-month. Several genes identified by microarray as differentially expressed only at 6- or 12-month follow-ups were evaluated by RT-PCR for differential expression across both follow-up time-points to determine whether microarray assays may have underestimated true differences in gene expression as previously observed (11).

Table S1. Genes differentially expressed in relationship to baseline affect. Gene transcripts associated with high (positive) and low (negative) Affect Balance Scale scores, as assessed by analysis of covariance controlling for age, ethnicity, tumor grade, disease stage, and tumor estrogen receptor and progesterone receptor status.

Upregulated (Positive affect-related)

Gene Symbol	**Ratio (Positive / Negative)**
HLA-DRB5	4.83
HBG2	2.92
HBG1	2.46
HBA1	2.09
SCGB3A1	2.08
HLA-DRB1	2.02
LOC731682	1.93
HBD	1.91
AHSP	1.87
ALAS2	1.77
HBM	1.68
LOC652479	1.68
CLEC2B	1.66
STRADB	1.66
HBA2	1.65
CA1	1.61
SNCA	1.59
C16orf87	1.57
LOC728105	1.54
HRK	1.53
FOLR3	1.51
LOC651309	1.51
PTGR2	1.51
RAXL1	1.51

Downregulated (Negative affect-related)

Gene Symbol	**Ratio (Positive / Negative)**
PMEPA1	0.67
RNF19B	0.67
PDCD1	0.67
PPP1R11	0.67
IL23A	0.67
KRT86	0.67
LOC440280	0.67
ACSL1	0.66
KIR2DL4	0.66
SERPINB8	0.66
RBM47	0.66
RRAD	0.66

RAB10	0.66
C14orf109	0.66
DFNA5	0.66
HIST1H2BG	0.66
PLCXD1	0.66
PDE4D	0.66
AHSA1	0.66
DEFB1	0.66
HAMP	0.66
CCL3L1	0.66
FOSB	0.66
C2orf77	0.66
NFE2L2	0.66
RAB7B	0.66
FASLG	0.66
EREG	0.66
RASD1	0.66
TNFRSF4	0.66
DNCL1	0.66
FAM43A	0.66
EMP1	0.66
CRISPLD2	0.65
CXCR6	0.65
OASL	0.65
CNOT6L	0.65
GK	0.65
TACSTD2	0.65
PVRIG	0.65
HK2	0.65
SERPINB9	0.65
SLC16A10	0.65
MIR886	0.65
STARD8	0.65
ARMET	0.65
MIR155HG	0.65
CD300A	0.65
CRLS1	0.65
ANKRD37	0.64
CASP1	0.64
RNU11	0.64
KIR2DL3	0.64
SC5DL	0.64
LOC652226	0.64
EIF4A3	0.64
TNFAIP8	0.64
MAP3K8	0.64
NEU1	0.64
RILPL2	0.64
IL1B	0.64
ATP6V1D	0.64

AVPI1	0.64
ATP6V1B2	0.64
ERO1L	0.64
SOD2	0.64
FFAR2	0.63
HES4	0.63
C3AR1	0.63
ZC3H12A	0.63
LOC728069	0.63
CHST7	0.63
DYNLL1	0.63
RNU4-2	0.63
CCL3	0.63
MRPL18	0.63
ANXA5	0.63
HSPA6	0.63
C19orf59	0.63
SLC39A8	0.63
CXCR7	0.63
BASP1	0.63
CXCL9	0.63
SAMSN1	0.63
NDRG1	0.62
TNFSF8	0.62
LOC651524	0.62
DLL1	0.62
HCP5	0.62
SH2D1B	0.62
SYTL3	0.62
IL1R1	0.62
IFIT3	0.62
IFNG	0.62
C15orf48	0.61
CD55	0.61
HIST2H2AA4	0.61
HSPA5	0.61
ADORA2A	0.61
HIST2H2AA3	0.60
LOC653610	0.60
HIST1H1C	0.60
ZNF331	0.60
CRIP2	0.60
TMEM88	0.60
IL4I1	0.60
NP	0.60
GZMB	0.60
OBFC2A	0.60
MAT2A	0.59
PRF1	0.59
CLIC3	0.59

RIN2	0.59
CXCL10	0.58
SPP1	0.58
CCL4L2	0.58
KIR3DS1	0.58
LMNA	0.58
HIST2H2AC	0.58
ELL2	0.58
TKTL1	0.57
CHST15	0.57
TREM1	0.57
ARL5B	0.57
PHACTR1	0.57
LYPD3	0.57
IL18RAP	0.56
LHFPL2	0.56
NPC1	0.56
MMP9	0.56
AQP9	0.56
PPP1R10	0.56
KCNJ2	0.56
MYOM2	0.56
CYB5D1	0.55
B4GALT5	0.55
MSC	0.55
DRAM1	0.54
ETS2	0.54
PLAUR	0.54
MIR1974	0.54
MXD1	0.54
FAM108C1	0.54
THBS1	0.54
TNF	0.54
RNU4-1	0.54
LDLR	0.54
PLA2G7	0.53
CCL7	0.53
TNFRSF21	0.52
ZC3H12C	0.52
GPR132	0.52
MS4A7	0.51
NEU4	0.49
SPSB1	0.49
OLR1	0.49
IL1RN	0.47
PLAU	0.46
IRG1	0.45
HSPA1A	0.45
PTGS2	0.44
IRAK2	0.44

RNF144B	0.44
SLC16A3	0.43
GPR84	0.42
HLA-A29.1	0.39
CCRL2	0.39
HSPH1	0.39
LRRC50	0.37
SERPINB2	0.36
GJB2	0.36
LOC730249	0.33
IL6	0.31
HSPA1B	0.29
TNFAIP6	0.28
CCL20	0.27
IL1A	0.26

Table S2. Genes differentially expressed in response to cognitive-behavioral stress management (CBSM) over follow-up (average of 6- and 12-month vs. baseline). Group differences in change over time (average of 6- and 12-month follow-up vs. baseline) estimated by mixed effect repeated measures analysis of covariance controlling for age, ethnicity, tumor grade, disease stage, tumor estrogen receptor and progesterone receptor status, use of radiation therapy, and use of chemotherapy.

Upregulated by CBSM

Gene Symbol	**Ratio (CBSM / Control)**
IFIT1	2.08
IFIT3	2.04
ISG15	1.93
SAMD9L	1.93
RSAD2	1.86
IFIT2	1.81
TNFSF10	1.76
IFI44	1.74
XAF1	1.71
OAS3	1.71
IFI44L	1.68
HERC5	1.66
LOC100008589	1.63
OAS2	1.62
LAG3	1.62
SNORA12	1.60
STAT2	1.58
C19orf66	1.56
PTGR2	1.56
IL32	1.56
GVIN1	1.55
IFNG	1.54
NCF1	1.54
RN7SK	1.52
MX2	1.52
MIR155HG	1.52
PARP12	1.51
STAT1	1.51
TRIM22	1.50

Downregulated by CBSM

Gene Symbol	**Ratio (CBSM / Control)**
SC5DL	0.66
GPR183	0.66
ABCG1	0.66
MIR302C	0.66
SPSB1	0.66

C13orf15	0.66
EMP1	0.66
KIAA1009	0.66
TNFRSF21	0.65
C3AR1	0.65
AGPAT9	0.65
GNA15	0.65
GJB2	0.64
MXD1	0.64
SLC43A2	0.64
C15orf48	0.63
TMEM158	0.63
NLRP3	0.63
MMP9	0.63
RGS1	0.63
ADORA2B	0.63
DHRS9	0.62
LOC651524	0.62
LMNA	0.61
GPR132	0.61
CXCR7	0.61
ALAS2	0.61
ASPH	0.61
LYPD3	0.61
IL6	0.59
CCL2	0.58
SLC16A6	0.58
THBD	0.58
CD300LB	0.58
CTSL1	0.57
AVPI1	0.57
ZNF331	0.57
CYP1B1	0.56
GPR84	0.56
IER3	0.55
OSM	0.55
C5AR1	0.55
HLA-A29.1	0.54
PHLDA1	0.54
CCL3L3	0.52
CCL4L2	0.52
PLAUR	0.52
G0S2	0.52
CCL7	0.51
CCL3L1	0.48
CCL3	0.48
THBS1	0.47
LOC728835	0.47
CXCL2	0.46
PTGS2	0.46

CXCL1	0.45
CCL4L1	0.41
SERPINB2	0.40
IL1A	0.40
IL1B	0.35
OLR1	0.34
CCL20	0.31

Table S3. Genes differentially expressed in cross-sectional analysis of 6-month follow-up samples. CBSM, cognitive-behavioral stress management.

Upregulated by CBSM

Gene Symbol	**Ratio (CBSM / Control)**
MIR1974	2.18
LOC649143	1.87
HLA-DRB5	1.74
TNF	1.67
IFIT3	1.65
TUBB2C	1.56
ISG15	1.56
IFI44L	1.55
LOC730249	1.54
IFIT1	1.53
LAG3	1.52

Downregulated by CBSM

Gene Symbol	**Ratio (CBSM / Control)**
EPB49	0.66
B3GNT5	0.66
LOC727908	0.66
LOC442232	0.66
Unnamed transcript	0.66
ZFP36L1	0.66
LOC643287	0.66
PTPLAD2	0.66
LOC649946	0.66
LOC347376	0.66
SH3YL1	0.65
RAP1BL	0.65
TNS1	0.65
HECA	0.65
HLA-G	0.65
LOC643534	0.65
EVI5	0.65
MYL4	0.65
LOC441454	0.65
CAPZA1	0.65
LOC642076	0.65
LOC643960	0.65
PPM2C	0.65
40428	0.64
C20orf108	0.64
LOC641849	0.64
LOC729004	0.64
PTGES3	0.64

WDR40A	0.64
FAM162A	0.64
VPS26	0.64
HBE1	0.64
LOC100131787	0.64
LOC729342	0.64
LOC729208	0.63
LOC392285	0.63
LOC728602	0.63
LOC400750	0.63
SNCA	0.63
LOC441642	0.63
LOC387934	0.63
LOC100128836	0.63
HLA-A29.1	0.63
LOC641727	0.63
LOC401640	0.62
LOC642828	0.62
LOC730746	0.62
LOC729686	0.62
KRT1	0.62
LOC402644	0.62
CD164	0.62
OSBP2	0.62
HMGB1L1	0.62
LOC648099	0.61
LOC100129657	0.61
TMEM70	0.61
LOC728060	0.61
LOC388076	0.61
NAT13	0.61
LOC100128060	0.61
LOC729687	0.61
LOC641750	0.61
LOC654103	0.61
LOC402342	0.61
KRT8P9	0.61
LOC389293	0.61
LOC100131164	0.60
STRADB	0.60
LOC729677	0.60
TLR1	0.60
LOC100130775	0.60
LOC342994	0.60
LOC442727	0.60
CCL4L1	0.60
LOC641992	0.60
LOC389599	0.60
HBG1	0.59

BCL2A1	0.59
LOC346950	0.59
HBG2	0.59
LOC729510	0.59
LOC400304	0.59
TMEM191A	0.58
LOC728791	0.58
LOC651453	0.58
C18orf32	0.58
LOC100129982	0.58
LOC731682	0.58
LOC442162	0.58
LOC729255	0.58
SELENBP1	0.58
LOC728484	0.58
LOC441154	0.58
GPR183	0.58
LOC146053	0.57
LOC100129067	0.57
LOC387753	0.57
LOC644937	0.57
LOC727821	0.57
LOC728002	0.57
LOC646093	0.57
LOC100130802	0.57
APOBEC3A	0.56
LOC100130750	0.56
LOC100130154	0.56
LOC100134504	0.56
LOC440487	0.56
LOC728207	0.56
LOC653702	0.56
SLC4A1	0.56
EPB42	0.56
LOC441896	0.55
LOC647436	0.55
LOC100130624	0.55
LOC646672	0.55
LOC400389	0.54
LOC654350	0.54
PTMA	0.54
LOC653778	0.53
Unnamed transcript	0.53
LOC730382	0.53
LOC137107	0.53
LOC729484	0.52
RPL31P10	0.52
ALAS2	0.52
LOC643870	0.52

LOC440063	0.52
LOC100132199	0.51
HBD	0.51
LOC728026	0.51
LOC391825	0.51
LOC644338	0.51
LOC727970	0.51
LOC648740	0.51
LOC646784	0.51
LOC100131675	0.51
LOC390876	0.50
LOC391655	0.50
LOC100132547	0.50
LOC391769	0.50
HBM	0.50
LOC644037	0.50
LOC100129952	0.50
LOC728572	0.49
LOC729332	0.49
AHSP	0.49
LOC645737	0.48
LOC100132804	0.48
LOC652608	0.48
LOC645693	0.48
LOC728774	0.48
LOC100127893	0.48
LOC100134540	0.47
LOC126235	0.47
LOC643007	0.47
LOC100133277	0.47
LOC645018	0.47
LOC646527	0.47
RPL23AP13	0.47
LOC392501	0.46
LOC641848	0.46
LOC100129934	0.46
LOC388344	0.46
LOC645231	0.46
LOC442270	0.45
LOC646841	0.45
CA1	0.45
LOC100131672	0.45
LOC645630	0.45
LOC390578	0.45
LOC646966	0.45
LOC728843	0.45
LOC648343	0.45
LOC729760	0.44
LOC390735	0.44

LOC100131526	0.43
LOC100129243	0.43
LOC100131160	0.43
LOC646909	0.43
LOC643779	0.43
LOC100131572	0.43
LOC390183	0.43
LOC441550	0.43
LOC647074	0.42
LOC441032	0.42
LOC642073	0.41
LOC646949	0.40
LOC100133773	0.40
LOC402677	0.39
LOC439992	0.39
LOC649839	0.39
LOC644936	0.38
LOC648659	0.38
LOC643308	0.37
LOC642567	0.37
LOC642738	0.37
LOC401717	0.35
LOC400652	0.31
LOC100133329	0.29

Table S4. Genes differentially expressed in cross-sectional analysis of 12-month follow-up samples. CBSM, cognitive-behavioral stress management.

Upregulated by CBSM

Gene Symbol	**Ratio (CBSM / Control)**
HLA-DRB5	6.56
HSPA1B	5.04
HLA-DRB1	3.62
RPPH1	2.85
HSPA1A	2.80
HBG2	2.76
ISG15	2.69
CXCL10	2.64
HBG1	2.60
LOC100132564	2.46
RGS18	2.46
SAMD9L	2.32
HSPA6	2.25
LCN2	2.24
TNFSF10	2.24
GNG11	2.24
C7orf68	2.23
LOC100008589	2.22
STAT1	2.22
RSAD2	2.18
IFIT1	2.17
IFI44L	2.15
IFNG	2.14
IFIT3	2.11
LOC85389	2.10
SDPR	2.09
HBA1	2.09
LOC650557	2.09
AHSA2	2.06
SNORD46	2.06
EPSTI1	2.01
PFKFB4	2.00
RHOB	1.99
EGR1	1.99
PLIN2	1.98
PSMB9	1.95
CYP27A1	1.95
FAM46A	1.95
STAT2	1.94
F13A1	1.94
PPBP	1.94
OAS3	1.93
SNCA	1.92

XAF1	1.92
IFI35	1.92
SH3BGRL2	1.91
DHX58	1.91
C19orf66	1.90
UCP2	1.89
OAS2	1.89
LOC85390	1.88
GBP1	1.88
NRD1	1.87
TMEM140	1.87
GSDMD	1.87
ATHL1	1.86
MYOM2	1.86
GP9	1.85
40426	1.85
MGC13057	1.85
TUBB1	1.83
IFI44	1.82
HIST1H3H	1.82
RBPMS2	1.82
EIF5A	1.81
BNIP3	1.81
CFB	1.81
VCL	1.81
CCL8	1.80
ACRBP	1.80
FOS	1.80
PF4V1	1.79
UBA7	1.79
SNORD96A	1.78
DDIT3	1.77
SELL	1.77
TKTL1	1.77
IL8RBP	1.77
JUN	1.76
HIST1H3F	1.76
LAG3	1.76
C2orf89	1.75
FAM65B	1.75
TMEM107	1.75
HBA2	1.74
ITGA2B	1.74
PRO0628	1.74
LOC100130229	1.74
HERC5	1.74
APOBEC3A	1.74
FBXO6	1.74
PRKD2	1.74
SNORD35A	1.73

C21orf7	1.73
SNORA57	1.73
PSME2	1.71
HK2	1.71
MVP	1.71
UBE2L6	1.71
HIST2H2BE	1.71
LTA	1.71
NRGN	1.70
IRF7	1.70
HSD17B4	1.70
SLAMF1	1.70
SNORD99	1.70
TRAF3IP3	1.69
LOC100132394	1.68
TPI1	1.68
SNORD71	1.68
HSH2D	1.67
HAPLN3	1.67
FGD2	1.67
C17orf62	1.66
GBP5	1.66
SPP1	1.66
MICB	1.66
GBP2	1.66
MFGE8	1.66
CD19	1.65
ENO2	1.65
TAGAP	1.65
PARP12	1.65
HSD17B8	1.65
ECH1	1.65
MGC3020	1.65
HLA-DOA	1.65
LOC653610	1.65
HSPA7	1.64
C1orf57	1.64
OBFC1	1.64
TNFSF13B	1.64
IL32	1.64
HLA-DOB	1.64
HIST1H2BK	1.64
IFIT2	1.64
MX2	1.64
SNORD83B	1.64
HSPB1	1.64
BCKDK	1.64
ASGR1	1.64
MOV10	1.64
EVL	1.63

H1FX	1.63
SPATS2L	1.63
IRF3	1.63
SNORD56	1.63
LOC54103	1.63
PCMTD1	1.63
PIM2	1.63
BCL11A	1.63
CD27	1.63
MX1	1.63
HIST2H2AC	1.63
PDK4	1.62
LOC100134364	1.62
SNORA67	1.62
HERC6	1.62
TMEM126B	1.62
CDKN2D	1.62
TNFAIP2	1.61
Unnamed transcript	1.61
LARS	1.61
RCSD1	1.61
FCHSD2	1.61
PRIC285	1.61
APBB3	1.60
P2RX7	1.60
HIST1H2AC	1.60
MBD6	1.60
LILRA5	1.60
ZBP1	1.60
PARVG	1.60
SLC25A28	1.60
LASS6	1.60
NFE2	1.59
TRAFD1	1.59
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PLCB2	1.58
PARP14	1.58
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ERO1L	1.58
ISG20	1.58
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CMTM8	1.57
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LY6E	1.57
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SNORD84	1.57
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ATF3	1.57
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SP140	1.56
TYMP	1.56
LOC648470	1.56
PABPC1L	1.56
CD69	1.56
DNAJB1	1.56
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CHKB	1.56
IFITM1	1.56
RNF31	1.56
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MPL	1.56
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USP18	1.53
IL23A	1.53
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GBP4	1.50
DRAP1	1.50
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PI3	1.50
HBQ1	1.50
C7orf59	1.50
RGL1	1.50

Downregulated by CBSM

Gene Symbol	**Ratio (CBSM / Control)**
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LOC441896	0.66
MAP3K8	0.66
LDOC1L	0.66
ARID4B	0.66
LOC646672	0.66
LOC100131160	0.66
YES1	0.66
PGRMC2	0.66
SIPA1L3	0.66
ADNP2	0.66
B3GALTL	0.66
LOC729332	0.66

PPIB	0.66
BTBD15	0.66
MIR130A	0.66
LOC729677	0.66
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MTMR6	0.66
PDXK	0.66
ANXA2P1	0.66
TGFBRAP1	0.66
CXCR6	0.66
LOC100131675	0.66
GAPDHL6	0.66
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ZNF571	0.66
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NEK6	0.65
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PTRF	0.65
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LOC100129934	0.57
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SAMSN1	0.55
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MMP9	0.53
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IL1R1	0.44
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ZNF331	0.43
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SLC7A5	0.42
CCL7	0.41
EMP1	0.41
ELL2	0.40
RPS26P10	0.40
ADORA2B	0.40
AUTS2	0.39
AGPAT9	0.39
SERPINB2	0.39
LOC731682	0.38
OLR1	0.37
C19orf59	0.36
LOC644936	0.31
CYP1B1	0.26

Table S5. RT-PCR confirmation of differential gene expression.

Transcript	**Microarray DEG***	**RT-PCR Fold-difference****	***p***
JUN	12	1.73	0.0001
FOS	12	1.49	0.0001
EGR1	12	1.21	0.0001
CD164	12	1.36	0.0001
IFIT1	6, 12, Avg 6,12	2.04	0.0001
IFIT3	6, 12, Avg 6,12	2.43	0.0001
TNF	6	1.65	0.0001
MX1	12	1.75	0.0001
IRF7	12	2.35	0.0001
STAT1	12, Avg 6,12	1.19	0.0001
ISG15	12, Avg 6,12	1.46	0.0001
*MMP9****	12, Avg 6,12	0.60	0.0001

* Differentially expressed gene (DEG) according to microarray analysis at 6 = 6-month follow-up, 12 = 12-month follow-up, Avg 6,12 = average across 6- and 12-month follow-ups.

** CBSM / Control, averaging over both follow-up timepoints.

*** Downregulated by microarray.

CBSM, cognitive-behavioral stress management; RT-PCR, reverse transcriptase-polymerase chain reaction.

CONSORT Statement - Checklist ☑

Items to include when reporting a randomized trial

PAPER SECTION **And topic**	Item	**Descriptor**	**Reported on Page #**
TITLE & ABSTRACT	1	How participants were allocated to interventions (*e.g.*, "random allocation", "randomized", or "randomly assigned").	2
INTRODUCTION Background	2	Scientific background and explanation of rationale.	4-6
METHODS Participants	3	Eligibility criteria for participants and the settings and locations where the data were collected.	7
Interventions	4	Precise details of the interventions intended for each group and how and when they were actually administered.	7 (and prior report ref 29)
Objectives	5	Specific objectives and hypotheses.	5-6
Outcomes	6	Clearly defined primary and secondary outcome measures and, when applicable, any methods used to enhance the quality of measurements (*e.g.*, multiple observations, training of assessors).	8-10
Sample size	7	How sample size was determined and, when applicable, explanation of any interim analyses and stopping rules.	11 (and prior report ref 29
Randomization -- Sequence generation	8	Method used to generate the random allocation sequence, including details of any restrictions (*e.g.*, blocking, stratification)	7 and detailed in NIH Clinical Trial NCT01422551)
Randomization -- Allocation concealment	9	Method used to implement the random allocation sequence (*e.g.*, numbered containers or central telephone), clarifying whether the sequence was concealed until interventions were assigned.	NIH Clinical Trial NCT01422551
Randomization -- Implementation	10	Who generated the allocation sequence, who enrolled participants, and who assigned participants to their groups.	Noted in ref 26 - 29
Blinding (masking)	11	Whether or not participants, those administering the interventions, and those assessing the outcomes were blinded to group assignment. If done, how the success of blinding was evaluated.	Noted in NIH Clinical Trial NCT01422551
Statistical methods	12	Statistical methods used to compare groups for primary outcome(s); Methods for additional analyses, such as subgroup analyses and adjusted analyses.	8,9
RESULTS Participant flow	13	Flow of participants through each stage (a diagram is strongly recommended). Specifically, for each group report the numbers of participants randomly assigned, receiving intended treatment, completing the study protocol, and analyzed for the primary outcome. Describe protocol deviations from study as planned, together with reasons.	11 (and diagram as supplemental Fig S1)
Recruitment	14	Dates defining the periods of recruitment and follow-up.	refs 26 and NIH Clinical Trial NCT01422551

Baseline data	15	Baseline demographic and clinical characteristics of each group.	11, 31
Numbers analyzed	16	Number of participants (denominator) in each group included in each analysis and whether the analysis was by "intention-to-treat". State the results in absolute numbers when feasible (*e.g.*, 10/20, not 50%).	8, 11
Outcomes and estimation	17	For each primary and secondary outcome, a summary of results for each group, and the estimated effect size and its precision (*e.g.*, 95% confidence interval).	11-15
Ancillary analyses	18	Address multiplicity by reporting any other analyses performed, including subgroup analyses and adjusted analyses, indicating those pre-specified and those exploratory.	N/A
Adverse events	19	All important adverse events or side effects in each intervention group.	none
DISCUSSION Interpretation	20	Interpretation of the results, taking into account study hypotheses, sources of potential bias or imprecision and the dangers associated with multiplicity of analyses and outcomes.	16 - 21
Generalizability	21	Generalizability (external validity) of the trial findings.	20
Overall evidence	22	General interpretation of the results in the context of current evidence.	16 - 20

Supplemental References

1. Cole SW, Hawkley LC, Arevalo JM, Sung CY, Rose RM, Cacioppo JT (2007): Social regulation of gene expression in human leukocytes. *Genome Biol* 8:1-13.
2. Cole S, Arevalo J, Takahashi R, Sloan EK, Lutgendorf S, Sood AK, *et al.* (2010): Computational identification of gene-social environment interaction at the human IL6 locus. *Proc Natl Acad Sci U S A* 107:5681-5686.
3. Bolstad BM, Irizarry RA, Astrand M, Speed TP (2003): A comparison of normalization methods for high density oligonucleotide array data based on variance and bias. *Bioinformatics* 19:185-193.
4. McCulloch CE, Searle SR, Neuhaus JM (2008): *Generalized, linear, and mixed models.* John Wiley & Sons: Hoboken, NJ.
5. Benjamini Y, Hochberg Y (1995): Controlling the false discovery rate: A practical and powerful approach to multiple testing. *Journal of the Royal Statistical Society, Series B* 57:289-300.
6. Beissbarth T, Speed TP (2004): GOstat: Find statistically overrepresented Gene Ontologies within a group of genes. *Bioinformatics* 20:1464-1465.
7. Cole SW, Yan W, Galic Z, Arevalo J, Zack JA (2005): Expression-based monitoring of transcription factor activity: The TELiS database. *Bioinformatics* 21:803-810.
8. Wingender E, Dietze P, Karas H, Knuppel R (1996): TRANSFAC: a database on transcription factors and their DNA binding sites. *Nucleic Acids Res* 24:238-241.
9. Antoni MH, Lechner S, Diaz A, Vargas S, Holley H, Phillips K, *et al.* (2009): Cognitive behavioral stress management effects on psychosocial and physiological adaptation in women undergoing treatment for breast cancer. *Brain Behav Immun* 23:580-591.
10. Cole SW, Hawkley LC, Arevalo JM, Cacioppo JT (2011): Transcript origin analysis identifies antigen-presenting cells as primary targets of socially regulated gene expression in leukocytes. *Proc Natl Acad Sci U S A* 108:3080-3085.

11. Cole SW, Galic Z, Zack JA (2003): Controlling false-negative errors in microarray differential expression analysis: a PRIM approach. *Bioinformatics* 19:1808-1816.