US 20090117565A1

(19) **United States**

(12) **Patent Application Publication**

Akil et al.

(10) **Pub. No.: US 2009/0117565 A1**

(43) **Pub. Date: May 7, 2009**

(54) **COMPOSITIONS AND METHODS FOR DIAGNOSIS AND TREATING MOOD DISORDERS**

(75) Inventors: **Huda Akil**, Ann Arbor, MI (US); **William E. Bunney, JR.**, Laguna Beach, CA (US); **Prabhakara V. Choudary**, Davis, CA (US); **Simon J. Evans**, Milan, MI (US); **Edward G. Jones**, Winters, CA (US); **Jun Li**, Palo Alto, CA (US); **Juan F. Lopez**, Ann Arbor, MI (US); **Robert C. Thompson**, Ann Arbor, MI (US); **Richard Myers**, Stanford, CA (US); **Hiroaki Tomita**, Irvine, CA (US); **Marquis P. Vawter**, Niguel, CA (US); **Stanley Watson**, Ann Arbor, MI (US)

Correspondence Address:
TOWNSEND AND TOWNSEND AND CREW, LLP
TWO EMBARCADERO CENTER, EIGHTH FLOOR
SAN FRANCISCO, CA 94111-3834 (US)

(73) Assignee: **The Board of Trustees of the Leland Stanford Junior University**, Palo Alto, CA (US)

(21) Appl. No.: **12/154,419**

(22) Filed: **May 21, 2008**

Related U.S. Application Data

(63) Continuation of application No. 10/701,263, filed on Nov. 3, 2003, now Pat. No. 7,410,759.

(60) Provisional application No. 60/431,454, filed on Dec. 6, 2002, provisional application No. 60/423,247, filed on Nov. 1, 2002.

Publication Classification

(51) **Int. Cl.**

C12Q 1/68	(2006.01)
A61K 38/02	(2006.01)

(52) **U.S. Cl.** **435/6**

(57) **ABSTRACT**

The present invention provides methods for diagnosing mental disorders such as mood disorders, including bipolar disorder I and II and major depression; The invention also provides methods of identifying modulators of such mental disorders as well as methods of using these modulators to treat patients suffering from such mental disorders.

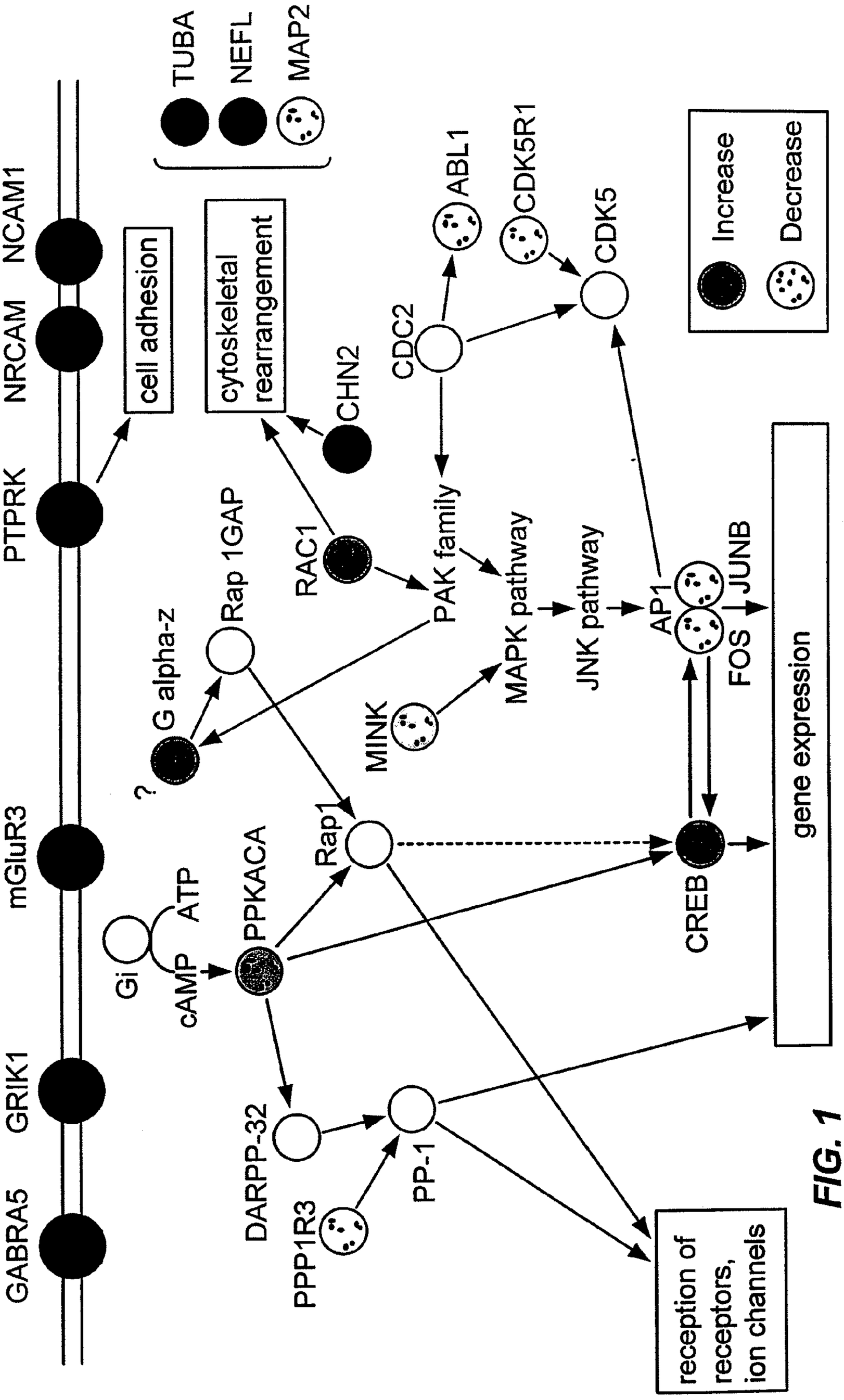

TUBA
NEFL
MAP2
NRCAM
NCAM1
cell adhesion
cytoskeletal rearrangement
CHN2
CDC2
ABL1
CDK5R1
CDK5
Increase
Decrease
PTPRK
Rap 1GAP
RAC1
PAK family
MAPK pathway
JNK pathway
AP1
FOS
JUNB
G alpha-z
?
MINK
mGluR3
Gi
ATP
cAMP
PPKACA
Rap1
CREB
gene expression
GRIK1
GABRA5
DARPP-32
PPP1R3
PP-1
reception of receptors, ion channels

FIG. 1

Summary of Functions for Signal Transduction Transcripts Differentially Expressed in MDD

- **GRB2 (growth receptor binding protein 2).** An adaptor protein that transduces signal from an activated growth factor receptor, culminating in the activation of Ras.

- **ITPKB (inositol-1,4,5-triphosphate-3 kinase B).** A member of a family of kinases involved in inositol triphosphate signal transduction.

- **PAK-1 (p21 activated kinase 1).** A protein that is regulated by small-GTP binding proteins, like Ras, and is believed to directly act on the JNK1/MAP kinase signal transduction cascade.

- **PKC, beta1 (protein kinase C, beta 1).** A protein kinase involved in several Ca^{++} and IP3 dependent signal transduction cascades.

- **RalGDS (Ral guanine nucleotide dissociation stimulator).** Stimulates dissociation of GDP from Ras and Ras-like (Ral) small G-proteins, thereby, increasing the rate of GTP-GDP exchange and facilitating activation of Ras/Ral proteins.

- **RAP1 (GTPase activating protein 1).** Stimulates GTPase activity of small GTP- binding proteins, thereby terminating their activity and functionally inhibiting Ras/Ral proteins.

- **RGS20 (regulator of G-protein signaling 20).** A GTPase activating protein that has specificity for the alpha z subunit of heterotrimeric G-proteins. Facilitation of GTPase activity would terminate signal transduction and thus act as a negative regulator.

FIG. 2

bFGF System Transcripts are Differential Expressed in PFC in Subjects with MDD

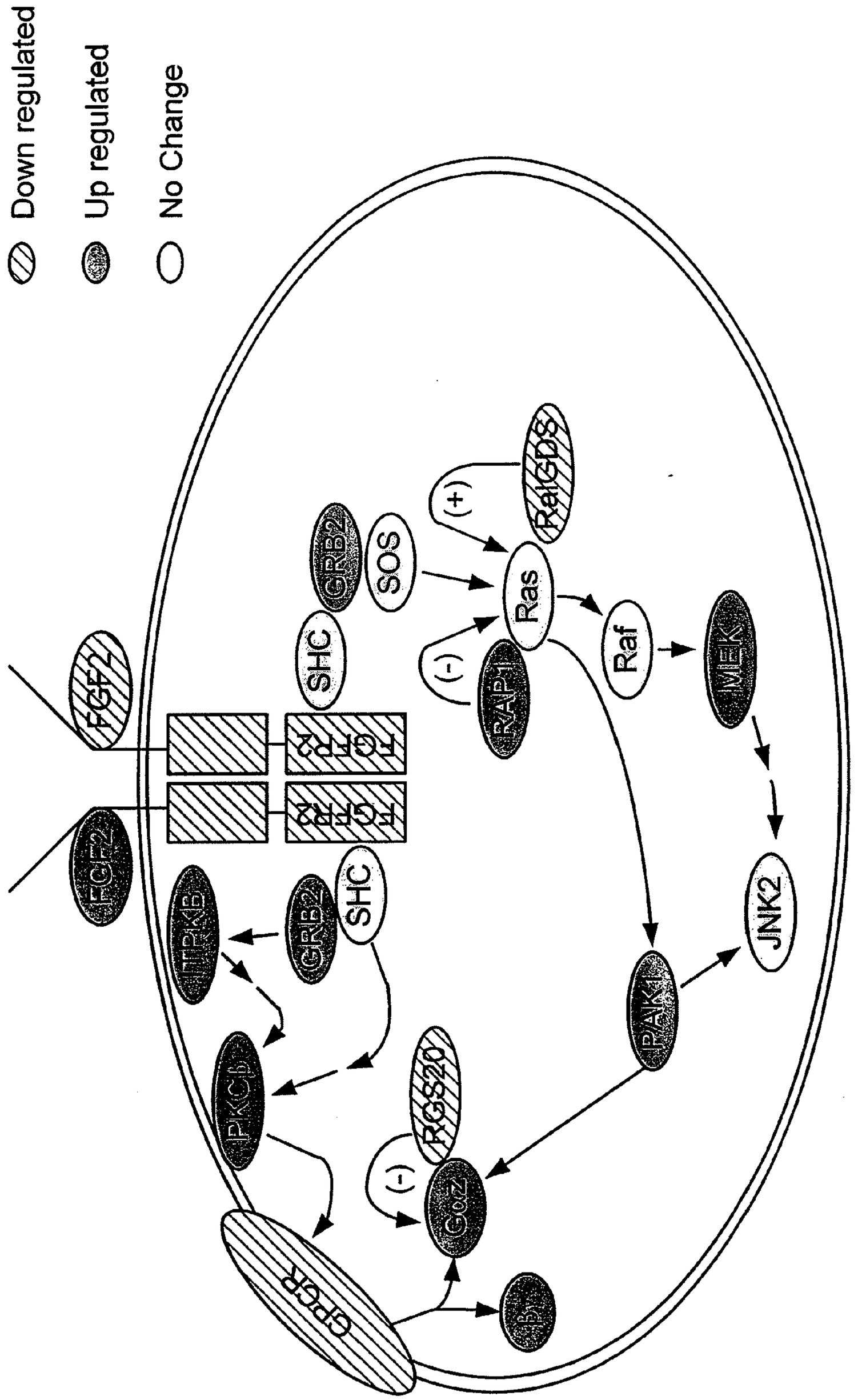


FIG. 3

Fold Change and p-values for bFGF system Transcripts in PFC for MDD Subjects

probe set ID	Gene	Accession #	Fold Change	p-value
1593_at	FGF2	JO4513	-1.4	0.04
1363_at	FGFR2	M87770	-1.4	0.02
1143_s_at	FGFR2	M87771	-1.2	0.04
33855_at	GRB2	M96995	-1.2	0.06
37272_at	ITPKB	X57206	-1.4	0.05
1557_at	PAK-1	U24152	-1.4	0.01
1558_g_at	PAK-1	U24152	-1.6	0.05
1217_g_at	PKC beta 1	X07109	-1.6	0.02
36550_at	RalGDS	ALO49538	-1.5	0.01
1270_at	RAP1	M64788	-1.2	0.06
41086_at	RGS 20	AF060877	-1.6	0.04

FIG. 4

Metabolic pathways significantly dysregulated

Pathway	GS/GOC	possibility of false positive
oxidative phosphorylation	8/95	0.0017
terpenoid biosynthesis	2/5	0.0057
proteasome	3/30	0.0360
ATP synthesis	3/32	0.0425
sterol, vitamin K/E, carotenoid biosynthesis	2/17	0.0643
Parkinson's disease	2/21	0.0934
sphingophospholipid	1/4	0.0955
nitrogen metabolism	2/23	0.1091
galactose metabolism	2/24	0.1172
glutathion metabolism	2/32	0.1866
glycine, serine, threonine metabolism	2/33	0.1957

GS/GOC; genes selected/genes on a chip

FIG. 5

Signaling & Metabolic Pathways Significantly Altered in AnCg of Bipolar Subjects

• 1.	Inositol Phosphate Metabolism	14/135
• 2.	Nicotinate and Nicotimade Metabolism	11/116
• 3.	Benzoate Degradation N-Ac CoA Ligation	11/118
• 4.	Alanine and Aspartate Metabolism	4/18
• 5.	Starch and Sucrose Metabolism	11/132
• 6.	Sphingoglycolipid Metabolism	11/134
• 7.	Glutamate Metabolism	4/21
• 8.	Phosphotydil-Isositol Signaling	8/101

FIG. 6

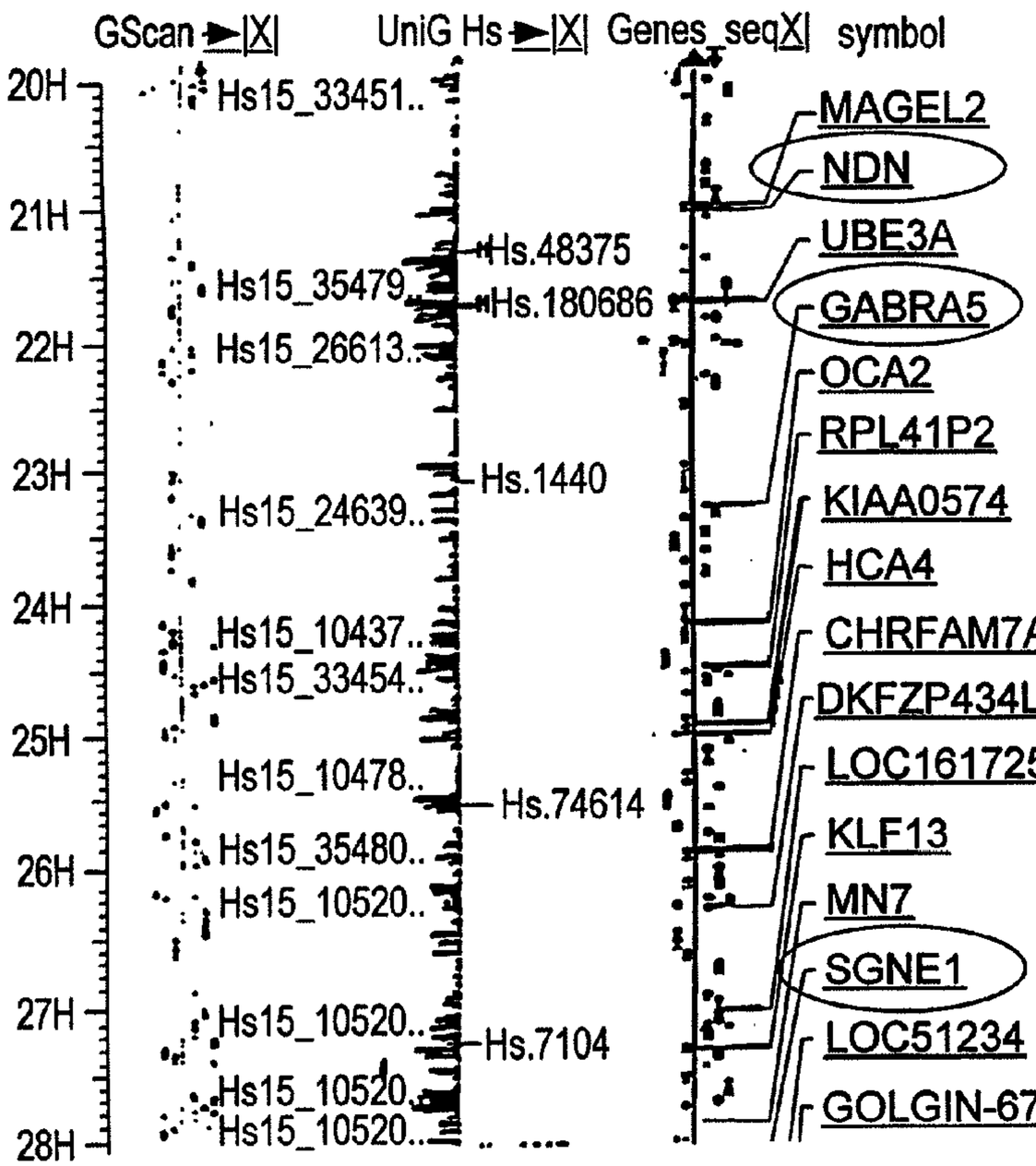
Three Over Expressed Genes Are Located in the Same Chromosomal Region

Symbol	Description	Cytogenetic Band (Mb)
SGNE1	Secretory granule, neuroendocrine protein 1 (7B2 protein)	15q13
NDN	Necdin homolog (mouse)	15q11.2
GABRA5	Gamma-aminobutyric acid (GABA) A receptor, alpha 5	15q11.2

FIG. 7

FIG. 8

Three of the 13 genes are on 15q11-13 within the Prader Willi region (SGNE1, GABRA5, and NDN)



orient.	links	evidence	cyto.	full name
▲	sv ev hm seq mm	C	15q11-q12	MAGE-like 2
▲	sv ev hm seq mm	C	15q11.2-q12	necdin homolog (mouse)
▲	sv ev hm seq mm	C	15q11-q12	ubiquitin protein ligase E3A (h
▼	sv ev hm seq mm	C	15q11.2-q12	gamma-aminobutyric acid (GA
▲	sv ev hm seq mm	C	15q11.2-q12	oculocutaneous albinism II (pi
▼	sv ev - seq mm		15q11-q12	ribosomal protein L41, pseud
▲	sv ev - seq mm	C	15q12	KIAA0574 protein
▲	sv ev - seq mm	C	15q12	hepatocellular carcinoma-asso
▲	sv ev - seq mm	C	15q14	CHRNA7 (cholinergic recept
▼	sv ev - seq mm	C	15q13.1	DKFZP434L187 protein
▼	sv ev - seq mm	C	15q12	cezanne 2
▲	sv ev hm seq mm	C	15q12	Kruppel-like factor 13
▼	sv ev - seq mm	C	15q11-q13	D15F37 (pseudogene)
▼	sv ev hm seq mm	?	15q13-q14	secretory granule, neuroendoc
▼	sv ev hm seq mm	C	15q13.1	hypothetical protein LOC512
▲	sv ev - seq mm	C	15q11.2	golgin-67

These 3 genes are found within a 7 Mb region that contains 10 other genes.

Genes Regulated in Human Postmortem Tissue (Dorsolateral PFC and Anterior Cingulate)

Gene	MDD DLPC	MDD AnCg	BD DLPC	BD AnCg
neuronal cell adhesion molecule	↑			
solute carrier family 14 (urea transporter) member	↓			
protein kinase C, beta 1	↑	↑		
NEL-like 1	↑			
phosphoribosyl pyrophosphate synthetase	↑			
solute carrier family 1(glial high affinity glutamate transporter) member		↓		
glutamate receptor, metabotropic 3				↑
Microtubule Associated Protein 2			↑	
Fribroblast Growth Factor 2		↓		
CABA A Receptor				↓
Inositol 1,4,5-triphosphate receptor		↑		

FIG. 9

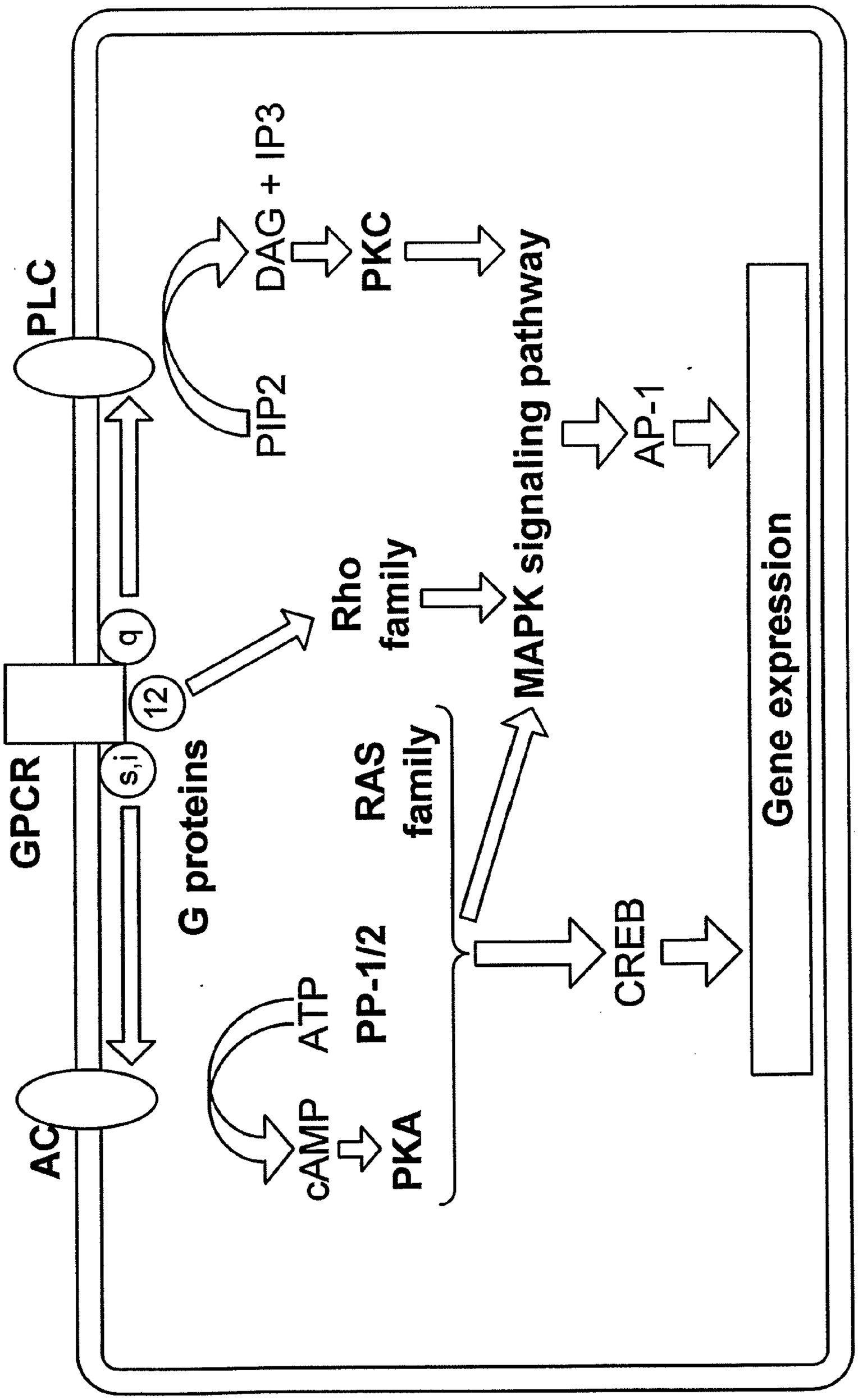

GRCR and cAMP/PI/Rho pathways
GPCR
s,i
q
12
G proteins
AC
PLC
DAG + IP3
PKC
PIP2
Rho family
RAS family
MAPK signaling pathway
AP-1
CREB
Gene expression
ATP
PP-1/2
cAMP
PKA

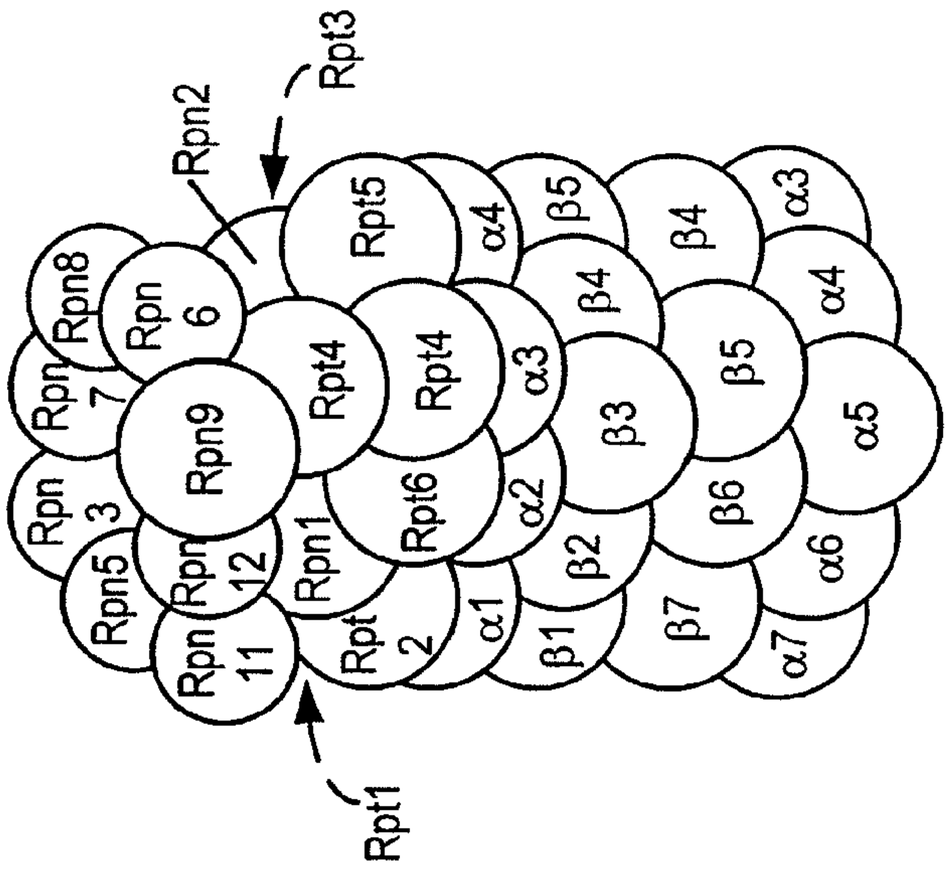
FIG. 10

Proteasome: Alterations in In Bipolar Ss, Anterior Cingulate

-28 subunits
-Four rings of seven.
-Outer rings: alpha subunits
-Inner Rings: beta subunits
-Beta: responsible for peptidase activity.
-Degrades Short-lived and Misfolded Proteins
-Role in Ubiquitination
-ATP Dependent Activity

Note: 11/31 genes are significant, mostly Beta

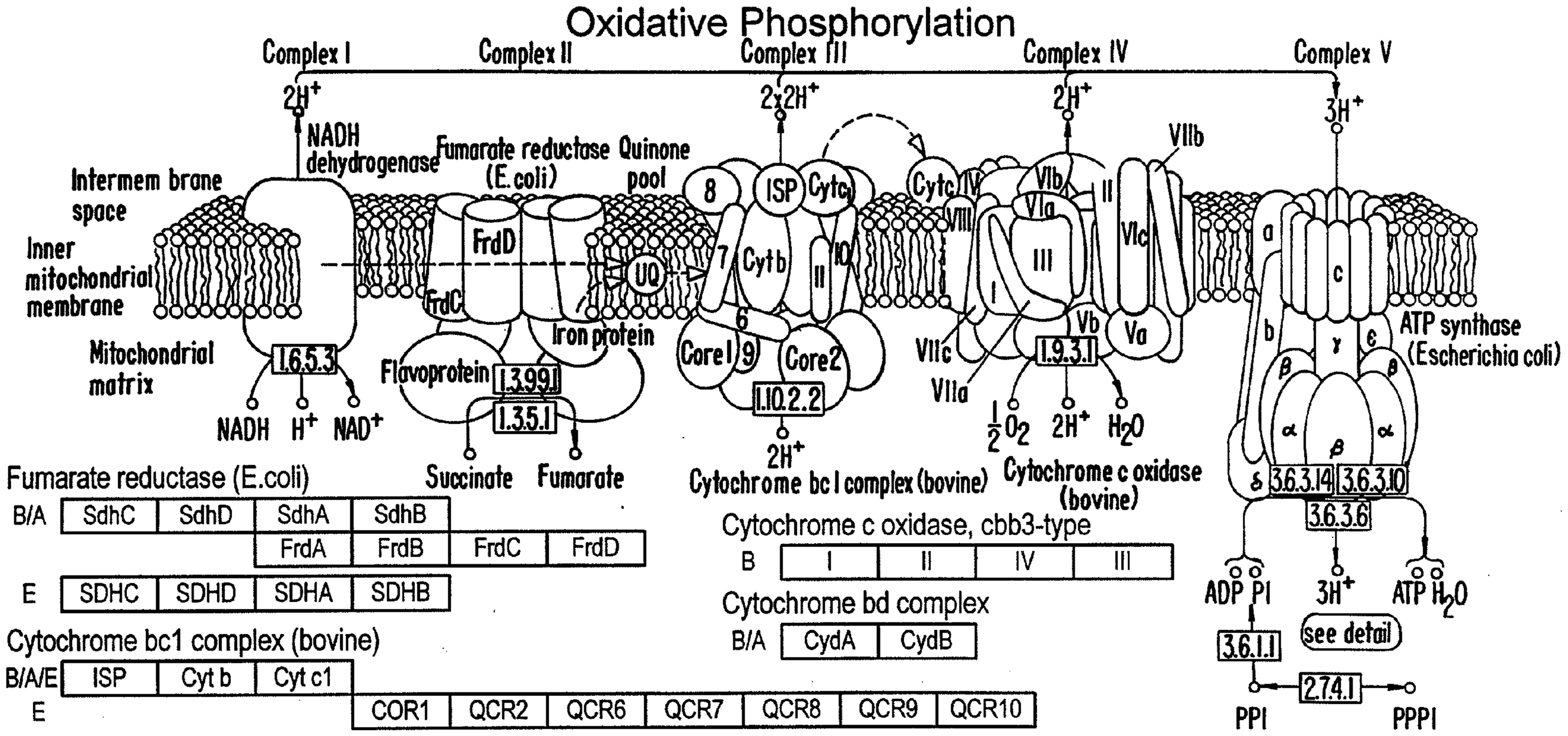
PROTEASOME



26S Proteasome (Saccharomyces cerevisiae)

Rpn1	Rpn2	Rpn3	Rpn4	Rpn5	Rpn6
Rpn7	Rpn8	Rpn9	Rpn10	Rpn11	Rpn12
Rpt1	Rpt2	Rpt3	Rpt4	Rpt5	Rpt6
α1	α2	α3	α4	α5	α6
β1	β2	β3	β4	β5	β6
					α7
					β7

FIG. 11



Fumarate reductase (E.coli)

B/A	SdhC	SdhD	SdhA	SdhB		
			FrdA	FrdB	FrdC	FrdD
E	SDHC	SDHD	SDHA	SDHB		

Cytochrome bc1 complex (bovine)

B/A/E	ISP	Cyt b	Cyt c1					
E	COR1	QCR2	QCR6	QCR7	QCR8	QCR9	QCR10	

Cytochrome c oxidase

E	COX10		COX3	COX1	COX2	COX4	COX5A	COX5B	COX6A	COX6B	COX6C	COX7A	COX7B	COX7C	COX8
B/A	CyoE	CyoD	CyoC	CyoB	CyoA										
		CoxD	CoxC	CoxA	CoxB										
		QoxD	QoxC	QoxB	QoxA										

Cytochrome c oxidase, cbb3-type

B	I	II	IV	III

Cytochrome bd complex

B/A	CydA	CydB

ATP5A1; ATP6V1A1; ATP6VOE; ATP6VOENDUFV1; ATP6V1E1; ATP5H; ATP5J; ATP6V0B; ATP5O; ATP5J2; ATP6VOD1
NDUFS7; NDUFB5; NDUFS2; NDUFC1; NDUFB3
COX7B; SDHB; UQCRC2

FIG. 12

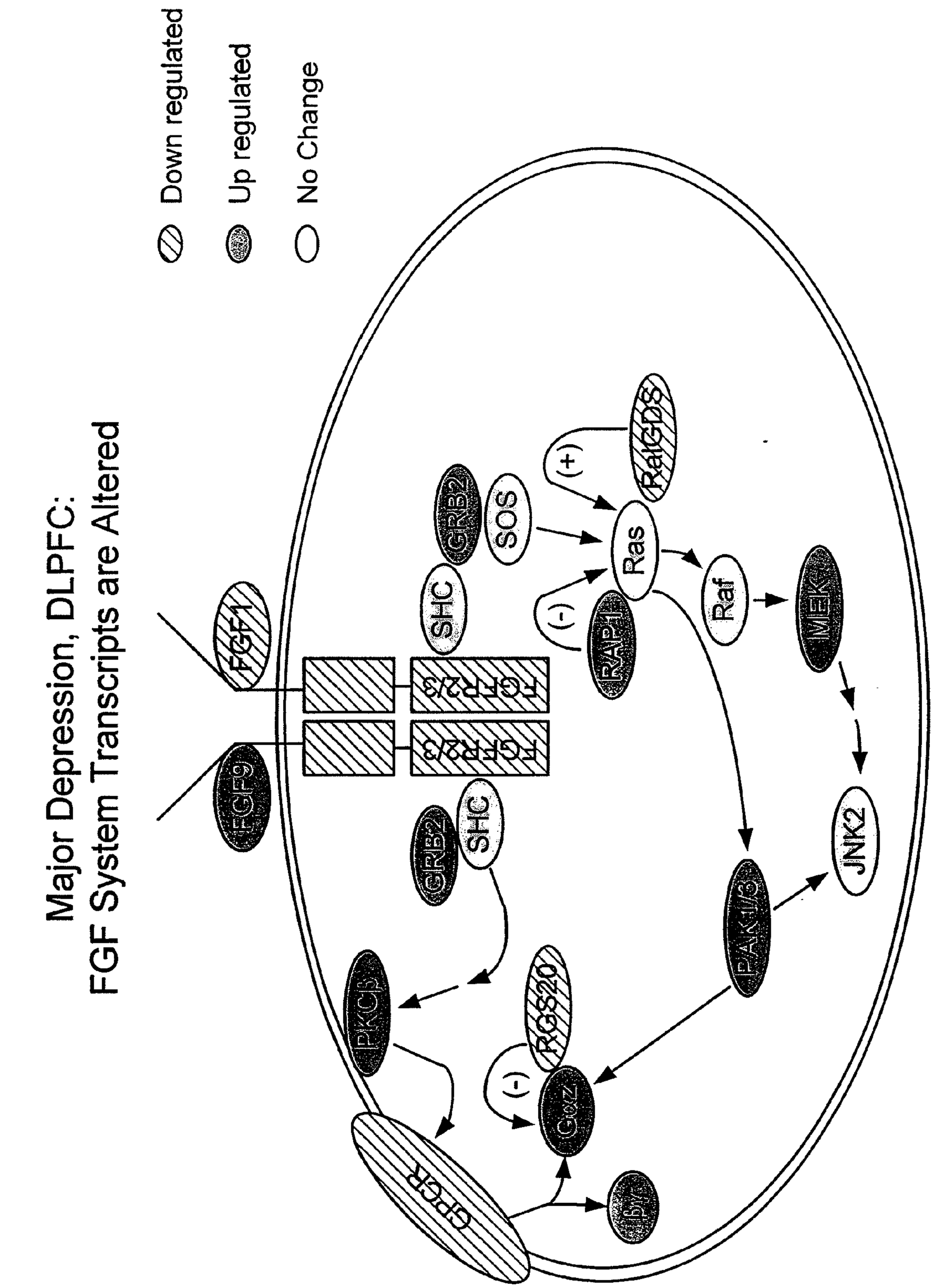

Down regulated
Up regulated
No Change
Major Depression, DLPFC:
FGF System Transcripts are Altered
FGF1
FGF9
FGFR2/3
FGFR2/3
GRB2
SHC
GRB2
SOS
SHC
(+)
RalGDS
(-)
RAP1
Ras
Raf
MEK
JNK2
PAK1/3
PKCβ
RGS20
(-)
GαZ
GPCR
Βγ

FIG. 13

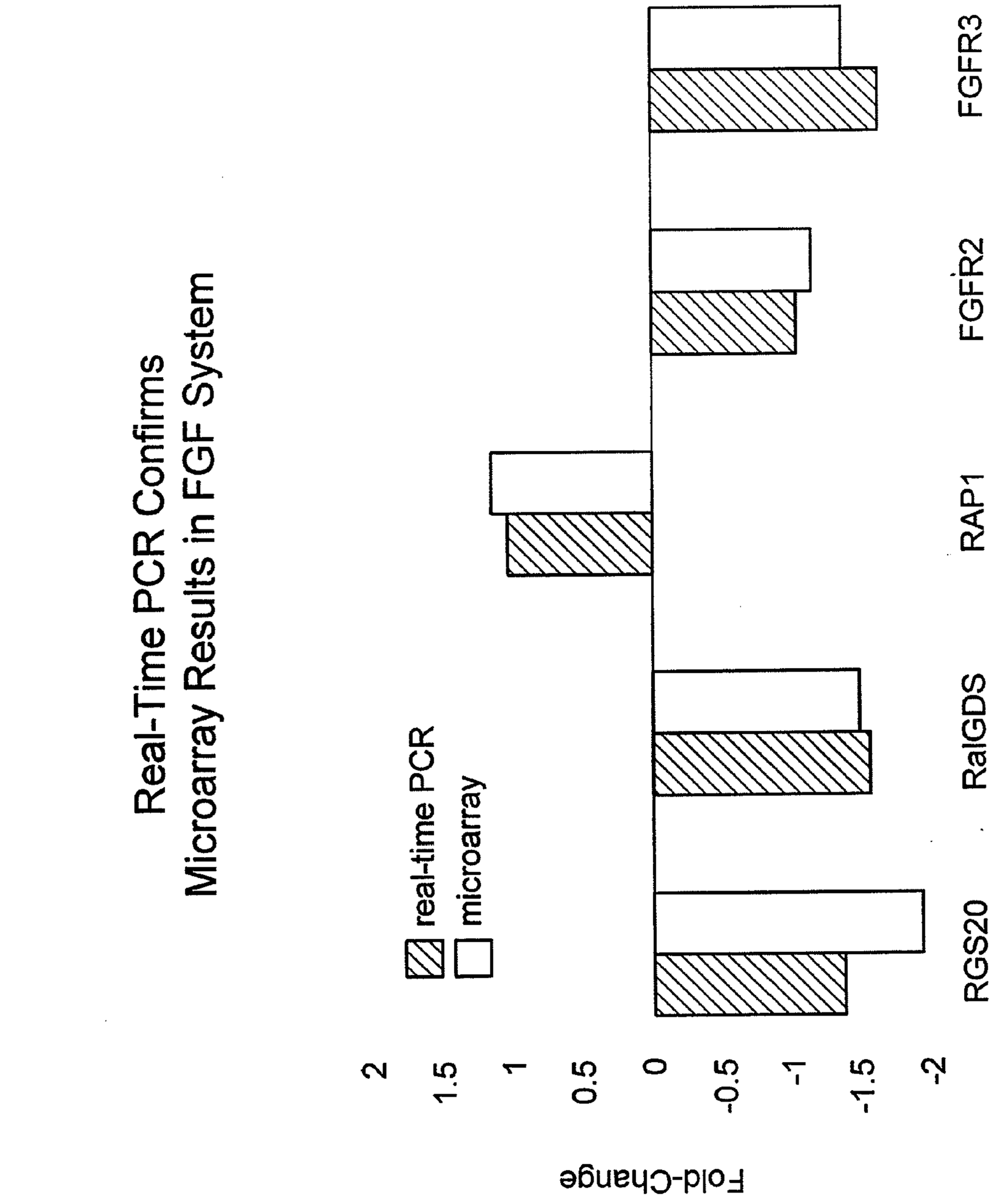

Real-Time PCR Confirms
Microarray Results in FGF System
real-time PCR
microarray
Fold-Change
2
1.5
1
0.5
0
-0.5
-1
-1.5
-2
RGS20
RalGDS
RAP1
FGFR2
FGFR3

FIG. 14

Signaling Pathways involving GABA & Glutamate

BPD_AnCg

- GABRA5 GABA A Receptor, alpha 5
- GRIA1 Glut Receptor, ionotropic, AMPA1
- GRIM3 Glut Receptor, metabotropic 3
- GRIA3 Glut Receptor, ionotropic, AMPA3
- GRIK1 Glut Receptor, ionotropic, kainate1

BPD_DLPFC

- GABRA5 GABA A Receptor, alpha 5
- GABBR1 GABA B Receptor 1
- GABARAPL2 GABA (A) Receptor-assoc. protein-like 2
- **(None Down)**

MDD_AnCg

- **(None UP)**
- SLC1A3 Solute carrier family 1 (glial high affinity glut transporter), member 3
- SLC1A2 Solute carrier family 1 (glial high affinity glut transporter), member 2
- GLUL Glutamate-ammonia ligase (glutamine synthase)

MDD_**DLPFC**

- GABARB2 GABA A receptor, beta 3
- GABARG2 GABA A receptor, gamma 2
- GRIA1 Glut Receptor, ionotropic, AMPA1
- GRIK5 Glut Receptor, ionotropic, kainite 5
- SLC1A3 Solute carrier family 1 (glial high affinity glut transporter), member 3
- SLC1A2 Solute carrier family 1 (glial high affinity glut transporter), member 2
- GLUL Glutamate-ammonia ligase (glutamine synthase)

Black: Up-regulated; Red: Down-regulated

FIG. 15

COMPOSITIONS AND METHODS FOR DIAGNOSIS AND TREATING MOOD DISORDERS

CROSS-REFERENCES TO RELATED APPLICATIONS

[0001] The present application claims the benefit of U.S. Ser. No. 60/423,247, filed Nov. 1, 2002 and U.S. Ser. No. 60/431,454, filed Dec. 6, 2002, the disclosures of which are hereby incorporated by reference in their entirety for all purposes.

STATEMENT AS TO RIGHTS TO INVENTIONS MADE UNDER FEDERALLY SPONSORED RESEARCH AND DEVELOPMENT

[0002] Not applicable.

BACKGROUND OF THE INVENTION

[0003] Clinical depression, including both bipolar disorders and major depression disorders, is a major public health problem, affecting an estimated 9.5% of the adult population of the United States each year. While it has been hypothesized that mental disorders, including mood disorders such as major depression and bipolar disorder as well as psychotic disorders such as schizophrenia, have complex genetic roots, little progress has been made in identifying gene sequences and gene products that play a role in causing these disorders, as is true for many diseases with a complex genetic origin (see, e.g. Burmeister, *Biol. Psychiatry* 45:522-532 (1999)). Relying on the discovery that certain genes expressed in particular brain pathways and regions are likely involved in the development of mental disorders, the present invention provides methods for diagnosis and treatment of mental disorders, as well as methods for identifying compounds effective in treating mental disorders.

BRIEF SUMMARY OF THE INVENTION

[0004] In order to further understand the neurobiology of mood disorders such as bipolar disorders (BP) and major depression disorders (MDD), the inventors of the present application have used DNA microarrays to study expression profiles of human post-mortem brains from patients diagnosed with BP or MDD. The work has focused on three brain regions: the anterior cingulated cortex (AnCg), the dorsolateral prefrontal cortex (DLPFC), and the cerebellum (CB).

[0005] The present invention demonstrates, for the first time, differential expression of the 72 nucleic acids listed in Table 2, the 16 nucleic acids listed in Table 3, or the 967 nucleic acids listed in Table 4, in the brains of patients suffering from mood disorders, such as bipolar disorder and major depression disorder, in comparison with normal control subjects. In addition, the present invention identifies biochemical pathways involved in mood disorders, where the proteins encoded by the nucleic acids listed in Table 2, 3, or 4 are components of the biochemical pathways (e.g., the bFGF signal transduction pathway, the GPCR and cAMP/PI/Rho pathways, the proteasome pathway, the oxidative phosphorylation pathway, Myelination, Cytochrome P450, or the GABA and glutamate pathways; see also FIGS. **1-5**, **10-13**, and **15**).

[0006] Finally, genes that are differentially expressed in MDD or BP and by gender are useful in diagnosing mood disorders, as the prevalence of certain mood disorders shows a gender bias. Differential expression by brain region similarly is a useful diagnostic and therapeutic tool, as certain mood disorders primarily affect certain brain regions.

[0007] This invention thus provides methods for determining whether a subject has or is predisposed for a mental disorder such as bipolar disorder or major depression disorder. The invention also provides methods of providing a prognosis and for monitoring disease progression and treatment. Furthermore, the present invention provides nucleic acid and protein targets for assays for drugs for the treatment of mental disorders such as bipolar disorder and major depression disorder.

[0008] In some embodiments, the methods comprise the steps of: (i) obtaining a biological sample from a subject; (ii) contacting the sample with a reagent that selectively associates with a polynucleotide or polypeptide encoded by a nucleic acid that hybridizes under stringent conditions to a nucleotide sequence listed in Table 2, 3 or 4; and (iii) detecting the level of reagent that selectively associates with the sample, thereby determining whether the subject has or is predisposed for a mental disorder.

[0009] In some embodiments, the reagent is an antibody. In some embodiments, the reagent is a nucleic acid. In some embodiments, the reagent associates with a polynucleotide. In some embodiments, the reagent associates with a polypeptide. In some embodiments, the polynucleotide comprises a nucleotide sequence of a gene listed in Table 2, 3, or 4. In some embodiment, the polypeptide comprises an amino acid sequence of a gene listed in Table 2, 3, or 4. In some embodiments, the level of reagent that associates with the sample is different (i.e., higher or lower) from a level associated with humans without a mental disorder. In some embodiments, the biological sample is obtained from amniotic fluid. In some embodiments, the mental disorder is a mood disorder. In some embodiments, the mood disorder is selected from the group consisting of bipolar disorder and major depression disorder.

[0010] The invention also provides methods of identifying a compound for treatment of a mental disorder. In some embodiments, the methods comprises the steps of: (i) contacting the compound with a polypeptide, which is encoded by a polynucleotide that hybridizes under stringent conditions to a nucleic acid comprising a nucleotide sequence of Table 2, 3, or 4; and (ii) determining the functional effect of the compound upon the polypeptide, thereby identifying a compound for treatment of a mental disorder.

[0011] In some embodiments, the contacting step is performed in vitro. In some embodiment, the polypeptide comprises an amino acid sequence of a gene listed in Table 2, 3, or 4. In some embodiments, the polypeptide is expressed in a cell or biological sample, and the cell or biological sample is contacted with the compound. In some embodiments, the mental disorder is a mood disorder or psychotic disorder. In some embodiments, the mood disorder is selected from the group consisting of bipolar disorder I and II and major depression. In some embodiments, the psychotic disorder is schizophrenia. In some embodiments, the methods further comprise administering the compound to an animal, e.g., an animal subjected to stress as a model for depression and determining the effect on the animal, e.g., an invertebrate, a vertebrate, or a mammal. In some embodiments, the determining step comprises testing the animal's mental function.

[0012] In some embodiments, the methods comprise the steps of (i) contacting the compound to a cell, the cell com-

prising a polynucleotide that hybridizes under stringent conditions to a nucleotide sequence of Table 2, 3, or 4; and (ii) selecting a compound that modulates expression of the polynucleotide, thereby identifying a compound for treatment of a mental disorder. In some embodiments, the polynucleotide comprises a nucleotide sequence listed in Table 2, 3, or 4. In some embodiment, the expression of the polynucleotide is enhanced. In some embodiments, the expression of the polynucleotide is decreased. In some embodiments, the methods further comprise administering the compound to an animal and determining the effect on the animal. In some embodiments, the determining step comprises testing the animal's mental function. In some embodiments, the mental disorder is a mood disorder or psychotic disorder. In some embodiments, the mood disorder is selected from the group consisting of bipolar disorder I and II and major depression. In some embodiments, the psychotic disorder is schizophrenia.

[0013] The invention also provides methods of treating a mental disorder in a subject. In some embodiments, the methods comprise the step of administering to the subject a therapeutically effective amount of a compound identified using the methods described above. In some embodiments, the mental disorder is a mood disorder or psychotic disorder. In some embodiments, the mood disorder is selected from the group consisting of bipolar disorder I and II and major depression. In some embodiments, the psychotic disorder is schizophrenia. In some embodiments, the compound is a small organic molecule, an antibody, an antisense molecule, aptamer, or a peptide.

[0014] The invention also provides methods of treating mental disorders in a subject, comprising the step of administering to the subject a therapeutically effective amount of a polypeptide, which is encoded by a polynucleotide that hybridizes under stringent conditions to a nucleic acid of Table 2, 3, or 4. In some embodiments, the polypeptide comprises an amino acid sequence encoded by a gene listed in Table 2, 3, or 4. In some embodiments, the mental disorder is a mood disorder or psychotic disorder. In some embodiments, the psychotic disorder is schizophrenia. In some embodiments, the mood disorder is a bipolar disorder or major depression.

[0015] The invention also provides methods of treating mental disorders in a subject, comprising the step of administering to the subject a therapeutically effective amount of a polynucleotide, which hybridizes under stringent conditions to a nucleic acid of Table 2, 3, or 4. In some embodiments, the mental disorder is a mood disorder or psychotic disorder. In some embodiments, the mood disorder is a bipolar disorder or major depression.

BRIEF DESCRIPTION OF THE DRAWINGS

[0016] Table 1: Table 1 lists genes differentially expressed in mood disorder subjects.

[0017] Table 2: Table 2 lists 72 genes differentially expressed in mood disorder subjects.

[0018] Table 3: Table 3 lists 16 genes differentially expressed in specific brain regions and mood disorder.

[0019] Table 4: Table 4 lists 967 genes differentially expressed in mood disorder subjects as determined by microarray analysis. Flag 1 indicates that the differential expression of the gene was confirmed by Real time PCR. Flag 2 indicates that differential expression of the gene was confirmed by anti-depressant studies. Flag 3 indicates that the gene belongs to an enriched gene ontology. Up and down indicates the direction of the changes compared to controls.

[0020] Table 5: Table 5 lists Real time PCR results on sample genes that are differentially expressed in mood disorder subjects.

[0021] Table 6: Table 6 lists anti-depressant treatment results for genes that are differentially expressed in mood disorder subjects.

[0022] Table 7: Tables 7A-D lists the gene ontology of selected genes differentially expressed in mood disorder subjects.

[0023] Table 8: Table 8 lists sample of genes that are differentially expressed in mood disorder subjects and are potential druggable targets.

[0024] FIG. **1** shows selected biochemical pathways for genes differentially expressed in mood disorder subjects.

[0025] FIG. **2** summarizes functions for signal transduction transcripts differentially expressed in MDD subjects.

[0026] FIG. **3** shows bFGF pathway transcripts differentially expressed in MDD subjects.

[0027] FIG. **4** shows values for differential expression of bFGF transcripts in MDD subjects.

[0028] FIG. **5** shows selected biochemical pathways that are dysregulated in mood disorders.

[0029] FIG. **6** shows selected biochemical pathways that are dysregulated in BP subjects.

[0030] FIG. **7** shows three genes overexpressed in mood disorder subjects that are located in the same chromosomal region.

[0031] FIG. **8** shows three genes overexpressed in mood disorder subjects that are located on 15q11-13 in the Prader-Willi region.

[0032] FIG. **9** shows certain genes regulated in human postmortem tissue and by antidepressants in rats.

[0033] FIG. **10** shows selected biochemical pathways (i.e., the GPCR and cAMP/PI/Rho pathways) for genes differentially expressed in mood disorder subjects. Two G protein coupled receptors, GPR37 and GPRC5B, are increased in both AnCg and DLPFC of BP patients, and decreased in MD. As downstream signaling pathways of GPCR, genes involved in cAMP pathway signaling are increased n BP patients, and decreased in MD. Genes involved in phosphatidylinositol pathways are deregulated specifically in MD.

[0034] FIG. **11** shows a selected biochemical pathway (i.e., the proteasome pathway) for genes differentially expressed in mood disorder subjects. The proteasome is an assembly of 28 alpha and beta subunits that functions to degrade proteins. The proteasome is involved in regulation of protein turnover and in particular oxidized proteins. There is an over representation of proteasome genes found in cortical regions of BP, but not in the cerebellum, suggesting that some functional compensation in the proteasome is occurring in BP patients.

[0035] FIG. **12** shows a selected biochemical pathway (i.e., the oxidative phosphorylation pathway) for genes differentially expressed in mood disorder subjects. The oxidative phosphorylation classification is involved in bioenergetics, metabolism, and as a byproduct can produce reactive oxygen species. This pathway is overly expressed in both bipolar and major depression, with differences between cortical regions and cerebellum.

[0036] FIG. **13** shows an example of a growth factor system (e.g., FGF) that is altered in mood disorders.

[0037] FIG. **14** shows RealTime PCR results which confirm that selected FGF-related genes first identified using microarray analysis are differentially expressed in mood disorders.

[0038] FIG. **15** shows selected genes in biochemical pathways involving GABA and glutamate that are differentially expressed in mood disorder subjects.

DEFINITIONS

[0039] A "mental disorder" or "mental illness" or "mental disease" or "psychiatric or neuropsychiatric disease or illness or disorder" refers to mood disorders (e.g., major depression, mania, and bipolar disorders), psychotic disorders (e.g., schizophrenia, schizoaffective disorder, schizophreniform disorder, delusional disorder, brief psychotic disorder, and shared psychotic disorder), personality disorders, anxiety disorders (e.g., obsessive-compulsive disorder) as well as other mental disorders such as substance-related disorders, childhood disorders, dementia, autistic disorder, adjustment disorder, delirium, multi-infarct dementia, and Tourette's disorder as described in Diagnostic and Statistical Manual of Mental Disorders, Fourth Edition, (DSM IV). Typically, such disorders have a complex genetic and/or a biochemical component.

[0040] "A psychotic disorder" refers to a condition that affects the mind, resulting in at least some loss of contact with reality. Symptoms of a psychotic disorder include, e.g., hallucinations, changed behavior that is not based on reality, delusions and the like. See, e.g., DSM IV. Schizophrenia, schizoaffective disorder, schizophreniform disorder, delusional disorder, brief psychotic disorder, substance-induced psychotic disorder, and shared psychotic disorder are examples of psychotic disorders.

[0041] "Schizophrenia" refers to a psychotic disorder involving a withdrawal from reality by an individual. Symptoms comprise for at least a part of a month two or more of the following symptoms: delusions (only one symptom is required if a delusion is bizarre, such as being abducted in a space ship from the sun); hallucinations (only one symptom is required if hallucinations are of at least two voices talking to one another or of a voice that keeps up a running commentary on the patient's thoughts or actions); disorganized speech (e.g., frequent derailment or incoherence); grossly disorganized or catatonic behavior; or negative symptoms, i.e., affective flattening, alogia, or avolition. Schizophrenia encompasses disorders such as, e.g., schizoaffective disorders. Diagnosis of schizophrenia is described in, e.g., DSM IV. Types of schizophrenia include, e.g., paranoid, disorganized, catatonic, undifferentiated, and residual.

[0042] A "mood disorder" refers to disruption of feeling tone or emotional state experienced by an individual for an extensive period of time. Mood disorders include major depression disorder (i.e., unipolar disorder), mania, dysphoria, bipolar disorder, dysthymia, cyclothymia and many others. See, e.g., Diagnostic and Statistical Manual of Mental Disorders, Fourth Edition, (DSM IV).

[0043] "Major depression disorder," "major depressive disorder," or "unipolar disorder" refers to a mood disorder involving any of the following symptoms: persistent sad, anxious, or "empty" mood; feelings of hopelessness or pessimism; feelings of guilt, worthlessness, or helplessness; loss of interest or pleasure in hobbies and activities that were once enjoyed, including sex; decreased energy, fatigue, being "slowed down"; difficulty concentrating, remembering, or making decisions; insomnia, early-morning awakening, or oversleeping; appetite and/or weight loss or overeating and weight gain; thoughts of death or suicide or suicide attempts; restlessness or irritability; or persistent physical symptoms that do not respond to treatment, such as headaches, digestive disorders, and chronic pain. Various subtypes of depression are described in, e.g., DSM IV.

[0044] "Bipolar disorder" is a mood disorder characterized by alternating periods of extreme moods. A person with bipolar disorder experiences cycling of moods that usually swing from being overly elated or irritable (mania) to sad and hopeless (depression) and then back again, with periods of normal mood in between. Diagnosis of bipolar disorder is described in, e.g., DSM IV. Bipolar disorders include bipolar disorder I (mania with or without major depression) and bipolar disorder II (hypomania with major depression), see, e.g., DSM IV.

[0045] An "agonist" refers to an agent that binds to a polypeptide or polynucleotide of the invention, stimulates, increases, activates, facilitates, enhances activation, sensitizes or up regulates the activity or expression of a polypeptide or polynucleotide of the invention.

[0046] An "antagonist" refers to an agent that inhibits expression of a polypeptide or polynucleotide of the invention or binds to, partially or totally blocks stimulation, decreases, prevents, delays activation, inactivates, desensitizes, or down regulates the activity of a polypeptide or polynucleotide of the invention.

[0047] "Inhibitors," "activators," and "modulators" of expression or of activity are used to refer to inhibitory, activating, or modulating molecules, respectively, identified using in vitro and in vivo assays for expression or activity, e.g., ligands, agonists, antagonists, and their homologs and mimetics. The term "modulator" includes inhibitors and activators. Inhibitors are agents that, e.g., inhibit expression of a polypeptide or polynucleotide of the invention or bind to, partially or totally block stimulation or enzymatic activity, decrease, prevent, delay activation, inactivate, desensitize, or down regulate the activity of a polypeptide or polynucleotide of the invention, e.g., antagonists. Activators are agents that, e.g., induce or activate the expression of a polypeptide or polynucleotide of the invention or bind to, stimulate, increase, open, activate, facilitate, enhance activation or enzymatic activity, sensitize or up regulate the activity of a polypeptide or polynucleotide of the invention, e.g., agonists. Modulators include naturally occurring and synthetic ligands, antagonists, agonists, small chemical molecules and the like. Assays to identify inhibitors and activators include, e.g., applying putative modulator compounds to cells, in the presence or absence of a polypeptide or polynucleotide of the invention and then determining the functional effects on a polypeptide or polynucleotide of the invention activity. Samples or assays comprising a polypeptide or polynucleotide of the invention that are treated with a potential activator, inhibitor, or modulator are compared to control samples without the inhibitor, activator, or modulator to examine the extent of effect. Control samples (untreated with modulators) are assigned a relative activity value of 100%. Inhibition is achieved when the activity value of a polypeptide or polynucleotide of the invention relative to the control is about 80%, optionally 50% or 25-1%. Activation is achieved when the activity value of a polypeptide or polynucleotide of the invention relative to the control is 110%, optionally 150%, optionally 200-500%, or 1000-3000% higher.

[0048] The term "test compound" or "drug candidate" or "modulator" or grammatical equivalents as used herein describes any molecule, either naturally occurring or synthetic, e.g., protein, oligopeptide (e.g., from about 5 to about 25 amino acids in length, preferably from about 10 to 20 or 12 to 18 amino acids in length, preferably 12, 15, or 18 amino acids in length), small organic molecule, polysaccharide, lipid, fatty acid, polynucleotide, oligonucleotide, etc. The test compound can be in the form of a library of test compounds, such as a combinatorial or randomized library that provides a sufficient range of diversity. Test compounds are optionally linked to a fusion partner, e.g., targeting compounds, rescue compounds, dimerization compounds, stabilizing compounds, addressable compounds, and other functional moieties. Conventionally, new chemical entities with useful properties are generated by identifying a test compound (called a "lead compound") with some desirable property or activity, e.g., inhibiting activity, creating variants of the lead compound, and evaluating the property and activity of those variant compounds. Often, high throughput screening (HTS) methods are employed for such an analysis.

[0049] A "small organic molecule" refers to an organic molecule, either naturally occurring or synthetic, that has a molecular weight of more than about 50 Daltons and less than about 2500 Daltons, preferably less than about 2000 Daltons, preferably between about 100 to about 1000 Daltons, more preferably between about 200 to about 500 Daltons.

[0050] "Determining the functional effect" refers to assaying for a compound that increases or decreases a parameter that is indirectly or directly under the influence of a polynucleotide or polypeptide of the invention (such as a polynucleotide of Table 2, 3, or 4 or a polypeptide encoded by a gene of Table 2, 3, or 4), e.g., measuring physical and chemical or phenotypic effects. Such functional effects can be measured by any means known to those skilled in the art, e.g., changes in spectroscopic (e.g., fluorescence, absorbance, refractive index), hydrodynamic (e.g., shape), chromatographic, or solubility properties for the protein; measuring inducible markers or transcriptional activation of the protein; measuring binding activity or binding assays, e.g. binding to antibodies; measuring changes in ligand binding affinity; measurement of calcium influx; measurement of the accumulation of an enzymatic product of a polypeptide of the invention or depletion of an substrate; measurement of changes in protein levels of a polypeptide of the invention; measurement of RNA stability; G-protein binding; GPCR phosphorylation or dephosphorylation; signal transduction, e.g., receptor-ligand interactions, second messenger concentrations (e.g., cAMP, IP3, or intracellular Ca^{2+}); identification of downstream or reporter gene expression (CAT, luciferase, β-gal, GFP and the like), e.g., via chemiluminescence, fluorescence, calorimetric reactions, antibody binding, inducible markers, and ligand binding assays.

[0051] Samples or assays comprising a nucleic acid or protein disclosed herein that are treated with a potential activator, inhibitor, or modulator are compared to control samples without the inhibitor, activator, or modulator to examine the extent of inhibition. Control samples (untreated with inhibitors) are assigned a relative protein activity value of 100%. Inhibition is achieved when the activity value relative to the control is about 80%, preferably 50%, more preferably 25-0%. Activation is achieved when the activity value relative to the control (untreated with activators) is 110%, more preferably 150%, more preferably 200-500% (i.e., two to five fold higher relative to the control), more preferably 1000-3000% higher.

[0052] "Biological sample" includes sections of tissues such as biopsy and autopsy samples, and frozen sections taken for histologic purposes. Such samples include blood, spinal fluid, sputum, tissue, lysed cells, brain biopsy, cultured cells, e.g., primary cultures, explants, and transformed cells, stool, urine, etc. A biological sample is typically obtained from a eukaryotic organism, most preferably a mammal such as a primate, e.g., chimpanzee or human; cow; dog; cat; a rodent, e.g., guinea pig, rat, mouse; rabbit; or a bird; reptile; or fish.

[0053] "Antibody" refers to a polypeptide substantially encoded by an immunoglobulin gene or immunoglobulin genes, or fragments thereof which specifically bind and recognize an analyte (antigen). The recognized immunoglobulin genes include the kappa, lambda, alpha, gamma, delta, epsilon and mu constant region genes, as well as the myriad immunoglobulin variable region genes. Light chains are classified as either kappa or lambda. Heavy chains are classified as gamma, mu, alpha, delta, or epsilon, which in turn define the immunoglobulin classes, IgG, IgM, IgA, IgD and IgE, respectively.

[0054] An exemplary immunoglobulin (antibody) structural unit comprises a tetramer. Each tetramer is composed of two identical pairs of polypeptide chains, each pair having one "light" (about 25 kD) and one "heavy" chain (about 50-70 kD). The N-terminus of each chain defines a variable region of about 100 to 110 or more amino acids primarily responsible for antigen recognition. The terms variable light chain (V_L) and variable heavy chain (V_H) refer to these light and heavy chains respectively.

[0055] Antibodies exist, e.g., as intact immunoglobulins or as a number of well-characterized fragments produced by digestion with various peptidases. Thus, for example, pepsin digests an antibody below the disulfide linkages in the hinge region to produce $F(ab)'_2$, a dimer of Fab which itself is a light chain joined to V_H—C_H1 by a disulfide bond. The $F(ab)'_2$ may be reduced under mild conditions to break the disulfide linkage in the hinge region, thereby converting the $F(ab)'_2$ dimer into an Fab' monomer. The Fab' monomer is essentially an Fab with part of the hinge region (see, Paul (Ed.) *Fundamental Immunology*, Third Edition, Raven Press, NY (1993)). While various antibody fragments are defined in terms of the digestion of an intact antibody, one of skill will appreciate that such fragments may be synthesized de novo either chemically or by utilizing recombinant DNA methodology. Thus, the term antibody, as used herein, also includes antibody fragments either produced by the modification of whole antibodies or those synthesized de novo using recombinant DNA methodologies (e.g., single chain Fv).

[0056] The terms "peptidomimetic" and "mimetic" refer to a synthetic chemical compound that has substantially the same structural and functional characteristics of the polynucleotides, polypeptides, antagonists or agonists of the invention. Peptide analogs are commonly used in the pharmaceutical industry as non-peptide drugs with properties analogous to those of the template peptide. These types of non-peptide compound are termed "peptide mimetics" or "peptidomimetics" (Fauchere, *Adv. Drug Res.* 15:29 (1986); Veber and Freidinger *TINS* p. 392 (1985); and Evans et al., *J. Med. Chem.* 30:1229 (1987), which are incorporated herein by reference). Peptide mimetics that are structurally similar to therapeutically useful peptides may be used to produce an

equivalent or enhanced therapeutic or prophylactic effect. Generally, peptidomimetics are structurally similar to a paradigm polypeptide (i.e., a polypeptide that has a biological or pharmacological activity), such as a CCX CKR, but have one or more peptide linkages optionally replaced by a linkage selected from the group consisting of, e.g., —CH_2NH—, —CH_2S—, —CH_2—CH_2—, —CH═CH— (cis and trans), —$COCH_2$—, —$CH(OH)CH_2$—, and —CH_2SO—. The mimetic can be either entirely composed of synthetic, non-natural analogues of amino acids, or, is a chimeric molecule of partly natural peptide amino acids and partly non-natural analogs of amino acids. The mimetic can also incorporate any amount of natural amino acid conservative substitutions as long as such substitutions also do not substantially alter the mimetic's structure and/or activity. For example, a mimetic composition is within the scope of the invention if it is capable of carrying out the binding or enzymatic activities of a polypeptide or polynucleotide of the invention or inhibiting or increasing the enzymatic activity or expression of a polypeptide or polynucleotide of the invention.

[0057] The term "gene" means the segment of DNA involved in producing a polypeptide chain; it includes regions preceding and following the coding region (leader and trailer) as well as intervening sequences (introns) between individual coding segments (exons).

[0058] The term "isolated," when applied to a nucleic acid or protein, denotes that the nucleic acid or protein is essentially free of other cellular components with which it is associated in the natural state. It is preferably in a homogeneous state although it can be in either a dry or aqueous solution. Purity and homogeneity are typically determined using analytical chemistry techniques such as polyacrylamide gel electrophoresis or high performance liquid chromatography. A protein that is the predominant species present in a preparation is substantially purified. In particular, an isolated gene is separated from open reading frames that flank the gene and encode a protein other than the gene of interest. The term "purified" denotes that a nucleic acid or protein gives rise to essentially one band in an electrophoretic gel. Particularly, it means that the nucleic acid or protein is at least 85% pure, more preferably at least 95% pure, and most preferably at least 99% pure.

[0059] The term "nucleic acid" or "polynucleotide" refers to deoxyribonucleotides or ribonucleotides and polymers thereof in either single- or double-stranded form. Unless specifically limited, the term encompasses nucleic acids containing known analogues of natural nucleotides that have similar binding properties as the reference nucleic acid and are metabolized in a manner similar to naturally occurring nucleotides. Unless otherwise indicated, a particular nucleic acid sequence also implicitly encompasses conservatively modified variants thereof (e.g., degenerate codon substitutions), alleles, orthologs, SNPs (haplotypes), and complementary sequences as well as the sequence explicitly indicated. Specifically, degenerate codon substitutions may be achieved by generating sequences in which the third position of one or more selected (or all) codons is substituted with mixed-base and/or deoxyinosine residues (Batzer et al., *Nucleic Acid Res.* 19:5081 (1991); Ohtsuka et al., *J. Biol. Chem.* 260:2605-2608 (1985); and Cassol et al. (1992); Rossolini et al., *Mol. Cell. Probes* 8:91-98 (1994)). The term nucleic acid is used interchangeably with gene, cDNA, and mRNA encoded by a gene.

[0060] The terms "polypeptide," "peptide," and "protein" are used interchangeably herein to refer to a polymer of amino acid residues. The terms apply to amino acid polymers in which one or more amino acid residue is an artificial chemical mimetic of a corresponding naturally occurring amino acid, as well as to naturally occurring amino acid polymers and non-naturally occurring amino acid polymers. As used herein, the terms encompass amino acid chains of any length, including full-length proteins (i.e., antigens), wherein the amino acid residues are linked by covalent peptide bonds.

[0061] The term "amino acid" refers to naturally occurring and synthetic amino acids, as well as amino acid analogs and amino acid mimetics that function in a manner similar to the naturally occurring amino acids. Naturally occurring amino acids are those encoded by the genetic code, as well as those amino acids that are later modified, e.g., hydroxyproline, γ-carboxyglutamate, and O-phosphoserine. Amino acid analogs refers to compounds that have the same basic chemical structure as a naturally occurring amino acid, i.e., an cl carbon that is bound to a hydrogen, a carboxyl group, an amino group, and an R group, e.g., homoserine, norleucine, methionine sulfoxide, methionine methyl sulfonium. Such analogs have modified R groups (e.g., norleucine) or modified peptide backbones, but retain the same basic chemical structure as a naturally occurring amino acid. "Amino acid mimetics" refers to chemical compounds that have a structure that is different from the general chemical structure of an amino acid, but that functions in a manner similar to a naturally occurring amino acid.

[0062] Amino acids may be referred to herein by either the commonly known three letter symbols or by the one-letter symbols recommended by the IUPAC-IUB Biochemical Nomenclature Commission. Nucleotides, likewise, may be referred to by their commonly accepted single-letter codes.

[0063] "Conservatively modified variants" applies to both amino acid and nucleic acid sequences. With respect to particular nucleic acid sequences, "conservatively modified variants" refers to those nucleic acids that encode identical or essentially identical amino acid sequences, or where the nucleic acid does not encode an amino acid sequence, to essentially identical sequences. Because of the degeneracy of the genetic code, a large number of functionally identical nucleic acids encode any given protein. For instance, the codons GCA, GCC, GCG and GCU all encode the amino acid alanine. Thus, at every position where an alanine is specified by a codon, the codon can be altered to any of the corresponding codons described without altering the encoded polypeptide. Such nucleic acid variations are "silent variations," which are one species of conservatively modified variations. Every nucleic acid sequence herein that encodes a polypeptide also describes every possible silent variation of the nucleic acid. One of skill will recognize that each codon in a nucleic acid (except AUG, which is ordinarily the only codon for methionine, and TGG, which is ordinarily the only codon for tryptophan) can be modified to yield a functionally identical molecule. Accordingly, each silent variation of a nucleic acid that encodes a polypeptide is implicit in each described sequence.

[0064] As to amino acid sequences, one of skill will recognize that individual substitutions, deletions or additions to a nucleic acid, peptide, polypeptide, or protein sequence which alters, adds or deletes a single amino acid or a small percentage of amino acids in the encoded sequence is a "conservatively modified variant" where the alteration results in the substitution of an amino acid with a chemically similar amino acid. Conservative substitution tables providing functionally

similar amino acids are well known in the art. Such conservatively modified variants are in addition to and do not exclude polymorphic variants, interspecies homologs, and alleles of the invention.

[0065] The following eight groups each contain amino acids that are conservative substitutions for one another:

1) Alanine (A), Glycine (G);

[0066] 2) Aspartic acid (D), Glutamic acid (E);

3) Asparagine (N), Glutamine (Q);

4) Arginine (R), Lysine (K);

5) Isoleucine (I), Leucine (L), Methionine (M), Valine (V);

6) Phenylalanine (F), Tyrosine (Y), Tryptophan (W);

7) Serine (S), Threonine (T); and

8) Cysteine (C), Methionine (M)

[0067] (see, e.g., Creighton, *Proteins* (1984)).

[0068] "Percentage of sequence identity" is determined by comparing two optimally aligned sequences over a comparison window, wherein the portion of the polynucleotide sequence in the comparison window may comprise additions or deletions (i.e., gaps) as compared to the reference sequence (which does not comprise additions or deletions) for optimal alignment of the two sequences. The percentage is calculated by determining the number of positions at which the identical nucleic acid base or amino acid residue occurs in both sequences to yield the number of matched positions, dividing the number of matched positions by the total number of positions in the window of comparison and multiplying the result by 100 to yield the percentage of sequence identity.

[0069] The terms "identical" or percent "identity," in the context of two or more nucleic acids or polypeptide sequences, refer to two or more sequences or subsequences that are the same or have a specified percentage of amino acid residues or nucleotides that are the same (i.e., 60% identity, optionally 65%, 70%, 75%, 80%, 85%, 90%, or 95% identity over a specified region), when compared and aligned for maximum correspondence over a comparison window, or designated region as measured using one of the following sequence comparison algorithms or by manual alignment and visual inspection. Such sequences are then said to be "substantially identical." This definition also refers to the complement of a test sequence. Optionally, the identity exists over a region that is at least about 50 nucleotides in length, or more preferably over a region that is 100 to 500 or 1000 or more nucleotides in length.

[0070] For sequence comparison, typically one sequence acts as a reference sequence, to which test sequences are compared. When using a sequence comparison algorithm, test and reference sequences are entered into a computer, subsequence coordinates are designated, if necessary, and sequence algorithm program parameters are designated. Default program parameters can be used, or alternative parameters can be designated. The sequence comparison algorithm then calculates the percent sequence identities for the test sequences relative to the reference sequence, based on the program parameters.

[0071] A "comparison window", as used herein, includes reference to a segment of any one of the number of contiguous positions selected from the group consisting of from 20 to 600, usually about 50 to about 200, more usually about 100 to about 150 in which a sequence may be compared to a reference sequence of the same number of contiguous positions after the two sequences are optimally aligned. Methods of alignment of sequences for comparison are well known in the art. Optimal alignment of sequences for comparison can be conducted, e.g., by the local homology algorithm of Smith and Waterman (1970) *Adv. Appl. Math.* 2:482c, by the homology alignment algorithm of Needleman and Wunsch (1970) *J. Mol. Biol.* 48:443, by the search for similarity method of Pearson and Lipman (1988) *Proc. Nat.'l. Acad. Sci. USA* 85:2444, by computerized implementations of these algorithms (GAP, BESTFIT, FASTA, and TFASTA in the Wisconsin Genetics Software Package, Genetics Computer Group, 575 Science Dr., Madison, Wis.), or by manual alignment and visual inspection (see, e.g., Ausubel et al., *Current Protocols in Molecular Biology* (1995 supplement)).

[0072] An example of an algorithm that is suitable for determining percent sequence identity and sequence similarity are the BLAST and BLAST 2.0 algorithms, which are described in Altschul et al. (1977) *Nuc. Acids Res.* 25:3389-3402, and Altschul et al. (1990) *J. Mol. Biol.* 215:403-410, respectively. Software for performing BLAST analyses is publicly available through the National Center for Biotechnology Information. This algorithm involves first identifying high scoring sequence pairs (HSPs) by identifying short words of length W in the query sequence, which either match or satisfy some positive-valued threshold score T when aligned with a word of the same length in a database sequence. T is referred to as the neighborhood word score threshold (Altschul et al., supra). These initial neighborhood word hits act as seeds for initiating searches to find longer HSPs containing them. The word hits are extended in both directions along each sequence for as far as the cumulative alignment score can be increased. Cumulative scores are calculated using, for nucleotide sequences, the parameters M (reward score for a pair of matching residues; always >0) and N (penalty score for mismatching residues; always <0). For amino acid sequences, a scoring matrix is used to calculate the cumulative score. Extension of the word hits in each direction are halted when: the cumulative alignment score falls off by the quantity X from its maximum achieved value; the cumulative score goes to zero or below, due to the accumulation of one or more negative-scoring residue alignments; or the end of either sequence is reached. The BLAST algorithm parameters W, T, and X determine the sensitivity and speed of the alignment. The BLASTN program (for nucleotide sequences) uses as defaults a wordlength (W) of 11, an expectation (E) or 10, M=5, N=−4 and a comparison of both strands. For amino acid sequences, the BLASTP program uses as defaults a wordlength of 3, and expectation (E) of 10, and the BLOSUM62 scoring matrix (see Henikoff and Henikoff (1989) *Proc. Natl. Acad. Sci. USA* 89:10915) alignments (B) of 50, expectation (E) of 10, M=5, N=−4, and a comparison of both strands.

[0073] The BLAST algorithm also performs a statistical analysis of the similarity between two sequences (see, e.g., Karlin and Altschul (1993) *Proc. Natl. Acad. Sci. USA* 90:5873-5787). One measure of similarity provided by the BLAST algorithm is the smallest sum probability (P(N)), which provides an indication of the probability by which a match between two nucleotide or amino acid sequences would occur by chance. For example, a nucleic acid is considered similar to a reference sequence if the smallest sum probability in a comparison of the test nucleic acid to the

reference nucleic acid is less than about 0.2, more preferably less than about 0.01, and most preferably less than about 0.001.

[0074] An indication that two nucleic acid sequences or polypeptides are substantially identical is that the polypeptide encoded by the first nucleic acid is immunologically cross reactive with the antibodies raised against the polypeptide encoded by the second nucleic acid, as described below. Thus, a polypeptide is typically substantially identical to a second polypeptide, for example, where the two peptides differ only by conservative substitutions. Another indication that two nucleic acid sequences are substantially identical is that the two molecules or their complements hybridize to each other under stringent conditions, as described below. Yet another indication that two nucleic acid sequences are substantially identical is that the same primers can be used to amplify the sequence.

[0075] The phrase "selectively (or specifically) hybridizes to" refers to the binding, duplexing, or hybridizing of a molecule only to a particular nucleotide sequence under stringent hybridization conditions when that sequence is present in a complex mixture (e.g., total cellular or library DNA or RNA).

[0076] The phrase "stringent hybridization conditions" refers to conditions under which a probe will hybridize to its target subsequence, typically in a complex mixture of nucleic acid, but to no other sequences. Stringent conditions are sequence-dependent and will be different in different circumstances. Longer sequences hybridize specifically at higher temperatures. An extensive guide to the hybridization of nucleic acids is found in Tijssen, *Techniques in Biochemistry and Molecular Biology—Hybridization with Nucleic Probes*, "Overview of principles of hybridization and the strategy of nucleic acid assays" (1993). Generally, stringent conditions are selected to be about 5-10° C. lower than the thermal melting point (T_m) for the specific sequence at a defined ionic strength pH. The T_m is the temperature (under defined ionic strength, pH, and nucleic concentration) at which 50% of the probes complementary to the target hybridize to the target sequence at equilibrium (as the target sequences are present in excess, at T_m, 50% of the probes are occupied at equilibrium). Stringent conditions will be those in which the salt concentration is less than about 1.0 M sodium ion, typically about 0.01 to 1.0 M sodium ion concentration (or other salts) at pH 7.0 to 8.3 and the temperature is at least about 30° C. for short probes (e.g., 10 to 50 nucleotides) and at least about 60° C. for long probes (e.g., greater than 50 nucleotides). Stringent conditions may also be achieved with the addition of destabilizing agents such as formamide. For selective or specific hybridization, a positive signal is at least two times background, optionally 10 times background hybridization. Exemplary stringent hybridization conditions can be as following: 50% formamide, 5×SSC, and 1% SDS, incubating at 42° C., or 5×SSC, 1% SDS, incubating at 65° C., with wash in 0.2×SSC, and 0.1% SDS at 65° C. Such washes can be performed for 5, 15, 30, 60, 120, or more minutes. Nucleic acids that hybridize to the genes listed in Tables 1-8 are encompassed by the invention.

[0077] Nucleic acids that do not hybridize to each other under stringent conditions are still substantially identical if the polypeptides that they encode are substantially identical. This occurs, for example, when a copy of a nucleic acid is created using the maximum codon degeneracy permitted by the genetic code. In such cases, the nucleic acids typically hybridize under moderately stringent hybridization conditions. Exemplary "moderately stringent hybridization conditions" include a hybridization in a buffer of 40% formamide, 1 M NaCl, 1% SDS at 37° C., and a wash in 1×SSC at 45° C. Such washes can be performed for 5, 15, 30, 60, 120, or more minutes. A positive hybridization is at least twice background. Those of ordinary skill will readily recognize that alternative hybridization and wash conditions can be utilized to provide conditions of similar stringency.

[0078] For PCR, a temperature of about 36° C. is typical for low stringency amplification, although annealing temperatures may vary between about 32° C. and 48° C. depending on primer length. For high stringency PCR amplification, a temperature of about 62° C. is typical, although high stringency annealing temperatures can range from about 50° C. to about 65° C., depending on the primer length and specificity. Typical cycle conditions for both high and low stringency amplifications include a denaturation phase of 90° C.-95° C. for 30 sec-2 min., an annealing phase lasting 30 sec.-2 min., and an extension phase of about 72° C. for 1-2 min. Protocols and guidelines for low and high stringency amplification reactions are provided, e.g., in Innis et al., *PCR Protocols, A Guide to Methods and Applications* (1990).

[0079] The phrase "a nucleic acid sequence encoding" refers to a nucleic acid that contains sequence information for a structural RNA such as rRNA, a tRNA, or the primary amino acid sequence of a specific protein or peptide, or a binding site for a trans-acting regulatory agent. This phrase specifically encompasses degenerate codons (i.e., different codons which encode a single amino acid) of the native sequence or sequences which may be introduced to conform with codon preference in a specific host cell.

[0080] The term "recombinant" when used with reference, e.g., to a cell, or nucleic acid, protein, or vector, indicates that the cell, nucleic acid, protein or vector, has been modified by the introduction of a heterologous nucleic acid or protein or the alteration of a native nucleic acid or protein, or that the cell is derived from a cell so modified. Thus, for example, recombinant cells express genes that are not found within the native (nonrecombinant) form of the cell or express native genes that are otherwise abnormally expressed, under-expressed or not expressed at all.

[0081] The term "heterologous" when used with reference to portions of a nucleic acid indicates that the nucleic acid comprises two or more subsequences that are not found in the same relationship to each other in nature. For instance, the nucleic acid is typically recombinantly produced, having two or more sequences from unrelated genes arranged to make a new functional nucleic acid, e.g., a promoter from one source and a coding region from another source. Similarly, a heterologous protein indicates that the protein comprises two or more subsequences that are not found in the same relationship to each other in nature (e.g., a fusion protein).

[0082] An "expression vector" is a nucleic acid construct, generated recombinantly or synthetically, with a series of specified nucleic acid elements that permit transcription of a particular nucleic acid in a host cell. The expression vector can be part of a plasmid, virus, or nucleic acid fragment. Typically, the expression vector includes a nucleic acid to be transcribed operably linked to a promoter.

[0083] The phrase "specifically (or selectively) binds to an antibody" or "specifically (or selectively) immunoreactive with", when referring to a protein or peptide, refers to a binding reaction which is determinative of the presence of the protein in the presence of a heterogeneous population of

proteins and other biologics. Thus, under designated immunoassay conditions, the specified antibodies bind to a particular protein and do not bind in a significant amount to other proteins present in the sample. Specific binding to an antibody under such conditions may require an antibody that is selected for its specificity for a particular protein. For example, antibodies raised against a protein having an amino acid sequence encoded by any of the polynucleotides of the invention can be selected to obtain antibodies specifically immunoreactive with that protein and not with other proteins, except for polymorphic variants. A variety of immunoassay formats may be used to select antibodies specifically immunoreactive with a particular protein. For example, solid-phase ELISA immunoassays, Western blots, or immunohistochemistry are routinely used to select monoclonal antibodies specifically immunoreactive with a protein. See, Harlow and Lane *Antibodies, A Laboratory Manual*, Cold Spring Harbor Publications, NY (1988) for a description of immunoassay formats and conditions that can be used to determine specific immunoreactivity. Typically, a specific or selective reaction will be at least twice the background signal or noise and more typically more than 10 to 100 times background.

[0084] One who is "predisposed for a mental disorder" as used herein means a person who has an inclination or a higher likelihood of developing a mental disorder when compared to an average person in the general population.

DETAILED DESCRIPTION OF THE INVENTION

I. Introduction

[0085] To understand the complex genetic basis of mental disorders, the present invention provides studies that have been conducted to investigate the expression patterns of genes that are differentially expressed specifically in central nervous system of subjects with mood disorders. The large spectrum of symptoms associated with mental disorders is likely a reflection of the complex genetic basis and complex gene expression patterns in patients with mental disorders. Different combinations of the genes disclosed herein can be responsible for one or more mental disorders. Furthermore, brain pathways or circuits as well as subcellular pathways are important for understanding the development and diagnosis of mental disorders. The selected brain regions described herein (AnCng, DLPFC, and CB) are implicated in the clinical symptoms of mental disorders such as mood disorders. Brain imaging studies focusing on particular brain regions, cytoarchitectural changes in brain regions, expression of key neurotransmitters or related molecules in brain regions, and subcellular pathways in brain regions all contribute to the development of mental disorders, and thus are an important consideration in the diagnosis and therapeutic uses described herein.

[0086] The present invention demonstrates the altered expression (either higher or lower expression) of the genes of Tables 1-8 at the mRNA level in selected brain regions of patients diagnosed with mood disorders (e.g., bipolar disorder and major depression disorder) in comparison with normal individuals. This invention thus provides methods for diagnosis of mental disorders such as mood disorders (e.g., bipolar disorder, major depression, and the like), psychotic disorders (e.g., schizophrenia, and the like), and other mental disorders by detecting the level of a transcript or translation product of the genes listed in Tables 1-8 as well as their corresponding biochemical pathways. The chromosomal location of such genes can be used to discover other genes in the region that are linked to development of a particular disorder.

[0087] The invention further provides methods of identifying a compound useful for the treatment of such disorders by selecting compounds that modulates the functional effect of the translation products or the expression of the transcripts described herein. The invention also provides for methods of treating patients with such mental disorders, e.g., by administering the compounds of the invention or by gene therapy.

[0088] The genes and the polypeptides that they encode, which are associated with mood disorders such as bipolar disease and major depression, are useful for facilitating the design and development of various molecular diagnostic tools such as GeneChip™ containing probe sets specific for all or selected mental disorders, including but not limited to mood disorders, and as an ante- and/or post-natal diagnostic tool for screening newborns in concert with genetic counseling. Other diagnostic applications include evaluation of disease susceptibility, prognosis, and monitoring of disease or treatment process, as well as providing individualized medicine via predictive drug profiling systems, e.g., by correlating specific genomic motifs with the clinical response of a patient to individual drugs. In addition, the present invention is useful for multiplex SNP or haplotype profiling, including but not limited to the identification of pharmacogenetic targets at the gene, mRNA, protein, and pathway level.

[0089] The genes and the polypeptides that they encode, described herein, as also useful as drug targets for the development of therapeutic drugs for the treatment or prevention of mental disorders, including but not limited to mood disorders. Mental disorders have a high co-morbidity with other neurological disorders, such as Parkinson's disease or Alzheimer's. Therefore, the present invention can be used for diagnosis and treatment of patients with multiple disease states that include a mental disorder such as a mood disorder.

[0090] For example, antidepressants belong to different classes, e.g., desipramine, bupropion, and fluoxetine are in general equally effect for the treatment of clinical depression, but act by different mechanisms. The similar effectiveness of the drugs for treatment of mood disorders suggests that they act through a yet as unidentified common pathway. We disclose herein that different classes of antidepressants (specific serotonin reuptake inhibitors, like fluoxetine and tricyclic antidepressants, like desipramine) regulate a common gene, and/or a common group of genes as well as a unique set of genes when the human and animal results herein are compared.

II. General Recombinant Nucleic Acid Methods for Use with the Invention

[0091] In numerous embodiments of the present invention, polynucleotides of the invention will be isolated and cloned using recombinant methods. Such polynucleotides include, e.g., those listed in Tables 1-8, which can be used for, e.g., protein expression or during the generation of variants, derivatives, expression cassettes, to monitor gene expression, for the isolation or detection of sequences of the invention in different species, for diagnostic purposes in a patient, e.g., to detect mutations or to detect expression levels of nucleic acids or polypeptides of the invention. In some embodiments, the sequences of the invention are operably linked to a heterologous promoter. In one embodiment, the nucleic acids of

the invention are from any mammal, including, in particular, e.g., a human, a mouse, a rat, a primate, etc.

A. General Recombinant Nucleic Acids Methods

[0092] This invention relies on routine techniques in the field of recombinant genetics. Basic texts disclosing the general methods of use in this invention include Sambrook et al., *Molecular Cloning, A Laboratory Manual* (3rd ed. 2001); Kriegler, *Gene Transfer and Expression. A Laboratory Manual* (1990); and *Current Protocols in Molecular Biology* (Ausubel et al., eds., 1994)).

[0093] For nucleic acids, sizes are given in either kilobases (kb) or base pairs (bp). These are estimates derived from agarose or acrylamide gel electrophoresis, from sequenced nucleic acids, or from published DNA sequences. For proteins, sizes are given in kilodaltons (kDa) or amino acid residue numbers. Proteins sizes are estimated from gel electrophoresis, from sequenced proteins, from derived amino acid sequences, or from published protein sequences.

[0094] Oligonucleotides that are not commercially available can be chemically synthesized according to the solid phase phosphoramidite triester method first described by Beaucage & Caruthers, *Tetrahedron Letts.* 22:1859-1862 (1981), using an automated synthesizer, as described in Van Devanter et. al., *Nucleic Acids Res.* 12:6159-6168 (1984). Purification of oligonucleotides is by either native acrylamide gel electrophoresis or by anion-exchange HPLC as described in Pearson & Reanier, *J. Chrom.* 255:137-149 (1983).

[0095] The sequence of the cloned genes and synthetic oligonucleotides can be verified after cloning using, e.g., the chain termination method for sequencing double-stranded templates of Wallace et al., *Gene* 16:21-26 (1981).

B. Cloning Methods for the Isolation of Nucleotide Sequences Encoding Desired Proteins

[0096] In general, the nucleic acids encoding the subject proteins are cloned from DNA sequence libraries that are made to encode cDNA or genomic DNA. The particular sequences can be located by hybridizing with an oligonucleotide probe, the sequence of which can be derived from the sequences of the genes listed in Tables 1-8, which provide a reference for PCR primers and defines suitable regions for isolating specific probes. Alternatively, where the sequence is cloned into an expression library, the expressed recombinant protein can be detected immunologically with antisera or purified antibodies made against a polypeptide comprising an amino acid sequence encoded by a gene listed in Table 1-8.

[0097] Methods for making and screening genomic and cDNA libraries are well known to those of skill in the art (see, e.g., Gubler and Hoffman *Gene* 25:263-269 (1983); Benton and Davis *Science,* 196:180-182 (1977); and Sambrook, supra). Brain cells are an example of suitable cells to isolate RNA and cDNA sequences of the invention.

[0098] Briefly, to make the cDNA library, one should choose a source that is rich in mRNA. The mRNA can then be made into cDNA, ligated into a recombinant vector, and transfected into a recombinant host for propagation, screening and cloning. For a genomic library, the DNA is extracted from a suitable tissue and either mechanically sheared or enzymatically digested to yield fragments of preferably about 5-100 kb. The fragments are then separated by gradient centrifugation from undesired sizes and are constructed in bacteriophage lambda vectors. These vectors and phage are packaged in vitro, and the recombinant phages are analyzed by plaque hybridization. Colony hybridization is carried out as generally described in Grunstein et al., *Proc. Natl. Acad. Sci. USA.,* 72:3961-3965 (1975).

[0099] An alternative method combines the use of synthetic oligonucleotide primers with polymerase extension on an mRNA or DNA template. Suitable primers can be designed from specific sequences of the invention. This polymerase chain reaction (PCR) method amplifies the nucleic acids encoding the protein of interest directly from mRNA, cDNA, genomic libraries or cDNA libraries. Restriction endonuclease sites can be incorporated into the primers. Polymerase chain reaction or other in vitro amplification methods may also be useful, for example, to clone nucleic acids encoding specific proteins and express said proteins, to synthesize nucleic acids that will be used as probes for detecting the presence of mRNA encoding a polypeptide of the invention in physiological samples, for nucleic acid sequencing, or for other purposes (see, U.S. Pat. Nos. 4,683,195 and 4,683,202). Genes amplified by a PCR reaction can be purified from agarose gels and cloned into an appropriate vector.

[0100] Appropriate primers and probes for identifying polynucleotides of the invention from mammalian tissues can be derived from the sequences provided herein. For a general overview of PCR, see, Innis et al. *PCR Protocols. A Guide to Methods and Applications, Academic Press*, San Diego (1990).

[0101] Synthetic oligonucleotides can be used to construct genes. This is done using a series of overlapping oligonucleotides, usually 40-120 bp in length, representing both the sense and anti-sense strands of the gene. These DNA fragments are then annealed, ligated and cloned.

[0102] A gene encoding a polypeptide of the invention can be cloned using intermediate vectors before transformation into mammalian cells for expression. These intermediate vectors are typically prokaryote vectors or shuttle vectors. The proteins can be expressed in either prokaryotes, using standard methods well known to those of skill in the art, or eukaryotes as described infra.

III. Purification of Proteins of the Invention

[0103] Either naturally occurring or recombinant polypeptides of the invention can be purified for use in functional assays. Naturally occurring polypeptides, e.g., polypeptides encoded by genes listed in Tables 1-8, can be purified, for example, from mouse or human tissue such as brain or any other source of an ortholog. Recombinant polypeptides can be purified from any suitable expression system.

[0104] The polypeptides of the invention may be purified to substantial purity by standard techniques, including selective precipitation with such substances as ammonium sulfate; column chromatography, immunopurification methods, and others (see, e.g., Scopes, *Protein Purification Principles and Practice* (1982); U.S. Pat. No. 4,673,641; Ausubel et al., supra; and Sambrook et al., supra).

[0105] A number of procedures can be employed when recombinant polypeptides are purified. For example, proteins having established molecular adhesion properties can be reversible fused to polypeptides of the invention. With the appropriate ligand, the polypeptides can be selectively adsorbed to a purification column and then freed from the column in a relatively pure form. The fused protein is then removed by enzymatic activity. Finally the polypeptide can be purified using immunoaffinity columns.

A. Purification of Proteins from Recombinant Bacteria

[0106] When recombinant proteins are expressed by the transformed bacteria in large amounts, typically after promoter induction, although expression can be constitutive, the proteins may form insoluble aggregates. There are several protocols that are suitable for purification of protein inclusion bodies. For example, purification of aggregate proteins (hereinafter referred to as inclusion bodies) typically involves the extraction, separation and/or purification of inclusion bodies by disruption of bacterial cells typically, but not limited to, by incubation in a buffer of about 100-150 μg/ml lysozyme and 0.1% Nonidet P40, a non-ionic detergent. The cell suspension can be ground using a Polytron grinder (Brinkman Instruments, Westbury, N.Y.). Alternatively, the cells can be sonicated on ice. Alternate methods of lysing bacteria are described in Ausubel et al. and Sambrook et al., both supra, and will be apparent to those of skill in the art.

[0107] The cell suspension is generally centrifuged and the pellet containing the inclusion bodies resuspended in buffer which does not dissolve but washes the inclusion bodies, e.g., 20 mM Tris-HCl (pH 7.2), 1 mM EDTA, 150 mM NaCl and 2% Triton-X 100, a non-ionic detergent. It may be necessary to repeat the wash step to remove as much cellular debris as possible. The remaining pellet of inclusion bodies may be resuspended in an appropriate buffer (e.g., 20 mM sodium phosphate, pH 6.8, 150 mM NaCl). Other appropriate buffers will be apparent to those of skill in the art.

[0108] Following the washing step, the inclusion bodies are solubilized by the addition of a solvent that is both a strong hydrogen acceptor and a strong hydrogen donor (or a combination of solvents each having one of these properties). The proteins that formed the inclusion bodies may then be renatured by dilution or dialysis with a compatible buffer. Suitable solvents include, but are not limited to, urea (from about 4 M to about 8 M), formamide (at least about 80%, volume/volume basis), and guanidine hydrochloride (from about 4 M to about 8 M). Some solvents that are capable of solubilizing aggregate-forming proteins, such as SDS (sodium dodecyl sulfate) and 70% formic acid, are inappropriate for use in this procedure due to the possibility of irreversible denaturation of the proteins, accompanied by a lack of immunogenicity and/or activity. Although guanidine hydrochloride and similar agents are denaturants, this denaturation is not irreversible and renaturation may occur upon removal (by dialysis, for example) or dilution of the denaturant, allowing re-formation of the immunologically and/or biologically active protein of interest. After solubilization, the protein can be separated from other bacterial proteins by standard separation techniques.

[0109] Alternatively, it is possible to purify proteins from bacteria periplasm. Where the protein is exported into the periplasm of the bacteria, the periplasmic fraction of the bacteria can be isolated by cold osmotic shock in addition to other methods known to those of skill in the art (see, Ausubel et al., supra). To isolate recombinant proteins from the periplasm, the bacterial cells are centrifuged to form a pellet. The pellet is resuspended in a buffer containing 20% sucrose. To lyse the cells, the bacteria are centrifuged and the pellet is resuspended in ice-cold 5 mM $MgSO_4$ and kept in an ice bath for approximately 10 minutes. The cell suspension is centrifuged and the supernatant decanted and saved. The recombinant proteins present in the supernatant can be separated from the host proteins by standard separation techniques well known to those of skill in the art.

B. Standard Protein Separation Techniques For Purifying Proteins

1. Solubility Fractionation

[0110] Often as an initial step, and if the protein mixture is complex, an initial salt fractionation can separate many of the unwanted host cell proteins (or proteins derived from the cell culture media) from the recombinant protein of interest. The preferred salt is ammonium sulfate. Ammonium sulfate precipitates proteins by effectively reducing the amount of water in the protein mixture. Proteins then precipitate on the basis of their solubility. The more hydrophobic a protein is, the more likely it is to precipitate at lower ammonium sulfate concentrations. A typical protocol is to add saturated ammonium sulfate to a protein solution so that the resultant ammonium sulfate concentration is between 20-30%. This will precipitate the most hydrophobic proteins. The precipitate is discarded (unless the protein of interest is hydrophobic) and ammonium sulfate is added to the supernatant to a concentration known to precipitate the protein of interest. The precipitate is then solubilized in buffer and the excess salt removed if necessary, through either dialysis or diafiltration. Other methods that rely on solubility of proteins, such as cold ethanol precipitation, are well known to those of skill in the art and can be used to fractionate complex protein mixtures.

2. Size Differential Filtration

[0111] Based on a calculated molecular weight, a protein of greater and lesser size can be isolated using ultrafiltration through membranes of different pore sizes (for example, Amicon or Millipore membranes). As a first step, the protein mixture is ultrafiltered through a membrane with a pore size that has a lower molecular weight cut-off than the molecular weight of the protein of interest. The retentate of the ultrafiltration is then ultrafiltered against a membrane with a molecular cut off greater than the molecular weight of the protein of interest. The recombinant protein will pass through the membrane into the filtrate. The filtrate can then be chromatographed as described below.

3. Column Chromatography

[0112] The proteins of interest can also be separated from other proteins on the basis of their size, net surface charge, hydrophobicity and affinity for ligands. In addition, antibodies raised against proteins can be conjugated to column matrices and the proteins immunopurified. All of these methods are well known in the art.

[0113] It will be apparent to one of skill that chromatographic techniques can be performed at any scale and using equipment from many different manufacturers (e.g., Pharmacia Biotech).

IV. Detection of Gene Expression

[0114] Those of skill in the art will recognize that detection of expression of polynucleotides of the invention has many uses. For example, as discussed herein, detection of the level of polypeptides or polynucleotides of the invention in a patient is useful for diagnosing mental disorders including mood disorders or psychotic disorders or a predisposition for a mood disorder or psychotic disorder. Moreover, detection of

gene expression is useful to identify modulators of expression of the polypeptides or polynucleotides of the invention.

[0115] A variety of methods of specific DNA and RNA measurement using nucleic acid hybridization techniques are known to those of skill in the art (see, Sambrook, supra). Some methods involve an electrophoretic separation (e.g., Southern blot for detecting DNA, and Northern blot for detecting RNA), but measurement of DNA and RNA can also be carried out in the absence of electrophoretic separation (e.g., by dot blot). Southern blot of genomic DNA (e.g., from a human) can be used for screening for restriction fragment length polymorphism (RFLP) to detect the presence of a genetic disorder affecting a polypeptide of the invention.

[0116] The selection of a nucleic acid hybridization format is not critical. A variety of nucleic acid hybridization formats are known to those skilled in the art. For example, common formats include sandwich assays and competition or displacement assays. Hybridization techniques are generally described in Hames and Higgins *Nucleic Acid Hybridization, A Practical Approach*, IRL Press (1985); Gall and Pardue, *Proc. Natl. Acad. Sci. U.S.A.,* 63:378-383 (1969); and John et al. *Nature,* 223:582-587 (1969).

[0117] Detection of a hybridization complex may require the binding of a signal-generating complex to a duplex of target and probe polynucleotides or nucleic acids. Typically, such binding occurs through ligand and anti-ligand interactions as between a ligand-conjugated probe and an anti-ligand conjugated with a signal. The binding of the signal generation complex is also readily amenable to accelerations by exposure to ultrasonic energy.

[0118] The label may also allow indirect detection of the hybridization complex. For example, where the label is a hapten or antigen, the sample can be detected by using antibodies. In these systems, a signal is generated by attaching fluorescent or enzyme molecules to the antibodies or in some cases, by attachment to a radioactive label (see, e.g., Tijssen, "*Practice and Theory of Enzyme Immunoassays," Laboratory Techniques in Biochemistry and Molecular Biology*, Burdon and van Knippenberg Eds., Elsevier (1985), pp. 9-20).

[0119] The probes are typically labeled either directly, as with isotopes, chromophores, lumiphores, chromogens, or indirectly, such as with biotin, to which a streptavidin complex may later bind. Thus, the detectable labels used in the assays of the present invention can be primary labels (where the label comprises an element that is detected directly or that produces a directly detectable element) or secondary labels (where the detected label binds to a primary label, e.g., as is common in immunological labeling). Typically, labeled signal nucleic acids are used to detect hybridization. Complementary nucleic acids or signal nucleic acids may be labeled by any one of several methods typically used to detect the presence of hybridized polynucleotides. The most common method of detection is the use of autoradiography with ^{3}H, ^{125}I, ^{35}S, ^{14}C, or ^{32}P-labeled probes or the like.

[0120] Other labels include, e.g., ligands that bind to labeled antibodies, fluorophores, chemiluminescent agents, enzymes, and antibodies which can serve as specific binding pair members for a labeled ligand. An introduction to labels, labeling procedures and detection of labels is found in Polak and Van Noorden *Introduction to Immunocytochemistry,* 2nd ed., Springer Verlag, NY (1997); and in Haugland *Handbook of Fluorescent Probes and Research Chemicals*, a combined handbook and catalogue Published by Molecular Probes, Inc. (1996).

[0121] In general, a detector which monitors a particular probe or probe combination is used to detect the detection reagent label. Typical detectors include spectrophotometers, phototubes and photodiodes, microscopes, scintillation counters, cameras, film and the like, as well as combinations thereof. Examples of suitable detectors are widely available from a variety of commercial sources known to persons of skill in the art. Commonly, an optical image of a substrate comprising bound labeling moieties is digitized for subsequent computer analysis.

[0122] Most typically, the amount of RNA is measured by quantifying the amount of label fixed to the solid support by binding of the detection reagent. Typically, the presence of a modulator during incubation will increase or decrease the amount of label fixed to the solid support relative to a control incubation which does not comprise the modulator, or as compared to a baseline established for a particular reaction type. Means of detecting and quantifying labels are well known to those of skill in the art.

[0123] In preferred embodiments, the target nucleic acid or the probe is immobilized on a solid support. Solid supports suitable for use in the assays of the invention are known to those of skill in the art. As used herein, a solid support is a matrix of material in a substantially fixed arrangement.

[0124] A variety of automated solid-phase assay techniques are also appropriate. For instance, very large scale immobilized polymer arrays (VLSIPS™), available from Affymetrix, Inc. (Santa Clara, Calif.) can be used to detect changes in expression levels of a plurality of genes involved in the same regulatory pathways simultaneously. See, Tijssen, supra., Fodor et al. (1991) *Science,* 251: 767-777; Sheldon et al. (1993) *Clinical Chemistry* 39(4): 718-719, and Kozal et al. (1996) *Nature Medicine* 2(7): 753-759.

[0125] Detection can be accomplished, for example, by using a labeled detection moiety that binds specifically to duplex nucleic acids (e.g., an antibody that is specific for RNA-DNA duplexes). One preferred example uses an antibody that recognizes DNA-RNA heteroduplexes in which the antibody is linked to an enzyme (typically by recombinant or covalent chemical bonding). The antibody is detected when the enzyme reacts with its substrate, producing a detectable product. Coutlee et al. (1989) *Analytical Biochemistry* 181: 153-162; Bogulavski (1986) et al. *J. Immunol. Methods* 89:123-130; Prooijen-Knegt (1982) *Exp. Cell Res.* 141:397-407; Rudkin (1976) *Nature* 265:472-473, Stollar (1970) *Proc. Nat'l Acad. Sci. USA* 65:993-1000; Ballard (1982) *Mol. Immunol.* 19:793-799; Pisetsky and Caster (1982) *Mol. Immunol.* 19:645-650; Viscidi et al. (1988) *J. Clin. Microbial.* 41:199-209; and Kiney et al. (1989) *J. Clin. Microbiol.* 27:6-12 describe antibodies to RNA duplexes, including homo and heteroduplexes. Kits comprising antibodies specific for DNA:RNA hybrids are available, e.g., from Digene Diagnostics, Inc. (Beltsville, Md.).

[0126] In addition to available antibodies, one of skill in the art can easily make antibodies specific for nucleic acid duplexes using existing techniques, or modify those antibodies that are commercially or publicly available. In addition to the art referenced above, general methods for producing polyclonal and monoclonal antibodies are known to those of skill in the art (see, e.g., Paul (3rd ed.) *Fundamental Immunology* Raven Press, Ltd., NY (1993); Coligan *Current Protocols in Immunology* Wiley/Greene, NY (1991); Harlow and Lane *Antibodies: A Laboratory Manual* Cold Spring Harbor Press, NY (1988); Stites et al. (eds.) *Basic and Clinical Immunology*

(4th ed.) Lange Medical Publications, Los Altos, Calif., and references cited therein; *Goding Monoclonal Antibodies: Principles and Practice* (2d ed.) Academic Press, New York, N.Y., (1986); and Kohler and Milstein *Nature* 256: 495-497 (1975)). Other suitable techniques for antibody preparation include selection of libraries of recombinant antibodies in phage or similar vectors (see, Huse et al. *Science* 246:1275-1281 (1989); and Ward et al. *Nature* 341:544-546 (1989)). Specific monoclonal and polyclonal antibodies and antisera will usually bind with a K_D of at least about 0.1 μM, preferably at least about 0.01 μM or better, and most typically and preferably, 0.001 μM or better.

[0127] The nucleic acids used in this invention can be either positive or negative probes. Positive probes bind to their targets and the presence of duplex formation is evidence of the presence of the target. Negative probes fail to bind to the suspect target and the absence of duplex formation is evidence of the presence of the target. For example, the use of a wild type specific nucleic acid probe or PCR primers may serve as a negative probe in an assay sample where only the nucleotide sequence of interest is present.

[0128] The sensitivity of the hybridization assays may be enhanced through use of a nucleic acid amplification system that multiplies the target nucleic acid being detected. Examples of such systems include the polymerase chain reaction (PCR) system, in particular RT-PCR or real time PCR, and the ligase chain reaction (LCR) system. Other methods recently described in the art are the nucleic acid sequence based amplification (NASBA, Cangene, Mississauga, Ontario) and Q Beta Replicase systems. These systems can be used to directly identify mutants where the PCR or LCR primers are designed to be extended or ligated only when a selected sequence is present. Alternatively, the selected sequences can be generally amplified using, for example, nonspecific PCR primers and the amplified target region later probed for a specific sequence indicative of a mutation.

[0129] An alternative means for determining the level of expression of the nucleic acids of the present invention is in situ hybridization. In situ hybridization assays are well known and are generally described in Angerer et al., *Methods Enzymol.* 152:649-660 (1987). In an in situ hybridization assay, cells or tissue, preferentially human cells or tissue from the cerebellum or the hippocampus, are fixed to a solid support, typically a glass slide. If DNA is to be probed, the cells are denatured with heat or alkali. The cells are then contacted with a hybridization solution at a moderate temperature to permit annealing of specific probes that are labeled. The probes are preferably labeled with radioisotopes or fluorescent reporters.

V. Immunological Detection of the Polypeptides of the Invention

[0130] In addition to the detection of polynucleotide expression using nucleic acid hybridization technology, one can also use immunoassays to detect polypeptides of the invention. Immunoassays can be used to qualitatively or quantitatively analyze polypeptides. A general overview of the applicable technology can be found in Harlow & Lane, *Antibodies. A Laboratory Manual* (1988).

A. Antibodies to Target Polypeptides or Other Immunogens

[0131] Methods for producing polyclonal and monoclonal antibodies that react specifically with a protein of interest or other immunogen are known to those of skill in the art (see, e.g., Coligan, supra; and Harlow and Lane, supra; Stites et al., supra and references cited therein; Goding, supra; and Kohler and Milstein *Nature,* 256:495-497 (1975)). Such techniques include antibody preparation by selection of antibodies from libraries of recombinant antibodies in phage or similar vectors (see, Huse et al., supra; and Ward et al., supra). For example, in order to produce antisera for use in an immunoassay, the protein of interest or an antigenic fragment thereof, is isolated as described herein. For example, a recombinant protein is produced in a transformed cell line. An inbred strain of mice or rabbits is immunized with the protein using a standard adjuvant, such as Freund's adjuvant, and a standard immunization protocol. Alternatively, a synthetic peptide derived from the sequences disclosed herein and conjugated to a carrier protein can be used as an immunogen.

[0132] Polyclonal sera are collected and titered against the immunogen in an immunoassay, for example, a solid phase immunoassay with the immunogen immobilized on a solid support. Polyclonal antisera with a titer of 10^4 or greater are selected and tested for their cross-reactivity against unrelated proteins or even other homologous proteins from other organisms, using a competitive binding immunoassay. Specific monoclonal and polyclonal antibodies and antisera will usually bind with a K_D of at least about 0.1 mM, more usually at least about 1 μM, preferably at least about 0.1 μM or better, and most preferably, 0.01 μM or better.

[0133] A number of proteins of the invention comprising immunogens may be used to produce antibodies specifically or selectively reactive with the proteins of interest. Recombinant protein is the preferred immunogen for the production of monoclonal or polyclonal antibodies. Naturally occurring protein, such as one comprising an amino acid sequence encoded by a gene listed in Table 1-8 may also be used either in pure or impure form. Synthetic peptides made using the protein sequences described herein may also be used as an immunogen for the production of antibodies to the protein. Recombinant protein can be expressed in eukaryotic or prokaryotic cells and purified as generally described supra. The product is then injected into an animal capable of producing antibodies. Either monoclonal or polyclonal antibodies may be generated for subsequent use in immunoassays to measure the protein.

[0134] Methods of production of polyclonal antibodies are known to those of skill in the art. In brief, an immunogen, preferably a purified protein, is mixed with an adjuvant and animals are immunized. The animal's immune response to the immunogen preparation is monitored by taking test bleeds and determining the titer of reactivity to the polypeptide of interest. When appropriately high titers of antibody to the immunogen are obtained, blood is collected from the animal and antisera are prepared. Further fractionation of the antisera to enrich for antibodies reactive to the protein can be done if desired (see, Harlow and Lane, supra).

[0135] Monoclonal antibodies may be obtained using various techniques familiar to those of skill in the art. Typically, spleen cells from an animal immunized with a desired antigen are immortalized, commonly by fusion with a myeloma cell (see, Kohler and Milstein, *Eur. J. Immunol.* 6:511-519 (1976)). Alternative methods of immortalization include, e.g., transformation with Epstein Barr Virus, oncogenes, or retroviruses, or other methods well known in the art. Colonies arising from single immortalized cells are screened for production of antibodies of the desired specificity and affinity for

the antigen, and yield of the monoclonal antibodies produced by such cells may be enhanced by various techniques, including injection into the peritoneal cavity of a vertebrate host. Alternatively, one may isolate DNA sequences which encode a monoclonal antibody or a binding fragment thereof by screening a DNA library from human B cells according to the general protocol outlined by Huse et al., supra.

[0136] Once target protein specific antibodies are available, the protein can be measured by a variety of immunoassay methods with qualitative and quantitative results available to the clinician. For a review of immunological and immunoassay procedures in general see, Stites, supra. Moreover, the immunoassays of the present invention can be performed in any of several configurations, which are reviewed extensively in Maggio Enzyme Immunoassay, CRC Press, Boca Raton, Fla. (1980); Tijssen, supra; and Harlow and Lane, supra.

[0137] Immunoassays to measure target proteins in a human sample may use a polyclonal antiserum that was raised to the protein (e.g., one has an amino acid sequence encoded by a gene listed in Table 1-8) or a fragment thereof. This antiserum is selected to have low cross-reactivity against different proteins and any such cross-reactivity is removed by immunoabsorption prior to use in the immunoassay.

B. Immunological Binding Assays

[0138] In a preferred embodiment, a protein of interest is detected and/or quantified using any of a number of well-known immunological binding assays (see, e.g., U.S. Pat. Nos. 4,366,241; 4,376,110; 4,517,288; and 4,837,168). For a review of the general immunoassays, see also Asai *Methods in Cell Biology Volume* 37: Antibodies in Cell Biology, Academic Press, Inc. NY (1993); Stites, supra. Immunological binding assays (or immunoassays) typically utilize a "capture agent" to specifically bind to and often immobilize the analyte (in this case a polypeptide of the present invention or antigenic subsequences thereof). The capture agent is a moiety that specifically binds to the analyte. In a preferred embodiment, the capture agent is an antibody that specifically binds, for example, a polypeptide of the invention. The antibody may be produced by any of a number of means well known to those of skill in the art and as described above.

[0139] Immunoassays also often utilize a labeling agent to specifically bind to and label the binding complex formed by the capture agent and the analyte. The labeling agent may itself be one of the moieties comprising the antibody/analyte complex. Alternatively, the labeling agent may be a third moiety, such as another antibody, that specifically binds to the antibody/protein complex.

[0140] In a preferred embodiment, the labeling agent is a second antibody bearing a label. Alternatively, the second antibody may lack a label, but it may, in turn, be bound by a labeled third antibody specific to antibodies of the species from which the second antibody is derived. The second antibody can be modified with a detectable moiety, such as biotin, to which a third labeled molecule can specifically bind, such as enzyme-labeled streptavidin.

[0141] Other proteins capable of specifically binding immunoglobulin constant regions, such as protein A or protein G, can also be used as the label agents. These proteins are normal constituents of the cell walls of streptococcal bacteria. They exhibit a strong non-immunogenic reactivity with immunoglobulin constant regions from a variety of species (see, generally, Kronval, et al. *J. Immunol.,* 111:1401-1406 (1973); and Akerstrom, et al. *J. Immunol.,* 135:2589-2542 (1985)).

[0142] Throughout the assays, incubation and/or washing steps may be required after each combination of reagents. Incubation steps can vary from about 5 seconds to several hours, preferably from about 5 minutes to about 24 hours. The incubation time will depend upon the assay format, analyte, volume of solution, concentrations, and the like. Usually, the assays will be carried out at ambient temperature, although they can be conducted over a range of temperatures, such as 10° C. to 40° C.

1. Non-Competitive Assay Formats

[0143] Immunoassays for detecting proteins of interest from tissue samples may be either competitive or noncompetitive. Noncompetitive immunoassays are assays in which the amount of captured analyte (in this case the protein) is directly measured. In one preferred "sandwich" assay, for example, the capture agent (e.g., antibodies specific for a polypeptide encoded by a gene listed in Table 1-8) can be bound directly to a solid substrate where it is immobilized. These immobilized antibodies then capture the polypeptide present in the test sample. The polypeptide thus immobilized is then bound by a labeling agent, such as a second antibody bearing a label. Alternatively, the second antibody may lack a label, but it may, in turn, be bound by a labeled third antibody specific to antibodies of the species from which the second antibody is derived. The second can be modified with a detectable moiety, such as biotin, to which a third labeled molecule can specifically bind, such as enzyme-labeled streptavidin.

2. Competitive Assay Formats

[0144] In competitive assays, the amount of analyte (such as a polypeptide encoded by a gene listed in Table 1-8) present in the sample is measured indirectly by measuring the amount of an added (exogenous) analyte displaced (or competed away) from a capture agent (e.g., an antibody specific for the analyte) by the analyte present in the sample. In one competitive assay, a known amount of, in this case, the protein of interest is added to the sample and the sample is then contacted with a capture agent, in this case an antibody that specifically binds to a polypeptide of the invention. The amount of immunogen bound to the antibody is inversely proportional to the concentration of immunogen present in the sample. In a particularly preferred embodiment, the antibody is immobilized on a solid substrate. For example, the amount of the polypeptide bound to the antibody may be determined either by measuring the amount of subject protein present in a protein/antibody complex or, alternatively, by measuring the amount of remaining uncomplexed protein. The amount of protein may be detected by providing a labeled protein molecule.

[0145] Immunoassays in the competitive binding format can be used for cross-reactivity determinations. For example, a protein of interest can be immobilized on a solid support. Proteins are added to the assay which compete with the binding of the antisera to the immobilized antigen. The ability of the above proteins to compete with the binding of the antisera to the immobilized protein is compared to that of the protein of interest. The percent cross-reactivity for the above proteins is calculated, using standard calculations. Those antisera with

less than 10% cross-reactivity with each of the proteins listed above are selected and pooled. The cross-reacting antibodies are optionally removed from the pooled antisera by immunoabsorption with the considered proteins, e.g., distantly related homologs.

[0146] The immunoabsorbed and pooled antisera are then used in a competitive binding immunoassay as described above to compare a second protein, thought to be perhaps a protein of the present invention, to the immunogen protein. In order to make this comparison, the two proteins are each assayed at a wide range of concentrations and the amount of each protein required to inhibit 50% of the binding of the antisera to the immobilized protein is determined. If the amount of the second protein required is less than 10 times the amount of the protein partially encoded by a sequence herein that is required, then the second protein is said to specifically bind to an antibody generated to an immunogen consisting of the target protein.

3. Other Assay Formats

[0147] In a particularly preferred embodiment, western blot (immunoblot) analysis is used to detect and quantify the presence of a polypeptide of the invention in the sample. The technique generally comprises separating sample proteins by gel electrophoresis on the basis of molecular weight, transferring the separated proteins to a suitable solid support (such as, e.g., a nitrocellulose filter, a nylon filter, or a derivatized nylon filter) and incubating the sample with the antibodies that specifically bind the protein of interest. For example, the antibodies specifically bind to a polypeptide of interest on the solid support. These antibodies may be directly labeled or alternatively may be subsequently detected using labeled antibodies (e.g., labeled sheep anti-mouse antibodies) that specifically bind to the antibodies against the protein of interest.

[0148] Other assay formats include liposome immunoassays (LIA), which use liposomes designed to bind specific molecules (e.g., antibodies) and release encapsulated reagents or markers. The released chemicals are then detected according to standard techniques (see, Monroe et al. (1986) *Amer. Clin. Prod. Rev.* 5:34-41).

4. Labels

[0149] The particular label or detectable group used in the assay is not a critical aspect of the invention, as long as it does not significantly interfere with the specific binding of the antibody used in the assay. The detectable group can be any material having a detectable physical or chemical property. Such detectable labels have been well developed in the field of immunoassays and, in general, most labels useful in such methods can be applied to the present invention. Thus, a label is any composition detectable by spectroscopic, photochemical, biochemical, immunochemical, electrical, optical or chemical means. Useful labels in the present invention include magnetic beads (e.g., Dynabeads™), fluorescent dyes (e.g., fluorescein isothiocyanate, Texas red, rhodamine, and the like), radiolabels (e.g., ^{3}H, ^{125}I, ^{35}S, ^{14}C, or ^{32}P), enzymes (e.g., horse radish peroxidase, alkaline phosphatase and others commonly used in an ELISA), and colorimetric labels such as colloidal gold or colored glass or plastic (e.g., polystyrene, polypropylene, latex, etc.) beads.

[0150] The label may be coupled directly or indirectly to the desired component of the assay according to methods well known in the art. As indicated above, a wide variety of labels may be used, with the choice of label depending on the sensitivity required, the ease of conjugation with the compound, stability requirements, available instrumentation, and disposal provisions.

[0151] Non-radioactive labels are often attached by indirect means. The molecules can also be conjugated directly to signal generating compounds, e.g., by conjugation with an enzyme or fluorescent compound. A variety of enzymes and fluorescent compounds can be used with the methods of the present invention and are well-known to those of skill in the art (for a review of various labeling or signal producing systems which may be used, see, e.g., U.S. Pat. No. 4,391,904).

[0152] Means of detecting labels are well known to those of skill in the art. Thus, for example, where the label is a radioactive label, means for detection include a scintillation counter or photographic film as in autoradiography. Where the label is a fluorescent label, it may be detected by exciting the fluorochrome with the appropriate wavelength of light and detecting the resulting fluorescence. The fluorescence may be detected visually, by means of photographic film, by the use of electronic detectors such as charge-coupled devices (CCDs) or photomultipliers and the like. Similarly, enzymatic labels may be detected by providing the appropriate substrates for the enzyme and detecting the resulting reaction product. Finally simple calorimetric labels may be detected directly by observing the color associated with the label. Thus, in various dipstick assays, conjugated gold often appears pink, while various conjugated beads appear the color of the bead.

[0153] Some assay formats do not require the use of labeled components. For instance, agglutination assays can be used to detect the presence of the target antibodies. In this case, antigen-coated particles are agglutinated by samples comprising the target antibodies. In this format, none of the components need to be labeled and the presence of the target antibody is detected by simple visual inspection.

VI. Screening for Modulators of Polypeptides and Polynucleotides of the Invention

[0154] Modulators of polypeptides or polynucleotides of the invention, i.e. agonists or antagonists of their activity or modulators of polypeptide or polynucleotide expression, are useful for treating a number of human diseases, including mood disorders or psychotic disorders. Administration of agonists, antagonists or other agents that modulate expression of the polynucleotides or polypeptides of the invention can be used to treat patients with mood disorders or psychotic disorders.

A. Screening Methods

[0155] A number of different screening protocols can be utilized to identify agents that modulate the level of expression or activity of polypeptides and polynucleotides of the invention in cells, particularly mammalian cells, and especially human cells. In general terms, the screening methods involve screening a plurality of agents to identify an agent that modulates the polypeptide activity by binding to a polypeptide of the invention, modulating inhibitor binding to the polypeptide or activating expression of the polypeptide or polynucleotide, for example.

1. Binding Assays

[0156] Preliminary screens can be conducted by screening for agents capable of binding to a polypeptide of the inven-

tion, as at least some of the agents so identified are likely modulators of polypeptide activity. The binding assays usually involve contacting a polypeptide of the invention with one or more test agents and allowing sufficient time for the protein and test agents to form a binding complex. Any binding complexes formed can be detected using any of a number of established analytical techniques. Protein binding assays include, but are not limited to, methods that measure co-precipitation, co-migration on non-denaturing SDS-polyacrylamide gels, and co-migration on Western blots (see, e.g., Bennet and Yamamura, (1985) "Neurotransmitter, Hormone or Drug Receptor Binding Methods," in *Neurotransmitter Receptor Binding* (Yamamura, H. I., et al., eds.), pp. 61-89. The protein utilized in such assays can be naturally expressed, cloned or synthesized.

[0157] Binding assays are also useful, e.g., for identifying endogenous proteins that interact with a polypeptide of the invention. For example, antibodies, receptors or other molecules that bind a polypeptide of the invention can be identified in binding assays.

2. Expression Assays

[0158] Certain screening methods involve screening for a compound that up or down-regulates the expression of a polypeptide or polynucleotide of the invention. Such methods generally involve conducting cell-based assays in which test compounds are contacted with one or more cells expressing a polypeptide or polynucleotide of the invention and then detecting an increase or decrease in expression (either transcript, translation product, or catalytic product). Some assays are performed with peripheral cells, or other cells, that express an endogenous polypeptide or polynucleotide of the invention.

[0159] Polypeptide or polynucleotide expression can be detected in a number of different ways. As described infra, the expression level of a polynucleotide of the invention in a cell can be determined by probing the mRNA expressed in a cell with a probe that specifically hybridizes with a transcript (or complementary nucleic acid derived therefrom) of a polynucleotide of the invention. Probing can be conducted by lysing the cells and conducting Northern blots or without lysing the cells using in situ-hybridization techniques. Alternatively, a polypeptide of the invention can be detected using immunological methods in which a cell lysate is probed with antibodies that specifically bind to a polypeptide of the invention.

[0160] Other cell-based assays are reporter assays conducted with cells that do not express a polypeptide or polynucleotide of the invention. Certain of these assays are conducted with a heterologous nucleic acid construct that includes a promoter of a polynucleotide of the invention that is operably linked to a reporter gene that encodes a detectable product. A number of different reporter genes can be utilized. Some reporters are inherently detectable. An example of such a reporter is green fluorescent protein that emits fluorescence that can be detected with a fluorescence detector. Other reporters generate a detectable product. Often such reporters are enzymes. Exemplary enzyme reporters include, but are not limited to, β-glucuronidase, chloramphenicol acetyl transferase (CAT); Alton and Vapnek (1979) *Nature* 282:864-869), luciferase, β-galactosidase, green fluorescent protein (GFP) and alkaline phosphatase (Toh, et al. (1980) *Eur. J. Biochem.* 182:231-238; and Hall et al. (1983) *J. Mol. Appl. Gen.* 2:101).

[0161] In these assays, cells harboring the reporter construct are contacted with a test compound. A test compound that either activates the promoter by binding to it or triggers a cascade that produces a molecule that activates the promoter causes expression of the detectable reporter. Certain other reporter assays are conducted with cells that harbor a heterologous construct that includes a transcriptional control element that activates expression of a polynucleotide of the invention and a reporter operably linked thereto. Here, too, an agent that binds to the transcriptional control element to activate expression of the reporter or that triggers the formation of an agent that binds to the transcriptional control element to activate reporter expression, can be identified by the generation of signal associated with reporter expression.

[0162] The level of expression or activity can be compared to a baseline value. As indicated above, the baseline value can be a value for a control sample or a statistical value that is representative of expression levels for a control population (e.g., healthy individuals not having or at risk for mood disorders or psychotic disorders). Expression levels can also be determined for cells that do not express a polynucleotide of the invention as a negative control. Such cells generally are otherwise substantially genetically the same as the test cells.

[0163] A variety of different types of cells can be utilized in the reporter assays. Cells that express an endogenous polypeptide or polynucleotide of the invention include, e.g., brain cells, including cells from the cerebellum, anterior cingulate cortex, or dorsolateral prefrontal cortex. Such brain regions are part of brain circuits or pathways that are implicated in mood disorders. Cells that do not endogenously express polynucleotides of the invention can be prokaryotic, but are preferably eukaryotic. The eukaryotic cells can be any of the cells typically utilized in generating cells that harbor recombinant nucleic acid constructs. Exemplary eukaryotic cells include, but are not limited to, yeast, and various higher eukaryotic cells such as the COS, CHO and HeLa cell lines, and stem cells.

[0164] Various controls can be conducted to ensure that an observed activity is authentic including running parallel reactions with cells that lack the reporter construct or by not contacting a cell harboring the reporter construct with test compound. Compounds can also be further validated as described below.

3. Catalytic Activity

[0165] Catalytic activity of polypeptides of the invention can be determined by measuring the production of enzymatic products or by measuring the consumption of substrates. Activity refers to either the rate of catalysis or the ability to the polypeptide to bind (K_m) the substrate or release the catalytic product (K_d).

[0166] Analysis of the activity of polypeptides of the invention are performed according to general biochemical analyses. Such assays include cell-based assays as well as in vitro assays involving purified or partially purified polypeptides or crude cell lysates. The assays generally involve providing a known quantity of substrate and quantifying product as a function of time.

4. Validation

[0167] Agents that are initially identified by any of the foregoing screening methods can be further tested to validate the apparent activity. Preferably such studies are conducted

with suitable animal models. The basic format of such methods involves administering a lead compound identified during an initial screen to an animal that serves as a model for humans and then determining if expression or activity of a polynucleotide or polypeptide of the invention is in fact upregulated. The animal models utilized in validation studies generally are mammals of any kind. Specific examples of suitable animals include, but are not limited to, primates, mice, and rats.

5. Animal Models

[0168] Animal models of mental disorders also find use in screening for modulators. In one embodiment, rat models of depression (both chronic and acute), in which the rats are subjected to stress, are used for screening. In one embodiment, invertebrate models such as Drosophila models can be used, screening for modulators of Drosophila orthologs of the human genes disclosed herein. In another embodiment, transgenic animal technology including gene knockout technology, for example as a result of homologous recombination with an appropriate gene targeting vector, or gene overexpression, will result in the absence, decreased or increased expression of a polynucleotide or polypeptide of the invention. The same technology can also be applied to make knockout cells. When desired, tissue-specific expression or knockout of a polynucleotide or polypeptide of the invention may be necessary. Transgenic animals generated by such methods find use as animal models of mental disorders and are useful in screening for modulators of mental disorders.

[0169] Knockout cells and transgenic mice can be made by insertion of a marker gene or other heterologous gene into an endogenous gene site in the mouse genome via homologous recombination. Such mice can also be made by substituting an endogenous polynucleotide of the invention with a mutated version of the polynucleotide, or by mutating an endogenous polynucleotide, e.g., by exposure to carcinogens.

[0170] For development of appropriate stem cells, a DNA construct is introduced into the nuclei of embryonic stem cells. Cells containing the newly engineered genetic lesion are injected into a host mouse embryo, which is re-implanted into a recipient female. Some of these embryos develop into chimeric mice that possess germ cells partially derived from the mutant cell line. Therefore, by breeding the chimeric mice it is possible to obtain a new line of mice containing the introduced genetic lesion (see, e.g., Capecchi et al., *Science* 244:1288 (1989)). Chimeric targeted mice can be derived according to Hogan et al., *Manipulating the Mouse Embryo: A Laboratory Manual*, Cold Spring Harbor Laboratory (1988) and *Teratocarcinomas and Embryonic Stem Cells: A Practical Approach*, Robertson, ed., IRL Press, Washington, D.C., (1987).

B. Modulators of Polypeptides or Polynucleotides of the Invention

[0171] The agents tested as modulators of the polypeptides or polynucleotides of the invention can be any small chemical compound, or a biological entity, such as a protein, sugar, nucleic acid or lipid. Alternatively, modulators can be genetically altered versions of a polypeptide or polynucleotide of the invention. Typically, test compounds will be small chemical molecules and peptides. Essentially any chemical compound can be used as a potential modulator or ligand in the assays of the invention, although most often compounds that can be dissolved in aqueous or organic (especially DMSO-based) solutions are used. The assays are designed to screen large chemical libraries by automating the assay steps and providing compounds from any convenient source to assays, which are typically run in parallel (e.g., in microtiter formats on microtiter plates in robotic assays). It will be appreciated that there are many suppliers of chemical compounds, including Sigma (St. Louis, Mo.), Aldrich (St. Louis, Mo.), Sigma-Aldrich (St. Louis, Mo.), Fluka Chemika-Biochemica Analytika (Buchs, Switzerland) and the like. Modulators also include agents designed to reduce the level of mRNA of the invention (e.g. antisense molecules, ribozymes, DNAzymes and the like) or the level of translation from an mRNA.

[0172] In one preferred embodiment, high throughput screening methods involve providing a combinatorial chemical or peptide library containing a large number of potential therapeutic compounds (potential modulator or ligand compounds). Such "combinatorial chemical libraries" or "ligand libraries" are then screened in one or more assays, as described herein, to identify those library members (particular chemical species or subclasses) that display a desired characteristic activity. The compounds thus identified can serve as conventional "lead compounds" or can themselves be used as potential or actual therapeutics.

[0173] A combinatorial chemical library is a collection of diverse chemical compounds generated by either chemical synthesis or biological synthesis, by combining a number of chemical "building blocks" such as reagents. For example, a linear combinatorial chemical library such as a polypeptide library is formed by combining a set of chemical building blocks (amino acids) in every possible way for a given compound length (i.e., the number of amino acids in a polypeptide compound). Millions of chemical compounds can be synthesized through such combinatorial mixing of chemical building blocks.

[0174] Preparation and screening of combinatorial chemical libraries is well known to those of skill in the art. Such combinatorial chemical libraries include, but are not limited to, peptide libraries (see, e.g., U.S. Pat. No. 5,010,175, Furka, *Int. J. Pept. Prot. Res.* 37:487-493 (1991) and Houghton et al., *Nature* 354:84-88 (1991)). Other chemistries for generating chemical diversity libraries can also be used. Such chemistries include, but are not limited to: peptoids (e.g., PCT Publication No. WO 91/19735), encoded peptides (e.g., PCT Publication WO 93/20242), random bio-oligomers (e.g., PCT Publication No. WO 92/00091), benzodiazepines (e.g., U.S. Pat. No. 5,288,514), diversomers such as hydantoins, benzodiazepines and dipeptides (Hobbs et al., *Proc. Nat. Acad. Sci. USA* 90:6909-6913 (11993)), vinylogous polypeptides (Hagihara et al., *J. Amer. Chem. Soc.* 114:6568 (1992)), non-peptidal peptidomimetics with glucose scaffolding (Hirschmann et al., *J. Amer. Chem. Soc.* 114:9217-9218 (1992)), analogous organic syntheses of small compound libraries (Chen et al., *J. Amer. Chem. Soc.* 116:2661 (1994)), oligocarbamates (Cho et al., *Science* 261:1303 (1993)), and/or peptidyl phosphonates (Campbell et al., *J. Org. Chem.* 59:658 (1994)), nucleic acid libraries (see Ausubel, Berger and Sambrook, all supra), peptide nucleic acid libraries (see, e.g., U.S. Pat. No. 5,539,083), antibody libraries (see, e.g., Vaughn et al., *Nature Biotechnology,* 14(3):309-314 (1996) and PCT/US96/10287), carbohydrate libraries (see, e.g., Liang et al., *Science,* 274:1520-1522 (1996) and U.S. Pat. No. 5,593,853), small organic molecule libraries (see, e.g., benzodiazepines, Baum C&EN, January 18, page 33 (1993); isoprenoids, U.S.

Pat. No. 5,569,588; thiazolidinones and metathiazanones, U.S. Pat. No. 5,549,974; pyrrolidines, U.S. Pat. Nos. 5,525,735 and 5,519,134; morpholino compounds, U.S. Pat. Nos. 5,506,337; benzodiazepines, 5,288,514, and the like).

[0175] Devices for the preparation of combinatorial libraries are commercially available (see, e.g., 357 MPS, 390 MPS, Advanced Chem Tech, Louisville Ky.; Symphony, Rainin, Woburn, Mass.; 433A Applied Biosystems, Foster City, Calif.; 9050 Plus, Millipore, Bedford, Mass.). In addition, numerous combinatorial libraries are themselves commercially available (see, e.g., ComGenex, Princeton, N.J.; Tripos, Inc., St. Louis, Mo.; 3D Pharmaceuticals, Exton, Pa.; Martek Biosciences, Columbia, Md., etc.).

C. Solid State and Soluble High Throughput Assays

[0176] In the high throughput assays of the invention, it is possible to screen up to several thousand different modulators or ligands in a single day. In particular, each well of a microtiter plate can be used to run a separate assay against a selected potential modulator, or, if concentration or incubation time effects are to be observed, every 5-10 wells can test a single modulator. Thus, a single standard microtiter plate can assay about 100 (e.g., 96) modulators. If 1536 well plates are used, then a single plate can easily assay from about 100 to about 1500 different compounds. It is possible to assay several different plates per day; assay screens for up to about 6,000-20,000 different compounds are possible using the integrated systems of the invention. More recently, microfluidic approaches to reagent manipulation have been developed.

[0177] The molecule of interest can be bound to the solid state component, directly or indirectly, via covalent or non-covalent linkage, e.g., via a tag. The tag can be any of a variety of components. In general, a molecule that binds the tag (a tag binder) is fixed to a solid support, and the tagged molecule of interest is attached to the solid support by interaction of the tag and the tag binder.

[0178] A number of tags and tag binders can be used, based upon known molecular interactions well described in the literature. For example, where a tag has a natural binder, for example, biotin, protein A, or protein G, it can be used in conjunction with appropriate tag binders (avidin, streptavidin, neutravidin, the Fc region of an immunoglobulin, etc.). Antibodies to molecules with natural binders such as biotin are also widely available and appropriate tag binders (see, SIGMA Immunochemicals 1998 catalogue SIGMA, St. Louis Mo.).

[0179] Similarly, any haptenic or antigenic compound can be used in combination with an appropriate antibody to form a tag/tag binder pair. Thousands of specific antibodies are commercially available and many additional antibodies are described in the literature. For example, in one common configuration, the tag is a first antibody and the tag binder is a second antibody which recognizes the first antibody. In addition to antibody-antigen interactions, receptor-ligand interactions are also appropriate as tag and tag-binder pairs, such as agonists and antagonists of cell membrane receptors (e.g., cell receptor-ligand interactions such as transferrin, c-kit, viral receptor ligands, cytokine receptors, chemokine receptors, interleukin receptors, immunoglobulin receptors and antibodies, the cadherin family, the integrin family, the selectin family, and the like; see, e.g., Pigott & Power, *The Adhesion Molecule Facts Book I* (1993)). Similarly, toxins and venoms, viral epitopes, hormones (e.g., opiates, steroids, etc.), intracellular receptors (e.g., which mediate the effects of various small ligands, including steroids, thyroid hormone, retinoids and vitamin D; peptides), drugs, lectins, sugars, nucleic acids (both linear and cyclic polymer configurations), oligosaccharides, proteins, phospholipids and antibodies can all interact with various cell receptors.

[0180] Synthetic polymers, such as polyurethanes, polyesters, polycarbonates, polyureas, polyamides, polyethyleneimines, polyarylene sulfides, polysiloxanes, polyimides, and polyacetates can also form an appropriate tag or tag binder. Many other tag/tag binder pairs are also useful in assay systems described herein, as would be apparent to one of skill upon review of this disclosure.

[0181] Common linkers such as peptides, polyethers, and the like can also serve as tags, and include polypeptide sequences, such as poly-Gly sequences of between about 5 and 200 amino acids. Such flexible linkers are known to those of skill in the art. For example, poly(ethelyne glycol) linkers are available from Shearwater Polymers, Inc., Huntsville, Ala. These linkers optionally have amide linkages, sulfhydryl linkages, or heterofunctional linkages.

[0182] Tag binders are fixed to solid substrates using any of a variety of methods currently available. Solid substrates are commonly derivatized or functionalized by exposing all or a portion of the substrate to a chemical reagent which fixes a chemical group to the surface which is reactive with a portion of the tag binder. For example, groups which are suitable for attachment to a longer chain portion would include amines, hydroxyl, thiol, and carboxyl groups. Aminoalkylsilanes and hydroxyalkylsilanes can be used to functionalize a variety of surfaces, such as glass surfaces. The construction of such solid phase biopolymer arrays is well described in the literature (see, e.g., Merrifield, *J. Am. Chem. Soc.* 85:2149-2154 (1963) (describing solid phase synthesis of, e.g., peptides); Geysen et al., *J. Immun. Meth.* 102:259-274 (1987) (describing synthesis of solid phase components on pins); Frank and Doring, *Tetrahedron* 44:60316040 (1988) (describing synthesis of various peptide sequences on cellulose disks); Fodor et al., *Science,* 251:767-777 (1991); Sheldon et al., *Clinical Chemistry* 39(4):718-719 (1993); and Kozal et al., *Nature Medicine* 2(7):753759 (1996) (all describing arrays of biopolymers fixed to solid substrates). Non-chemical approaches for fixing tag binders to substrates include other common methods, such as heat, cross-linking by UV radiation, and the like.

[0183] The invention provides in vitro assays for identifying, in a high throughput format, compounds that can modulate the expression or activity of the polynucleotides or polypeptides of the invention. In a preferred embodiment, the methods of the invention include such a control reaction. For each of the assay formats described, "no modulator" control reactions that do not include a modulator provide a background level of binding activity.

[0184] In some assays it will be desirable to have positive controls to ensure that the components of the assays are working properly. At least two types of positive controls are appropriate. First, a known activator of a polynucleotide or polypeptide of the invention can be incubated with one sample of the assay, and the resulting increase in signal resulting from an increased expression level or activity of polynucleotide or polypeptide determined according to the methods herein. Second, a known inhibitor of a polynucleotide or

polypeptide of the invention can be added, and the resulting decrease in signal for the expression or activity can be similarly detected.

D. Computer-Based Assays

[0185] Yet another assay for compounds that modulate the activity of a polypeptide or polynucleotide of the invention involves computer assisted drug design, in which a computer system is used to generate a three-dimensional structure of the polypeptide or polynucleotide based on the structural information encoded by its amino acid or nucleotide sequence. The input sequence interacts directly and actively with a pre-established algorithm in a computer program to yield secondary, tertiary, and quaternary structural models of the molecule. Similar analyses can be performed on potential receptors or binding partners of the polypeptides or polynucleotides of the invention. The models of the protein or nucleotide structure are then examined to identify regions of the structure that have the ability to bind, e.g., a polypeptide or polynucleotide of the invention. These regions are then used to identify polypeptides that bind to a polypeptide or polynucleotide of the invention.

[0186] The three-dimensional structural model of a protein is generated by entering protein amino acid sequences of at least 10 amino acid residues or corresponding nucleic acid sequences encoding a potential receptor into the computer system. The amino acid sequences encoded by the nucleic acid sequences provided herein represent the primary sequences or subsequences of the proteins, which encode the structural information of the proteins. At least 10 residues of an amino acid sequence (or a nucleotide sequence encoding 10 amino acids) are entered into the computer system from computer keyboards, computer readable substrates that include, but are not limited to, electronic storage media (e.g., magnetic diskettes, tapes, cartridges, and chips), optical media (e.g., CD ROM), information distributed by internet sites, and by RAM. The three-dimensional structural model of the protein is then generated by the interaction of the amino acid sequence and the computer system, using software known to those of skill in the art.

[0187] The amino acid sequence represents a primary structure that encodes the information necessary to form the secondary, tertiary, and quaternary structure of the protein of interest. The software looks at certain parameters encoded by the primary sequence to generate the structural model. These parameters are referred to as "energy terms," and primarily include electrostatic potentials, hydrophobic potentials, solvent accessible surfaces, and hydrogen bonding. Secondary energy terms include van der Waals potentials. Biological molecules form the structures that minimize the energy terms in a cumulative fashion. The computer program is therefore using these terms encoded by the primary structure or amino acid sequence to create the secondary structural model.

[0188] The tertiary structure of the protein encoded by the secondary structure is then formed on the basis of the energy terms of the secondary structure. The user at this point can enter additional variables such as whether the protein is membrane bound or soluble, its location in the body, and its cellular location, e.g., cytoplasmic, surface, or nuclear. These variables along with the energy terms of the secondary structure are used to form the model of the tertiary structure. In modeling the tertiary structure, the computer program matches hydrophobic faces of secondary structure with like, and hydrophilic faces of secondary structure with like.

[0189] Once the structure has been generated, potential ligand binding regions are identified by the computer system. Three-dimensional structures for potential ligands are generated by entering amino acid or nucleotide sequences or chemical formulas of compounds, as described above. The three-dimensional structure of the potential ligand is then compared to that of a polypeptide or polynucleotide of the invention to identify binding sites of the polypeptide or polynucleotide of the invention. Binding affinity between the protein and ligands is determined using energy terms to determine which ligands have an enhanced probability of binding to the protein.

[0190] Computer systems are also used to screen for mutations, polymorphic variants, alleles and interspecies homologs of genes encoding a polypeptide or polynucleotide of the invention. Such mutations can be associated with disease states or genetic traits and can be used for diagnosis. As described above, GeneChip™ and related technology can also be used to screen for mutations, polymorphic variants, alleles and interspecies homologs. Once the variants are identified, diagnostic assays can be used to identify patients having such mutated genes. Identification of the mutated a polypeptide or polynucleotide of the invention involves receiving input of a first amino acid sequence of a polypeptide of the invention (or of a first nucleic acid sequence encoding a polypeptide of the invention), e.g., any amino acid sequence having at least 60%, optionally at least 70% or 85%, identity with the amino acid sequence of interest, or conservatively modified versions thereof. The sequence is entered into the computer system as described above. The first nucleic acid or amino acid sequence is then compared to a second nucleic acid or amino acid sequence that has substantial identity to the first sequence. The second sequence is entered into the computer system in the manner described above. Once the first and second sequences are compared, nucleotide or amino acid differences between the sequences are identified. Such sequences can represent allelic differences in various polynucleotides, including SNPs and/or haplotypes, of the invention, and mutations associated with disease states and genetic traits.

VII. Compositions, Kits and Integrated Systems

[0191] The invention provides compositions, kits and integrated systems for practicing the assays described herein using polypeptides or polynucleotides of the invention, antibodies specific for polypeptides or polynucleotides of the invention, etc.

[0192] The invention provides assay compositions for use in solid phase assays; such compositions can include, for example, one or more polynucleotides or polypeptides of the invention immobilized on a solid support, and a labeling reagent. In each case, the assay compositions can also include additional reagents that are desirable for hybridization. Modulators of expression or activity of polynucleotides or polypeptides of the invention can also be included in the assay compositions.

[0193] The invention also provides kits for carrying out the therapeutic and diagnostic assays of the invention. The kits typically include a probe that comprises an antibody that specifically binds to polypeptides or polynucleotides of the invention, and a label for detecting the presence of the probe. The kits may include several polynucleotide sequences encoding polypeptides of the invention. Kits can include any of the compositions noted above, and optionally further

include additional components such as instructions to practice a high-throughput method of assaying for an effect on expression of the genes encoding the polypeptides of the invention, or on activity of the polypeptides of the invention, one or more containers or compartments (e.g., to hold the probe, labels, or the like), a control modulator of the expression or activity of polypeptides of the invention, a robotic armature for mixing kit components or the like.

[0194] The invention also provides integrated systems for high-throughput screening of potential modulators for an effect on the expression or activity of the polypeptides of the invention. The systems typically include a robotic armature which transfers fluid from a source to a destination, a controller which controls the robotic armature, a label detector, a data storage unit which records label detection, and an assay component such as a microtiter dish comprising a well having a reaction mixture or a substrate comprising a fixed nucleic acid or immobilization moiety.

[0195] A number of robotic fluid transfer systems are available, or can easily be made from existing components. For example, a Zymate XP (Zymark Corporation; Hopkinton, Mass.) automated robot using a Microlab 2200 (Hamilton; Reno, Nev.) pipetting station can be used to transfer parallel samples to 96 well microtiter plates to set up several parallel simultaneous STAT binding assays.

[0196] Optical images viewed (and, optionally, recorded) by a camera or other recording device (e.g., a photodiode and data storage device) are optionally further processed in any of the embodiments herein, e.g., by digitizing the image and storing and analyzing the image on a computer. A variety of commercially available peripheral equipment and software is available for digitizing, storing and analyzing a digitized video or digitized optical image, e.g., using PC, MACINTOSH®, or UNIX® based (e.g., SUN® work station) computers.

[0197] One conventional system carries light from the specimen field to a cooled charge-coupled device (CCD) camera, in common use in the art. A CCD camera includes an array of picture elements (pixels). The light from the specimen is imaged on the CCD. Particular pixels corresponding to regions of the specimen (e.g., individual hybridization sites on an array of biological polymers) are sampled to obtain light intensity readings for each position. Multiple pixels are processed in parallel to increase speed. The apparatus and methods of the invention are easily used for viewing any sample, e.g., by fluorescent or dark field microscopic techniques. Lasar based systems can also be used.

VIII. Administration and Pharmaceutical Compositions

[0198] Modulators of the polynucleotides or polypeptides of the invention (e.g., antagonists or agonists) can be administered directly to a mammalian subject for modulation of activity of those molecules in vivo. Administration is by any of the routes normally used for introducing a modulator compound into ultimate contact with the tissue to be treated and is well known to those of skill in the art. Although more than one route can be used to administer a particular composition, a particular route can often provide a more immediate and more effective reaction than another route.

[0199] Diseases that can be treated include the following, which include the corresponding reference number from Morrison, *DSM-IV Made Easy,* 1995: Schizophrenia, Catatonic, Subchronic, (295.21); Schizophrenia, Catatonic, Chronic (295.22); Schizophrenia, Catatonic, Subchronic with Acute Exacerbation (295.23); Schizophrenia, Catatonic, Chronic with Acute Exacerbation (295.24); Schizophrenia, Catatonic, in Remission (295.55); Schizophrenia, Catatonic, Unspecified (295.20); Schizophrenia, Disorganized, Subchronic (295.11); Schizophrenia, Disorganized, Chronic (295.12); Schizophrenia, Disorganized, Subchronic with Acute Exacerbation (295.13); Schizophrenia, Disorganized, Chronic with Acute Exacerbation (295.14); Schizophrenia, Disorganized, in Remission (295.15); Schizophrenia, Disorganized, Unspecified (295.10); Schizophrenia, Paranoid, Subchronic (295.31); Schizophrenia, Paranoid, Chronic (295.32); Schizophrenia, Paranoid, Subchronic with Acute Exacerbation (295.33); Schizophrenia, Paranoid, Chronic with Acute Exacerbation (295.34); Schizophrenia, Paranoid, in Remission (295.35); Schizophrenia, Paranoid, Unspecified (295.30); Schizophrenia, Undifferentiated, Subchronic (295.91); Schizophrenia, Undifferentiated, Chronic (295.92); Schizophrenia, Undifferentiated, Subchronic with Acute Exacerbation (295.93); Schizophrenia, Undifferentiated, Chronic with Acute Exacerbation (295.94); Schizophrenia, Undifferentiated, in Remission (295.95); Schizophrenia, Undifferentiated, Unspecified (295.90); Schizophrenia, Residual, Subchronic (295.61); Schizophrenia, Residual, Chronic (295.62); Schizophrenia, Residual, Subchronic with Acute Exacerbation (295.63); Schizophrenia, Residual, Chronic with Acute Exacerbation (295.94); Schizophrenia, Residual, in Remission (295.65); Schizophrenia, Residual, Unspecified (295.60); Delusional (Paranoid) Disorder (297.10); Brief Reactive Psychosis (298.80); Schizophreniform Disorder (295.40); Schizoaffective Disorder (295.70); Induced Psychotic Disorder (297.30); Psychotic Disorder NOS (Atypical Psychosis) (298.90); Personality Disorders, Paranoid (301.00); Personality Disorders, Schizoid (301.20); Personality Disorders, Schizotypal (301.22); Personality Disorders, Antisocial (301.70); Personality Disorders, Borderline (301.83) and bipolar disorders, maniac, hypomaniac, dysthymic or cyclothymic disorders, substance-induced mood disorders, major depression, psychotic disorders, including paranoid psychosis, catatonic psychosis, delusional psychosis, having schizoaffective disorder, and substance-induced psychotic disorder.

[0200] In some embodiments, modulators of polynucleotides or polypeptides of the invention can be combined with other drugs useful for treating mental disorders including useful for treating mood disorders, e.g., schizophrenia, bipolar disorders, or major depression. In some preferred embodiments, pharmaceutical compositions of the invention comprise a modulator of a polypeptide of polynucleotide of the invention combined with at least one of the compounds useful for treating schizophrenia, bipolar disorder, or major depression, e.g., such as those described in U.S. Pat. Nos. 6,297,262; 6,284,760; 6,284,771; 6,232,326; 6,187,752; 6,117,890; 6,239,162 or 6,166,008.

[0201] The pharmaceutical compositions of the invention may comprise a pharmaceutically acceptable carrier. Pharmaceutically acceptable carriers are determined in part by the particular composition being administered, as well as by the particular method used to administer the composition. Accordingly, there is a wide variety of suitable formulations of pharmaceutical compositions of the present invention (see, e.g., *Remington's Pharmaceutical Sciences,* 17^{th} ed. 1985)).

[0202] The modulators (e.g., agonists or antagonists) of the expression or activity of the a polypeptide or polynucleotide

of the invention, alone or in combination with other suitable components, can be made into aerosol formulations (i.e., they can be "nebulized") to be administered via inhalation or in compositions useful for injection. Aerosol formulations can be placed into pressurized acceptable propellants, such as dichlorodifluoromethane, propane, nitrogen, and the like.

[0203] Formulations suitable for administration include aqueous and non-aqueous solutions, isotonic sterile solutions, which can contain antioxidants, buffers, bacteriostats, and solutes that render the formulation isotonic, and aqueous and non-aqueous sterile suspensions that can include suspending agents, solubilizers, thickening agents, stabilizers, and preservatives. In the practice of this invention, can be administered or example, orally, nasally, topically, intravenously, intraperitoneally, or intrathecally. The formulations of compounds can be presented in unit-dose or multi-dose sealed containers, such as ampoules and vials. Solutions and suspensions can be prepared from sterile powders, granules, and tablets of the kind previously described. The modulators can also be administered as part of a prepared food or drug.

[0204] The dose administered to a patient, in the context of the present invention should be sufficient to effect a beneficial response in the subject over time. The optimal dose level for any patient will depend on a variety of factors including the efficacy of the specific modulator employed, the age, body weight, physical activity, and diet of the patient, on a possible combination with other drugs, and on the severity of the mental disorder. The size of the dose also will be determined by the existence, nature, and extent of any adverse side effects that accompany the administration of a particular compound or vector in a particular subject.

[0205] In determining the effective amount of the modulator to be administered a physician may evaluate circulating plasma levels of the modulator, modulator toxicity, and the production of anti-modulator antibodies. In general, the dose equivalent of a modulator is from about 1 ng/kg to 10 mg/kg for a typical subject.

[0206] For administration, modulators of the present invention can be administered at a rate determined by the LD-50 of the modulator, and the side effects of the modulator at various concentrations, as applied to the mass and overall health of the subject. Administration can be accomplished via single or divided doses.

IX. Gene Therapy Applications

[0207] A variety of human diseases can be treated by therapeutic approaches that involve stably introducing a gene into a human cell such that the gene is transcribed and the gene product is produced in the cell. Diseases amenable to treatment by this approach include inherited diseases, including those in which the defect is in a single or multiple genes. Gene therapy is also useful for treatment of acquired diseases and other conditions. For discussions on the application of gene therapy towards the treatment of genetic as well as acquired diseases, see, Miller, *Nature* 357:455-460 (1992); and Mulligan, *Science* 260:926-932 (1993).

[0208] In the context of the present invention, gene therapy can be used for treating a variety of disorders and/or diseases in which the polynucleotides and polypeptides of the invention has been implicated. For example, compounds, including polynucleotides, can be identified by the methods of the present invention as effective in treating a mental disorder. Introduction by gene therapy of these polynucleotides can then be used to treat, e.g., mental disorders including mood disorders and psychotic disorders.

A. Vectors for Gene Delivery

[0209] For delivery to a cell or organism, the polynucleotides of the invention can be incorporated into a vector. Examples of vectors used for such purposes include expression plasmids capable of directing the expression of the nucleic acids in the target cell. In other instances, the vector is a viral vector system wherein the nucleic acids are incorporated into a viral genome that is capable of transfecting the target cell. In a preferred embodiment, the polynucleotides can be operably linked to expression and control sequences that can direct expression of the gene in the desired target host cells. Thus, one can achieve expression of the nucleic acid under appropriate conditions in the target cell.

B. Gene Delivery Systems

[0210] Viral vector systems useful in the expression of the nucleic acids include, for example, naturally occurring or recombinant viral vector systems. Depending upon the particular application, suitable viral vectors include replication competent, replication deficient, and conditionally replicating viral vectors. For example, viral vectors can be derived from the genome of human or bovine adenoviruses, vaccinia virus, herpes virus, adeno-associated virus, minute virus of mice (MVM), HIV, sindbis virus, and retroviruses (including but not limited to Rous sarcoma virus), and MoMLV. Typically, the genes of interest are inserted into such vectors to allow packaging of the gene construct, typically with accompanying viral DNA, followed by infection of a sensitive host cell and expression of the gene of interest.

[0211] As used herein, "gene delivery system" refers to any means for the delivery of a nucleic acid of the invention to a target cell. In some embodiments of the invention, nucleic acids are conjugated to a cell receptor ligand for facilitated uptake (e.g., invagination of coated pits and internalization of the endosome) through an appropriate linking moiety, such as a DNA linking moiety (Wu et al., *J. Biol. Chem.* 263:14621-14624 (1988); WO 92/06180). For example, nucleic acids can be linked through a polylysine moiety to asialo-oromucocid, which is a ligand for the asialoglycoprotein receptor of hepatocytes.

[0212] Similarly, viral envelopes used for packaging gene constructs that include the nucleic acids of the invention can be modified by the addition of receptor ligands or antibodies specific for a receptor to permit receptor-mediated endocytosis into specific cells (see, e.g., WO 93/20221, WO 93/14188, and WO 94/06923). In some embodiments of the invention, the DNA constructs of the invention are linked to viral proteins, such as adenovirus particles, to facilitate endocytosis (Curiel et al., *Proc. Natl. Acad. Sci. U.S.A.* 88:8850-8854 (1991)). In other embodiments, molecular conjugates of the instant invention can include microtubule inhibitors (WO/9406922), synthetic peptides mimicking influenza virus hemagglutinin (Plank et al., *J. Biol. Chem.* 269:12918-12924 (1994)), and nuclear localization signals such as SV40 T antigen (WO93/19768).

[0213] Retroviral vectors are also useful for introducing the nucleic acids of the invention into target cells or organisms. Retroviral vectors are produced by genetically manipulating retroviruses. The viral genome of retroviruses is RNA. Upon infection, this genomic RNA is reverse transcribed into a

DNA copy which is integrated into the chromosomal DNA of transduced cells with a high degree of stability and efficiency. The integrated DNA copy is referred to as a provirus and is inherited by daughter cells as is any other gene. The wild type retroviral genome and the proviral DNA have three genes: the gag, the pol and the env genes, which are flanked by two long terminal repeat (LTR) sequences. The gag gene encodes the internal structural (nucleocapsid) proteins; the pol gene encodes the RNA directed DNA polymerase (reverse transcriptase); and the env gene encodes viral envelope glycoproteins. The 5' and 3' LTRs serve to promote transcription and polyadenylation of virion RNAs. Adjacent to the 5' LTR are sequences necessary for reverse transcription of the genome (the tRNA primer binding site) and for efficient encapsulation of viral RNA into particles (the Psi site) (see, Mulligan, In: *Experimental Manipulation of Gene Expression*, Inouye (ed), 155-173 (1983); Mann et al., Cell 33:153-159 (1983); Cone and Mulligan, *Proceedings of the National Academy of Sciences, U.S.A.,* 81:6349-6353 (1984)).

[0214] The design of retroviral vectors is well known to those of ordinary skill in the art. In brief, if the sequences necessary for encapsidation (or packaging of retroviral RNA into infectious virions) are missing from the viral genome, the result is a cis-acting defect which prevents encapsidation of genomic RNA. However, the resulting mutant is still capable of directing the synthesis of all virion proteins. Retroviral genomes from which these sequences have been deleted, as well as cell lines containing the mutant genome stably integrated into the chromosome are well known in the art and are used to construct retroviral vectors. Preparation of retroviral vectors and their uses are described in many publications including, e.g., European Patent Application EPA 0 178 220; U.S. Pat. No. 4,405,712, Gilboa *Biotechniques* 4:504-512 (1986); Mann et al., *Cell* 33:153-159 (1983); Cone and Mulligan *Proc. Natl. Acad. Sci. USA* 81:6349-6353 (1984); Eglitis et al. *Biotechniques* 6:608-614 (1988); Miller et al. *Biotechniques* 7:981-990 (1989); Miller (1992) sipra; Mulligan (1993), supra; and WO 92/07943.

[0215] The retroviral vector particles are prepared by recombinantly inserting the desired nucleotide sequence into a retrovirus vector and packaging the vector with retroviral capsid proteins by use of a packaging cell line. The resultant retroviral vector particle is incapable of replication in the host cell but is capable of integrating into the host cell genome as a proviral sequence containing the desired nucleotide sequence. As a result, the patient is capable of producing, for example, a polypeptide or polynucleotide of the invention and thus restore the cells to a normal phenotype.

[0216] Packaging cell lines that are used to prepare the retroviral vector particles are typically recombinant mammalian tissue culture cell lines that produce the necessary viral structural proteins required for packaging, but which are incapable of producing infectious virions. The defective retroviral vectors that are used, on the other hand, lack these structural genes but encode the remaining proteins necessary for packaging. To prepare a packaging cell line, one can construct an infectious clone of a desired retrovirus in which the packaging site has been deleted. Cells comprising this construct will express all structural viral proteins, but the introduced DNA will be incapable of being packaged. Alternatively, packaging cell lines can be produced by transforming a cell line with one or more expression plasmids encoding the appropriate core and envelope proteins. In these cells, the gag, pol, and env genes can be derived from the same or different retroviruses.

[0217] A number of packaging cell lines suitable for the present invention are also available in the prior art. Examples of these cell lines include Crip, GPE86, PA317 and PG13 (see Miller et al., *J. Virol.* 65:2220-2224 (1991)). Examples of other packaging cell lines are described in Cone and Mulligan *Proceedings of the National Academy of Sciences, USA,* 81:6349-6353 (1984); Danos and Mulligan *Proceedings of the National Academy of Sciences, USA,* 85:6460-6464 (1988); Eglitis et al. (1988), supra; and Miller (1990), supra.

[0218] Packaging cell lines capable of producing retroviral vector particles with chimeric envelope proteins may be used. Alternatively, amphotropic or xenotropic envelope proteins, such as those produced by PA317 and GPX packaging cell lines may be used to package the retroviral vectors.

[0219] In some embodiments of the invention, an antisense polynucleotide is administered which hybridizes to a gene encoding a polypeptide of the invention. The antisense polypeptide can be provided as an antisense oligonucleotide (see, e.g., Murayama et al., *Antisense Nucleic Acid Drug Dev.* 7:109-114 (1997)). Genes encoding an antisense nucleic acid can also be provided; such genes can be introduced into cells by methods known to those of skill in the art. For example, one can introduce an antisense nucleotide sequence in a viral vector, such as, for example, in hepatitis B virus (see, e.g., Ji et al., *J. Viral Hepat.* 4:167-173 (1997)), in adeno-associated virus (see, e.g., Xiao et al., *Brain Res.* 756:76-83 (1997)), or in other systems including, but not limited, to an HVJ (Sendai virus)-liposome gene delivery system (see, e.g., Kaneda et al., *Ann. NY Acad. Sci.* 811:299-308 (1997)), a "peptide vector" (see, e.g., Vidal et al., *CR Acad. Sci. III* 32:279-287 (1997)), as a gene in an episomal or plasmid vector (see, e.g., Cooper et al., *Proc. Natl. Acad. Sci. U.S.A.* 94:6450-6455 (1997), Yew et al. *Hum Gene Ther.* 8:575-584 (1997)), as a gene in a peptide-DNA aggregate (see, e.g., Niidome et al., *J. Biol. Chem.* 272:15307-15312 (1997)), as "naked DNA" (see, e.g., U.S. Pat. Nos. 5,580,859 and 5,589,466), in lipidic vector systems (see, e.g., Lee et al., *Crit. Rev Ther Drug Carrier Syst.* 14:173-206 (1997)), polymer coated liposomes (U.S. Pat. Nos. 5,213,804 and 5,013,556), cationic liposomes (Epand et al., U.S. Pat. Nos. 5,283,185; 5,578,475; 5,279,833; and 5,334,761), gas filled microspheres (U.S. Pat. No. 5,542,935), ligand-targeted encapsulated macromolecules (U.S. Pat. Nos. 5,108,921; 5,521,291; 5,554,386; and 5,166,320). In another embodiment, conditional expression systems, such as those typified by the tet-regulated systems and the RU-486 system, can be used (see, e.g., Gossen & Bujard, *PNAS* 89:5547 (1992); Oligino et al., *Gene Ther.* 5:491-496 (1998); Wang et al., *Gene Ther.* 4:432-441 (1997); Neering et al., *Blood* 88:1147-1155 (1996); and Rendahl et al., *Nat. Biotechnol.* 16:757-761 (1998)). These systems impart small molecule control on the expression of the target gene(s) of interest.

C. Pharmaceutical Formulations

[0220] When used for pharmaceutical purposes, the vectors used for gene therapy are formulated in a suitable buffer, which can be any pharmaceutically acceptable buffer, such as phosphate buffered saline or sodium phosphate/sodium sulfate, Tris buffer, glycine buffer, sterile water, and other buffers known to the ordinarily skilled artisan such as those described by Good et al. *Biochemistiy* 5:467 (1966).

[0221] The compositions can additionally include a stabilizer, enhancer, or other pharmaceutically acceptable carriers or vehicles. A pharmaceutically acceptable carrier can con-

tain a physiologically acceptable compound that acts, for example, to stabilize the nucleic acids of the invention and any associated vector. A physiologically acceptable compound can include, for example, carbohydrates, such as glucose, sucrose or dextrans; antioxidants, such as ascorbic acid or glutathione; chelating agents; low molecular weight proteins or other stabilizers or excipients. Other physiologically acceptable compounds include wetting agents, emulsifying agents, dispersing agents, or preservatives, which are particularly useful for preventing the growth or action of microorganisms. Various preservatives are well known and include, for example, phenol and ascorbic acid. Examples of carriers, stabilizers, or adjuvants can be found in Remington's *Pharmaceutical Sciences*, Mack Publishing Company, Philadelphia, Pa., 17th ed. (1985).

D. Administration of Formulations

[0222] The formulations of the invention can be delivered to any tissue or organ using any delivery method known to the ordinarily skilled artisan. In some embodiments of the invention, the nucleic acids of the invention are formulated in mucosal, topical, and/or buccal formulations, particularly mucoadhesive gel and topical gel formulations. Exemplary permeation enhancing compositions, polymer matrices, and mucoadhesive gel preparations for transdermal delivery are disclosed in U.S. Pat. No. 5,346,701.

E. Methods of Treatment

[0223] The gene therapy formulations of the invention are typically administered to a cell. The cell can be provided as part of a tissue, such as an epithelial membrane, or as an isolated cell, such as in tissue culture. The cell can be provided in vivo, ex vivo, or in vitro.

[0224] The formulations can be introduced into the tissue of interest in vivo or ex vivo by a variety of methods. In some embodiments of the invention, the nucleic acids of the invention are introduced into cells by such methods as microinjection, calcium phosphate precipitation, liposome fusion, or biolistics. In further embodiments, the nucleic acids are taken up directly by the tissue of interest.

[0225] In some embodiments of the invention, the nucleic acids of the invention are administered ex vivo to cells or tissues explanted from a patient, then returned to the patient. Examples of ex vivo administration of therapeutic gene constructs include Nolta et al., *Proc Natl. Acad. Sci. USA* 93(6): 2414-9 (1996); Koc et al., *Seminars in Oncology* 23 (1):46-65 (1996); Raper et al., *Annals of Surgery* 223(2):116-26 (1996); Dalesandro et al., *J. Thorac. Cardi. Surg.,* 11(2):416-22 (1996); and Makarov et al., *Proc. Natl. Acad. Sci. USA* 93(1): 402-6 (1996).

X. Diagnosis of Mood Disorders and Psychotic Disorders

[0226] The present invention also provides methods of diagnosing mood disorders (such as major depression or bipolar disorder), psychotic disorders (such as schizophrenia) Diagnosis involves determining the level of a polypeptide or polynucleotide of the invention in a patient and then comparing the level to a baseline or range. Typically, the baseline value is representative of a polypeptide or polynucleotide of the invention in a healthy person not suffering from a mood disorder or psychotic disorder or under the effects of medication or other drugs. Variation of levels of a polypeptide or polynucleotide of the invention from the baseline range (either up or down) indicates that the patient has a mood disorder or psychotic disorder or at risk of developing at least some aspects of a mood disorder or psychotic disorder. In some embodiments, the level of a polypeptide or polynucleotide of the invention are measured by taking a blood, urine or tissue sample from a patient and measuring the amount of a polypeptide or polynucleotide of the invention in the sample using any number of detection methods, such as those discussed herein, e.g., SNPs or haplotypes associated with this genes.

[0227] In some embodiments, the level of the enzymatic product of a polypeptide or polynucleotide of the invention is measured and compared to a baseline value of a healthy person or persons. Modulated levels of the product compared to the baseline indicates that the patient has a mood disorder or psychotic disorder or is at risk of developing at least some aspects of a mood disorder or psychotic disorder. Patient samples, for example, can be blood, saliva, CSF, urine or tissue samples.

[0228] It is understood that the examples and embodiments described herein are for illustrative purposes only and that various modifications or changes in light thereof will be suggested to persons skilled in the art and are to be included within the spirit and purview of this application and scope of the appended claims.

EXAMPLES

Example 1

Identification of Genes Dysregulated in Mood Disorders

[0229] A total of twenty mood disorder brains (9 bipolar and 11 major depression disorder patients) with twenty control brains were used in this study. Each brain pair (case and control) was matched on the basis of gender, age, and postmortem interval. Three brain regions, dorsolateral prefrontal cortex (DLPFC), anterior cingulate cortex (AnCg) and the cerebellum (CB) were extracted for RNA and subjected to microarray analysis using Affymetrix oligonucleotide GeneChips™. Each RNA sample was subjected to two independent analyses. The results were analyzed using multiple statistical tools and algorithms with various stringencies. Real time PCR analysis was used to confirm differential gene expression for selected genes. The genes identified using this study are listed in Tables 1, 2, and 3. Furthermore, biochemical pathways associated with the differentially expressed genes were identified (see FIGS. **1-5**).

[0230] The two cortical regions DLPFC and AnCg had similar gene expression profiles in controls but differed significantly in MDD and BP, demonstrating distinct gene expression profiles. BP subject showed more changes in AnCg compared to DLPFC whereas MDD show less profound changes in both cortical regions but had greater effects in the DLPFC than in the AnCg. For BP, several candidate genes were located in chromosomal region 15q11-13, which is associated with the Prader-Willi syndrome (see FIGS. **6-8**).

Example 2

Identification of Additional Genes Dysregulated in Mood Disorders

[0231] The RNA from three brain regions, dorsolateral prefrontal cortex (DLPFC), anterior cingulate cortex (AnCg) and

the cerebellum (CB) from deceased patients diagnosed with bipolar disease or major depression, and matched controls were extracted and subjected to microarray analysis using Affymetrix oligonucleotide GeneChips™. The patient's particular conditions in their terminal phase (agonal factors, e.g., seizure, coma, hypoxia, dehydration, and pyrexia) and the conditions of the brain tissue after death (postmortem factors, e.g., postmortem interval, and freezer interval) are two major influences on RNA preservation in postmortem brain tissue. Brain pH has been evaluated as an indicator for agonal status, and as an indicator of RNA preservation. The effect of agonal factors and pH were taken into account for quality control of the RNA. Two RNA samples were subjected to independent analyses. The results were analyzed using multiple statistical tools and algorithms with various stringencies. The 967 genes identified using this study are listed in Table 4. Real time PCR analysis was used to confirm differential gene expression for selected genes. Real time PCR confirmation of differential gene expression for selected genes is listed in Table 5.

[0232] Furthermore, biochemical pathways associated with the differentially expressed genes were identified. In particular, cortical areas in BP patients showed activation of several pathways, including the proteasome pathway, the oxidative phosphorylation pathway, the ATP synthesis pathway, and chaperones (i.e., heat shock proteins). In addition, signaling pathways dysregulated in BP include, e.g., G-coupled protein receptors, the phosphatidylinositol pathway, the cAMP pathway, the mitogen activated protein kinase pathway, cytoskeletal systems, and the cortical GABA and glutamate systems. In MD, dysregulated genes includes genes involved in transmission of nerve impulses, neurogenesis, and the fibroblast growth factor system (FGF). (see FIGS. **10-12**). Gene ontology (i.e., genetic signatures) for BP and MD can conveniently be used in developing diagnostic and therapeutic regiments for mood disorders.

Example 3

Identification of Additional Genes Dysregulated in Mood Disorders Using Rat Models of Depression and Anti-Depressant Treatment

[0233] Rats were exposed to chronic unpredictable stress treatments in parallel with chronic anti-depressants treatment (e.g., the tricyclic antidepressant desipramine and the specific serotonin reuptake inhibitor fluoxetine). Saline treated stressed rats (SS) and saline treated non-stressed rats (SN) were used as controls. In particular, saline treated stressed rats (SS) were compared to desipramine treated stressed rats (DS); saline treated stressed rats (SS) were compared to fluoxetine treated stressed rats (FS); saline treated non-stressed rats (SN) were compared to desipramine treated non-stressed rats (DN); saline treated non-stressed rats (SN) were compared to fluoxetine treated non-stressed rats (FN); and saline treated stressed rats (SS) were compared to saline treated non-stressed rats (SN). Gene expression changes in rat cortex following treatment were measured. The genes identified in this study are shown in Table 6. This data suggests that different classes of antidepressants, i.e., antidepressants with apparently different mechanisms of action may act through a common biochemical pathway.

[0234] The above examples are provided to illustrate the invention but not to limit its scope. Other variants of the invention will be readily apparent to one of ordinary skill in the art and are encompassed by the appended claims. All publications, databases, Genbank sequences, GO terms, patents, and patent applications cited herein are hereby incorporated by reference.

TABLE 1

GenBank Accession#	DLPFC-MDD Gene Description	OMIM	Chromosome Location
NM1964	Early growth response protein 1 (EGR1)	EGR1	5q31.1
NM599	human Insulin-like growth factor binding protein 5 (IGFBP5)	IGFBP5	2q33-34
M87771	Fibroblast growth factor receptor k-sam, Splice 3 (k-sam-III)	k-sam-III	10q26
Z24725	*H sapiens* Mitogen-inducible gene (mig-2)	mig-2	14q22.1
M64347	human Novel growth factor receptor (FGFR3)	FGFR3	4p16.3
M80634	human Keratinocyte growth factor receptor (FGFR2) (SEQ ID NO: 1)	FGFR2	10q26
Z14228	Nuclear mitotic apparatus protein 1, Alt. Splice Form 2 (NuMA Clone U4)	NUMA U4	11q13
X67951	human Proliferation-associated gene (PAGA)	PAGA	1p34.1

GenBank Accession #	DLPFC-MDD Gene Description	
AF036268	SH3-domain GRB2-like 2	OMIM - SH3 DOMAIN, GRB2-LIKE, 2; SH3GL2
AF060877	regulator of G-protein signalling 20	OMIM - REGULATOR OF G PROTEIN SIGNALING 20; RGS20
AL049538	ras association (RalGDS/AF-6) domain containing protein JC265	OMIM - RAL GUANINE NUCLEOTIDE DISSOCIATION STIMULATOR; RALGDS
D14838	fibroblast growth factor 9 (glia-activating factor)	OMIM - FIBROBLAST GROWTH FACTOR 9; FGF9
D26070	inositol 1,4,5-triphosphate receptor, type 1	OMIM - INOSITOL 1,4,5-TRIPHOSPHATE RECEPTOR, TYPE 1; ITPR1
J02902	protein phosphatase 2 (formerly 2A), regulatory subunit A (PR 65), alpha isoform	OMIM - PROTEIN PHOSPHATASE 2, STRUCTURAL/REGULATORY SUBUNIT A, ALPHA; PPP2R1A
J04513	fibroblast growth factor 2 (basic)	OMIM - FIBROBLAST GROWTH FACTOR 2; FGF2
L05624	mitogen-activated protein kinase kinase 1	OMIM - MITOGEN-ACTIVATED PROTEIN KINASE KINASE 1; MAP2K1
M64788	RAP1, GTPase activating protein 1	OMIM - RAP1, GTPase-ACTIVATING PROTEIN 1; RAP1GA1

TABLE 1-continued

M87771	fibroblast growth factor receptor 2 (bacteria-expressed kinase, keratinocyte growth factor receptor, craniofacial dysostosis 1, Crouzon syndrome, Pfeiffer syndrome, Jackson-Weiss syndrome)	OMIM - FIBROBLAST GROWTH FACTOR RECEPTOR 2; FGFR2
M96995	growth factor receptor-bound protein 2	OMIM - GROWTH FACTOR RECEPTOR-BOUND PROTEIN 2; GRB2
U09759	mitogen-activated protein kinase 9	OMIM - MITOGEN-ACTIVATED PROTEIN KINASE 9; MAPK9
U24152	p21/Cdc42/Rac1-activated kinase 1 (STE 20 homolog, yeast)	OMIM - p21/CDC42/RAC1-ACTIVATED KINASE 1; PAK1
U49857	transcriptional activator of the c-fos promoter	
W28432	Cluster Incl. W28432: 47f2 *Homo sapiens* cDNA /gb = W28432 /gi = 1308443/ug = Hs.92030 /len = 921	OMIM - NEUROTROPHIC TYROSINE KINASE, RECEPTOR, TYPE 2; NTRK2
X07109	protein kinase C, beta 1	OMIM - PROTEIN KINASE C, BETA-1; PRKCB1
X54938	inositol 1,4,5-trisphosphate 3-kinase A	OMIM - INOSITOL 1,4,5-TRISPHOSPHATE 3-KINASE A; ITPKA
Z71929	fibroblast growth factor receptor 2 (bacteria-expressed kinase, keratinocyte growth factor receptor, craniofacial dysostosis 1, Crouzon syndrome, Pfeiffer syndrome, Jackson-Weiss syndrome)	OMIM - FIBROBLAST GROWTH FACTOR RECEPTOR 2; FGFR2

GenBank Accession #	Antcg BP Description	Symbol
NM_004794	RAB33A, member RAS oncogene family	RAB33A
NM_002844	protein tyrosine phosphatase, receptor type, K	PTPRK
M14752	M14752 HUMABLA Human c-abl gene |Gen Bank==M14752	ABL1
NM_005252	v-fos FBJ murine osteosarcoma viral oncogene homolog	FOS
NM_002229	jun B proto-oncogene	JUNB
NM_014813	KIAA0806 gene product	KIAA0806
AB007943	AB007943: *Homo sapiens* mRNA for KIAA0474 protein |GenBank==AB007943	RAP1GA1
NM_004067	chimerin (chimaerin) 2	CHN2
NM_003676	degenerative spermatocyte homolog, lipid desaturase (*Drosophila*)	DEGS
NM_000830	glutamate receptor, ionotropic, kainate 1	GRIK1
NM_002487	necdin homolog (mouse)	NDN
NM_002921	retinal G protein coupled receptor	RGR
NM_001390	dystrobrevin, alpha	DTNA
NM_006000	tubulin, alpha 1 (testis specific)	TUBA1
NM_001634	S-adenosylmethionine decarboxylase 1	AMD1
NM_006931	solute carrier family 2 (facilitated glucose transporter), member 3	SLC2A3
NM_003832	phosphoserine phosphatase-like	PSPHL
NM_005010	neuronal cell adhesion molecule	NRCAM
NM_002073	guanine nucleotide binding protein (G protein), alpha z polypeptide	GNAZ
L24123	L24123: *Homo sapiens* NRF1 protein (NRF1) mRNA/cds = UNKNOWN /gb = L24123 /gi = 438646 /ug = Hs.83469 /len = 4992|Gen Bank==L24123	NFE2L1
NM_000810	gamma-aminobutyric acid (GABA) A receptor, alpha 5	GABRA5
NM_005398	protein phosphatase 1, regulatory (inhibitor) subunit 3C	PPP1R3C
AI526089	AI526089: DU3.2-7.H07.r *Homo sapiens* cDNA| GenBank==AI526089	COX5B
NM_000840	glutamate receptor, metabotropic 3	GRM3
NM_012249	ras-like protein TC10	TC10
NM_004791	integrin, beta-like 1 (with EGF-like repeat domains)	ITGBL1
NM_000615	neural cell adhesion molecule 1	NCAM1
NM_003916	adaptor-related protein complex 1, sigma 2 subunit	AP1S2
NM_001406	ephrin-B3	EFNB3
NM_001718	bone morphogenetic protein 6	BMP6
X66358	X66358 cds#2 HSSTHPKB *H. sapiens* mRNA KKIALRE for serine/threonine protein kinase|GenBank==X66358 DLPC-BP	CDKL1
D00654	actin, gamma 2, smooth muscle, enteric	ACTG2
U19599	U19599 HSU19599 Human (BAX delta) mRNA| GenBank==U19599	BAX

TABLE 1-continued

NM_006908	ras-related C3 botulinum toxin substrate 1 (rho family, small GTP binding protein Rac1)	RAC1
NM_002374	microtubule-associated protein 2	MAP2
AJ001612	phosphoserine phosphatase-like	PSPHL
NM_000293	phosphorylase kinase, beta	PHKB
NM_020217	hypothetical protein DKFZp547I014	DKFZp547I014
NM_004379	cAMP responsive element binding protein 1	CREB1
NM_032041	neurocalcin delta	NCALD
NM_015716	Misshapen/NIK-related kinase	MINK
AF059274	*Homo sapiens* cDNA FLJ37320 fis, clone BRAMY2018106	CSPG5
NM_006158	neurofilament, light polypeptide 68 kDa	NEFL
NM_002730	protein kinase, cAMP-dependent, catalytic, alpha	PRKACA
NM_003885	cyclin-dependent kinase 5, regulatory sub unit 1 (p35)	CDK5R1
NM_003020	Secretory granule, neuroendocrine protein 1 (SGNE1)(7B2 protein) located at chromosome band 15q13	

TABLE 2

NM1964	Early growth response protein 1 (EGR1)
NM599	human insulin-like growth factor binding protein 5 (IGFBP5)
M87771	Fibroblast growth factor receptor k-sam, Splice 3 (k-sam-III)
Z24725	*H sapiens* Mitogen-inducible gene (mig-2)
M64347	human Novel growth factor receptor (FGFR3)
M80634	human Keratinocyte growth factor receptor (FGFR2) (SEQ ID NO: 1)
Z14228	Nuclear mitotic apparatus protein 1, Alt. Splice Form 2 (NuMA Clone U4)
X67951	human Proliferation-associated gene (PAGA)
AF036268	SH3-domain GRB2-like 2
AF060877	regulator of G-protein signalling 20
AL049538	ras association (RaIGDS/AF-6) domain containing protein JC265
D14838	fibroblast growth factor 9 (glia-activating factor)
D26070	inositol 1,4,5-triphosphate receptor, type 1
J02902	protein phosphatase 2 (formerly 2A), regulatory subunit A (PR 65), alpha isoform
J04513	fibroblast growth factor 2 (basic)
L05624	mitogen-activated protein kinase kinase 1
M64788	RAP1, GTPase activating protein 1
M87771	fibroblast growth factor receptor 2 (bacteria-expressed kinase, keratinocyte growth factor receptor, craniofacial dysostosis 1, Crouzon syndrome, Pfeiffer syndrome, Jackson-Weiss syndrome)
M96995	growth factor receptor-bound protein 2
U09759	mitogen-activated protein kinase 9
U24152	p21/Cdc42/Rac1-activated kinase 1 (STE20 homolog, yeast)
U49857	transcriptional activator of the c-fos promoter
W28432	Cluster Incl. W28432: 47f2 *Homo sapiens* cDNA /gb = W28432 /gi = 1308443 /ug = Hs.92030 /len = 921
X07109	protein kinase C, beta 1
X54938	inositol 1,4,5-trisphosphate 3-kinase A
Z71929	fibroblast growth factor receptor 2 (bacteria-expressed kinase, keratinocyte growth factor receptor, craniofacial dysostosis 1, Crouzon syndrome, Pfeiffer syndrome, Jackson-Weiss syndrome)
NM_004794	RAB33A, member RAS oncogene family
NM_002844	protein tyrosine phosphatase, receptor type, K
M14752	M14752 HUMABLA Human c-abl gene|GenBank==M14752
NM_005252	v-fos FBJ murine osteosarcoma viral oncogene homolog
NM_002229	jun B proto-oncogene
NM_014813	KIAA0806 gene product
AB007943	AB007943: *Homo sapiens* mRNA for KIAA0474 protein|GenBank==AB007943
NM_004067	chimerin (chimaerin) 2
NM_003676	degenerative spermatocyte homolog, lipid desaturase (*Drosophila*)
NM_000830	glutamate receptor, ionotropic, kainate 1
NM_002487	necdin homolog (mouse)
NM_002921	retinal G protein coupled receptor
NM_001390	dystrobrevin, alpha
NM_006000	tubulin, alpha 1 (testis specific)

TABLE 2-continued

NM_001634	S-adenosylmethionine decarboxylase 1
NM_006931	solute carrier family 2 (facilitated glucose transporter), member 3
NM_003832	phosphoserine phosphatase-like
NM_005010	neuronal cell adhesion molecule
NM_002073	guanine nucleotide binding protein (G protein), alpha z polypeptide
L24123	L24123: *Homo sapiens* NRF1 protein (NRF1) mRNA /cds = UNKNOWN /gb = L24123 /gi = 438646 /ug = Hs.83469 /len = 4992|GenBank==L24123
NM_000810	gamma-aminobutyric acid (GABA) A receptor, alpha 5
NM_005398	protein phosphatase 1, regulatory (inhibitor) subunit 3C
AI526089	AI526089: DU3.2-7.H07.r *Homo sapiens* cDNA|GenBank==AI526089
NM_000840	glutamate receptor, metabotropic 3
NM_012249	ras-like protein TC10
NM_004791	integrin, beta-like 1 (with EGF-like repeat domains)
NM_000615	neural cell adhesion molecule 1
NM_003916	adaptor-related protein complex 1, sigma 2 subunit
NM_001406	ephrin-B3
NM_001718	bone morphogenetic protein 6
X66358	X66358 cds#2 HSSTHPKB *H. sapiens* mRNA KKIALRE for serine/threonine protein kinase|GenBank==X66358
D00654	actin, gamma 2, smooth muscle, enteric
U19599	U19599 HSU19599 Human (BAX delta) mRNA|GenBank==U19599
NM_006908	ras-related C3 botulinum toxin substrate 1 (rho family, small GTP binding protein Rac1)
NM_002374	microtubule-associated protein 2
AJ001612	phosphoserine phosphatase-like
NM_000293	phosphorylase kinase, beta
NM_020217	hypothetical protein DKFZp547I014
NM_004379	cAMP responsive element binding protein 1
NM_032041	neurocalcin delta
NM_015716	Misshapen/NIK-related kinase
AF059274	*Homo sapiens* cDNA FLJ37320 fis, clone BRAMY2018106
NM_006158	neurofilament, light polypeptide 68 kDa
NM_002730	protein kinase, cAMP-dependent, catalytic, alpha
NM_003885	cyclin-dependent kinase 5, regulatory subunit 1 (p35)

TABLE 3

Disorder/Region	Description	Acc. Numb.
MD DLPFC	carboxypeptidase D	U65090 ⍰
	prostaglandin D2 synthase (21 kD, brain)	AI207842 ⍰
	NEL-like 1 (chicken)	D83017 ⍰
	zinc finger protein 36, C3H type-like 1	X79067 ⍰
	phosphoribosyl pyrophosphate synthetase 1	X15331 ⍰
MD AnCng	solute carrier family 1 (glial high affinity glutamate transporter), member 3	D26443 ⍰
	clathrin, light polypeptide (Lcb)	M20470 ⍰
	aldolase A, fructose-bisphosphate	X05236 ⍰
	ubiquitin carboxyl-terminal esterase L1 (ubiquitin thiolesterase)	X04741 ⍰
BP AnCng	v-raf-1 murine leukemia viral oncogene homolog 1	X03484 ⍰
	cytochrome c oxidase subunit Vb	AI526089 ⍰
	proteasome (prosome, macropain) 26S subunit, non-ATPase, 1	D44466 ⍰
	tyrosine 3-monooxygenase/tryptophan 5-monooxygenase activation protein, theta polypeptide	X56468 ⍰
	nuclear receptor subfamily 4, group A, member 1	L13740 ⍰
	chondroitin sulfate proteoglycan 3 (neurocan)	AF02654 ⍰
	fatty acid binding protein 7, brain	AJ00296 ⍰
BP DLPFC	carboxypeptidase D	U65090 ⍰

⍰ indicates text missing or illegible when filed

TABLE 4

Summary

Genbank Accession No.	AnCg BP	AnCg MD	DLPFC BP	DLPFC MD	CB BP	CB MD	flags
D50310		up			up		1
L08485			up				1, 2, 3
U28964					up		1
AF016917				up		up	1, 3
L19182				down			1

TABLE 4-continued

Summary

Genbank Accession No.	AnCg BP	AnCg MD	DLPFC BP	DLPFC MD	CB BP	CB MD	flags
AJ001612		up	down	up	down	up	1, 3
U66879				up			1
J04046		up					1
X63575		up		up	up		1, 3
S74445				up			1, 2, 3
X71490	up		up	up	up		1
AF112471		up		up			1, 2, 3
AB006626		up					1
U37143		down		down			1, 3
AC004131	up	down	up	down		down	1
M29273		down	up	down			1, 2
X76220	up		up				1, 2
M12267			up				3
AF060877		down		down		down	1
AB018305				down			1, 2
U58334		down		down		down	1
AB020629	up	down	up	down			1, 3
U37122		down		down			1, 3
AL080061		down		down	down	down	1, 3
M34309		down		down			1
M80634		down		down			1
(SEO ID NO: 1)							
M64347		down		down			1
X57206		down		down		down	1, 3
X77196		down		down			1, 3
Z24725		down		down		down	1
AB018342		down		down			1, 3
Y10275			down			up	1, 3
AB007943	down						1
AL049538		down		down		down	1
M14758			down	down		down	3
X13839			down	down			3
X63432			up		up		3
X04098	up		up		up		3
AF006082	up					down	3
D67031		down		down		down	3
L22214			up				3
J03473			up				3
AJ236876	up	up		up			
AF072902		up		up			
U84011				down		down	
K02215	up				up	up	
AI800578		down		down		down	
AL049954				down			
R59606		down		down			
M80899		down		down			2, 3
U00957				up	up		
AA114830			up				
U81607			up				3
M90360	down		down				
X15414	up		up				3
U05861			up				
D17793		down		down			3
K03000				down		down	2
U46689			up			down	3
U24267	down		down	down			
M93405		down		down			3
U34252	up		up				3
X05236	up	up	up		up	up	
M21154	up		up		up		3
W63793			up				3
M63175			up		up		
AB028994		down		down			
U29926				down			
X81438	up	up	up	up			3
D14662		down		down		down	3
AF091077	down						3
X97074	up	up	up	up			2
D38293	up	up	up				3
J02611				down		down	
M12529		down		down			3

TABLE 4-continued

Summary

Genbank Accession No.	AnCg BP	AnCg MD	DLPFC BP	DLPFC MD	CB BP	CB MD	flags
D86981			up			down	3
U41518				down	down		
U34846		down		down	up		2, 3
D87468		down					2
L04510			up				2, 3
AF049884		up					3
U02570				down		down	
AB002292				down			
U50523		up					3
AI525393	up						3
AF006087	up			up			2, 3
AF006088			up				3
Z11501				up			
L08424		down		down			3
M27396		up	up				3
S67156		down		down			
AL096842		down		down			
AB018258				down			
J05096		down		down		down	2
M37457					up	up	2
W28508				down	up		
AF007876		down		down			
J04027					up		2, 3
L20977				up	up		3
W28589	up			up			3
AJ010953	up						2
D14710	up		up	up	up		3
U09813		up	up				3
AF087135		up	up				3
AA845575		up	up				3
AF047436		up	up				3
AA917672			up				3
X83218		up	up				3
D16469	up	up					
AL049929		up	up				3
D89052	up		up				3
AI318615			down				3
AI547262			up				3
AA056747			up				3
L09235		up		up	up		3
AA877795		up	up				2
X76228	up	up	up	up			3
W26326				up	up	up	
X79888			up				2
X66030				down			
M76125				down			
S82297		down		down			
AB021288		down		down			
V00567		down		down		down	
AF029893	up	up	up	up			3
AF082868		down		down			3
U50708				down			
U00115				down		down	
AL049257			up	down		down	
AB004066		down		down		down	
AF001383	up		up				2, 3
U68485	up						2, 3
AF002697			up		up		3
S78771	up			up	up	up	3
AC005306			down			up	
AB023169			up		up		
U72649		down		down		down	2
AF047472	up					down	
AB023171	up	down		down			
X94910	up	up	up			up	3
AF054175		up					3
AF014837			down	down			
AF009425				down	down	down	
X95592			up				2, 3
AB007948	up	up		up			
D86062	up						3

TABLE 4-continued

Summary

Genbank Accession No.	AnCg BP	AnCg MD	DLPFC BP	DLPFC MD	CB BP	CB MD	flags
AL080097		down		down		down	
AF006621		up	up	up			3
J03037		down	up	down		down	3
U79666			down				3
M76559				up			2
S60415		up					3
AF068862	up		up	up	up		2
U12022		up		up	up		
D45887	up				up		
AB020640		up					
U02390				up	up		2
U02390				up			
X85030		down		down			
U20325			up				2
AB002376		up				down	
AL035079	up		up				
AF070648		down		down			
L10822	up			up			
AF091433				down			
AF026166	up		up				3
X74801	up		up				3
AF026292	up						3
X69398	up				up		
M38690			up	down		down	
AF023158		down					
M37712				down			
AL031282	up					down	
M35543		up	up			down	
W27541		down		down			
U59325		up		up			
AF006484		down		down			3
X66364				up			2
L04658	up	up		up			
X77743	up		up				3
X66358				up			
U22398		down		down		down	
M16965				up	up	up	
W27184				up	up	up	
U65887	down						3
U60808	down						3
AI056696			up				3
U78516	down						
AL080084	up		up				3
AA189161	up		up				
AB023203	down					down	3
U03749		up					2
Y00064		up					
U07223	up		up				2
X70297			up				
W29042			up				3
D49738			up				3
Z30644			down				3
U89916		down		down			
M59287		down		down		down	
M59287		down		down		down	
AF039704			up				
X91788	up						3
M20469	up	up		up			3
M20470	up	up		up	up		3
AB020709			up	up			2, 3
S80562				down			2, 3
D13146	up		up				
M19650	up						3
Z21488				up	up		
AB020675		up					
AB014533	up				down	down	
M92642				down		down	
M58526		down		down			
U65928	up	up	up				3
AA149486	up	up					3

TABLE 4-continued

Summary

Genbank Accession No.	AnCg BP	AnCg MD	DLPFC BP	DLPFC MD	CB BP	CB MD	flags
M22760		up					3
M19961		up					3
AI526089	up	up	up				3
T57872	up	up					3
AA152406	down						3
AA978033		up					3
AB007618		up					3
N50520		up	up				3
U65090			up			down	
X51405			up				
S74445				up			2, 3
M27691	down						2
S68271	up		up				3
D10656	up				up		3
U49857		down		down			
U49857		down		down		down	
AL038340	up		up				
AL038340	up		up				
AF039397	up		up				
AF053641	up						3
M27826			down				
U89896			down		up	up	3
D32039				down	up		
X15998				down			
X15998				down			
AF026547		down		down		down	
M33146				down			2
L22569			up		up		
X16832		down		down			3
Y07593	up		up		up		
L06797		down				down	3
L47738		up		up	up		
M33318	down						3
Y11307				down	down	down	
M98529	up	up		up	up	up	
AB002379		down		down		down	
D15057			up				3
AL050152			up			down	
D31767			up				3
AL050084				down		down	
AB002367			up		down	down	3
AF086947		up		up			
U50733	up	up	up				3
W26651	up	up	up				
U48705				down			
L20817		down		down			
U59321				down	up		
AF000982			up				
AF007142				down		down	
U63825	up	up					3
AF021819			up				3
AL080115			up			down	
AL049944	up		up				
AL049934		down		down	down	down	
AL050390		up	up				
AL050272	up	up					
AL050159		down		down			
L08069	up	up	up	up			
L08069	up	up	up				
AI810807		up	up				2, 3
AI540958			up				3
AF000430	up			up	up		3
D50857		down		down		down	3
AF007875			up				3
U97105			up				
M97388	up		up				3
D83407		up	up				
S65738			up				3
U26742			down				3
U46744		down			up		3
U84551		down			up		

TABLE 4-continued

Summary

Genbank Accession No.	AnCg BP	AnCg MD	DLPFC BP	DLPFC MD	CB BP	CB MD	flags
U46746		down					3
X68277		down		down		down	3
L05147				up			
AB013382		down		down			3
U31930			up				3
U46461			down				3
D86550			up				2, 3
M91670	up			up			
M91670	up			up			
AL050282	up	up					
M31210		down					
D13168				down			
X70940		up	up				3
AB023159	up	up	up	up	up	up	
U03877		down		down			
U66406	up	up					3
AB011542			up				
L18960	down						
AF035280			up				3
U36764	up						3
U39067	up		up	up			3
U94855		up	up				3
U54558	up						3
AC002544			down		down		
D13748	up				up		
L36055			down				3
U49436		up			up		2,3
AL080199		down		down			
C18655				down			
AB002303		down		down			
X51956		up					2
L35594		down	up	down		down	2, 3
L35594		down	up	down		down	2, 3
D45421	down						2
AF103905				down			
U81984			down				3
D83492		up		up			
U12535				down			
M34309		down		down	down		3
X81625	up		up			down	3
J04058			up				3
AB028990			up				
J02931				down			
AJ002962	up		up				3
AA977580			up				2
W26480		down		down		down	
AF035284				down		down	
X87241		down		down			
AF000561			down				
M30448	up		up	up			
AB014596			up				3
D14697	up		up				3
U60060			up				3
U60061			up				3
X59065				down	down	down	
Z70276		up		up	up		3
U66198				up			3
D14838				up			2
Z69641		down		down		down	
Z69641		down					
M87770		down		down		down	
Z71929		down		down		down	
X55741	up		up	up			
AF070557			up				
W27472		down		down			
W26655	up		up				
AF052106		up		up			
AL049949		down		down	down	down	3
X02761	down			down			
V01512		down		down		down	
V01512		down		down	down	down	

TABLE 4-continued

Summary

Genbank Accession No.	AnCg BP	AnCg MD	DLPFC BP	DLPFC MD	CB BP	CB MD	flags
K00650		down				down	
AF032885		down		down			
M84562	down						
U32519			up				3
AB014560			up				3
AI565760			up				3
AJ225028			up				3
M82919				up	up		3
X15376				up	up		3
D86181	up					down	
S68805				down			
D00723			up			down	3
Y13286	up		up				3
S40719		down		down		down	3
D87467			up				
M65188		down					3
X52947		down		down		down	3
M57609			down	down			3
U33267	up	up	up				3
X76648		up					3
AB020645		up	up				2, 3
U08997	up	down					
U08997		down					
X59834		down		down		down	3
U43083	up			up			3
D90150	up	up					
AF017656			up	up	up		
AJ238764			up				3
AL049367				down			
AB020662	down	down		down		down	
AF047438	down						3
M22632		up	up				3
AF016004		down	up	down		down	3
D38449				up			3
U87460	up	down	up	down		down	
AJ011001				down			
X71973		up					2, 3
W28944	up		up				3
M81886	up				up		3
U10301				up			3
X82068	up						3
L19058	down						3
S40369				up		up	
X77748	up		up				2, 3
D87119		down		down	down	down	3
D87119		down		down	down	down	3
X04412	up	down	up	down		down	3
M16594	down				down		
J05459				up			2, 3
U90313	up	up	up				3
M95809			up				3
X03473				up			
D64142				up			
D64142				up			
L19779				up			
M37583		up	up				3
H15872	up		up				
AA255502	down				up	up	
M25079	down		down	down			
L48215	down			down			
AF019214				down	down	down	
AF029890	up	up	up				
U31814			up				3
AL034374		down	up	down		down	
AI391567			down				3
U51004		up					3
AB014555		down					3
X58536		down		down			
M32578				down			
U23803			up				3
X12671			up				

TABLE 4-continued

Summary

Genbank Accession No.	AnCg BP	AnCg MD	DLPFC BP	DLPFC MD	CB BP	CB MD	flags
M16342			up				3
D89678			up				3
U01923			up				3
W27191			down				3
X92814			up				2
X99209	up						
AF068754			up				3
M11717		down			down	down	
L26336	up	up	up	up	up		
L26336	up	up	up	up			
L26336	up	down	up	down		down	
L26336	up			down			
L12723	up						2
X87949			up				3
X13794	up	up	up				
Y00371	up	up	up	up			
L15189	up		up				2, 3
AL021937			up				
X15183			up				3
J04988	up	up		up			
W28616			up				
M22382			up				3
AI912041			up				2, 3
X57830		up					2, 3
AI434146			down				3
AF012023			up				
X77956				down		down	
AL022726				down			
U49283	up	up	up				
AA522698	up	up	up				3
X17025			up				3
U66042		up		up			
M24594	up						3
X16302		up		up			
AB017563				down			2
L42572			up				3
U26398				up	up		
X77567				up	up	up	3
U96876			up				2
X53586				down		down	
X07979				down			2
AL021786				down			
AA477898	up				up		
X54938	up	up		up	up		3
U23850		up			up		3
AB016492	up	up					
J04111	up	up					
X51345		down		down		down	
AF070523			up				
D79994		down		down		down	
L02840	down						3
U52155		down					3
U39196				up			
Y15065		up					3
D26067			up				3
D31887			up	up			3
D14663			up				3
AL049250	down	down	down				
D87074		up				down	
D87443	up					down	3
D87445				down			
AB002347			up				
AB002361	up						
AB007903			up				3
AB007963				down		down	
AB011095				down	down		
AB014526		down		down			
AB014544		down		down	down	down	
AB018335		down		down			
AB020637			up				
AB020661				down		down	

TABLE 4-continued

Summary

Genbank Accession No.	AnCg BP	AnCg MD	DLPFC BP	DLPFC MD	CB BP	CB MD	flags
AB023152	up						
AB023209		down		down			
AB023230		down		down			3
AB028972	up	up			up		
AB028977	down						3
AB029034				down		down	
AF070621			up				3
Y08319	up						3
AB002357			up				3
AF035621	up	up		up			3
U59919			up				3
AJ001685		down		down			
J04182	up		up			down	3
U36336		down	up	down		down	3
Y11395	up		up				
AL050126						down	2
M90424			down				3
X02152	up	up					3
X13794			up		up		
AI535946	up	up	up		up		
D55696	up						3
AF087693					up		2
X76488	up		up			down	3
U41060			up				3
X61118				down			
U79297	up		up				
AL039458		down		down			2
AB011540				down			
M63959	up	up	up		up		3
M92439	up	up	up	up			
AB012293	up	up		up			3
W26633	up		up				
U03100	up		up			down	3
U03100	up					down	3
D55649	up		up				3
AA420624				down			3
U01828				up	up		
U89330				up	up		
S76756	down						3
L05624	up	up		up			
U17743	up			up	up		3
U71087				up	up		3
Z11695	up			up	up		3
X14474					up		2, 3
X66867			down				
AF072250	up					down	
D84557	up					down	
X79440		up					3
S57212				up			
AW006742	down	down	down				
AI674208		down		down	up		
AI674208		down		down	up		
AF038186			up				
W26659			up				
AB014579			up		up		
M16279				down			2
D25217				down			
AI127424		up					3
AF001359			down				
X70326		down	up	down			3
AF041080	down		down		down	down	
AI670788		up	up				
Z48051			up	down			
U64565		down	up				3
D14812			up				2, 3
AI597616		up	up				3
Y11681	up	up	up	up	up		3
AL050361			up				3
Z98946				down			
AI547258		down		down			2
AF072928	up		up			down	3

TABLE 4-continued

Summary

Genbank Accession No.	AnCg BP	AnCg MD	DLPFC BP	DLPFC MD	CB BP	CB MD	flags
M55405	down		down				
AF001548	down		down	down			2
AF013570			down	down			2, 3
AF001548	down		down	down			2
D10667			down				2, 3
D10667			down				2, 3
J02854	down		down				3
AF020267				down			3
AB029029		up				down	
U42349	up	up	up				3
AF052142	up	up	up				3
AA126505	up						3
X77548						down	2
AF044209		up		up			
U35139	up	up	up				2, 3
D87953				down		down	
AF047185		up					3
AC002400	up	up	up				
AI345944		up					3
AA203354			up				2, 3
AF047181	up		up	up			3
AA527880	up						3
AA760866			up				3
AF050640	up		up				3
AI541336		up					3
AC005329	up		up				
AF053070	up	up	up	up			3
Y16241		down		down			3
D63878		down		down		down	
D23662		up					
X05608		up	up		up		
D83017		up	up	up			3
W27762	down		down				3
X64318			up				3
Z83840	up	up		up			
AB023192	up	up	up	up			3
AF019415		down		down			
AF019415		down		down			
X17620		up					2, 3
X73066	up	up	up				2, 3
AL038662		up					2, 3
X58965		up					2, 3
M86707	up	up		up			2
AI816034		up	up				
U97669	down					down	
W28770	up	up		up			3
AF002020	up					down	3
AF002020	up	down		down		down	3
AJ132583	up		up				3
U61849	up	up	up	up	up	up	2, 3
AI198311	up						2, 3
L13740	down	down	down	down	down	down	
L13740	down	down	down	down	down	down	
AB002341			up			down	2
U55258				up	up		2
U55258				up	up		2
X99076		up					
AB011150	up	up	up	up			3
U03985		up	up				2, 3
X55740				down			
AI018523		down		down		down	
X75958		down		down	up	down	2, 3
W28432		down		down		down	
Y10148		down		down			
AL050066	up	up		up			
U48250		down		down		down	
U48250		down					
M63623			up			down	
AF061034		up		up			3
AF061034		up					3
U63717			up	up			3

TABLE 4-continued

Summary

Genbank Accession No.	AnCg BP	AnCg MD	DLPFC BP	DLPFC MD	CB BP	CB MD	flags
U62961		up					3
AB017016	up	up		up			
M80482	up					down	
AB023211		down		down	down	down	
L13385			up				2, 3
D63391	up				up		3
U24152	up	up		up	up		2, 3
U24152		up		up	up		2, 3
AF068864	up	up	up	up			
AF068864	up	up	up	up			
AF005043		up	up				3
M93650				down			3
AI521453	up		up		up		3
X73424	up		up				3
AB020631				down		down	
D13892		up	up				3
D25547	up	up	up	up	up		3
U52969		up			up		3
AA535884				down			
U40370	up						2
AB007946		up					
S41458				down			
AF056490		down		down		down	3
L42451	up						3
X98248		up		up	up		
AB002345				down	down	down	2
AF093670			up			down	3
U41816	down						3
AL096719			up				3
V00572		up	up		up		2, 3
M83088	up						2, 3
X84908			up				
U45976			up			down	3
AF010312	up	down	up	down			
AL120815	up		up	down	down	down	
Z29090	down						3
U81802	up				up		3
U49070		up				up	
W28299	up	up		up			
AL050371				up			
D30037				up			
U03090				down			
U60644		up		up		up	3
U84573		down					3
M54927		down	up	down		down	3
M22299			up			down	3
X57398	up	up	up	up	up		
D11428	up	down	up	down		down	3
AF001601		down		down			
AL050161		down		down		down	
AF017786		down		down	up		2
AF016371	up						3
AF001691		up			down		3
Y18207		down		down			
Y18207		down					
N36638		down		down		down	3
Z50749		up					3
J02902		up					
J02902		up					
M64929	up		up				
M64929			up				
M29551		up					2
X89416	up			up			
U44772			up				3
AB014512	down						3
X67951			up				2
L19185	up	up	up				
U25182	up		up				
M33336			up		up		3
M33336			up		up		3
M33336			up				

TABLE 4-continued

Summary

Genbank Accession No.	AnCg BP	AnCg MD	DLPFC BP	DLPFC MD	CB BP	CB MD	flags
X07109		up		up			3
X06318		up		up			3
Z15108		up		up			3
U29185			up				2
AB011124		up			up	up	
X15331	up			up	up		3
D87258		down		down		down	
D87258		down		down		down	
J03077	up		up				3
M85169	up		up				3
L76517	up		up			down	3
D00760			up				2, 3
D00762	up		up				3
D00761	up		up				3
D26598	up						2, 3
D26600	up		up				3
D26600	up		up				3
D29011	up		up				2
D29012	up						3
D38048	up	up	up				3
AF035309		up					
D44466	up		up				2, 3
AL031177	up		up				
AB009398	up						2, 3
D78151	up						3
U51007		up					
D50063			up				3
D38047	up	up	up	up			3
D38047	up						3
AJ001612	down	up	down	up	down	up	3
D14694	up						
M98539		down		down		down	
AI207842		down				down	3
AI207842		down		down			3
U33284				up			
M14630				down			
M57399		down		down			2, 3
M57399	down	down		down			2, 3
X54131		down					
L77886	up						
Z48541	up			up			2
D64053	up						
M93426		down		down		down	2, 3
X63578					up		2
Z48054	down						3
AL031781		down					
AL031781		down		down		down	
AI540957			up				3
AF052113	up		up				3
M28209	up		up				3
AL050268			up				3
AL050268			up				3
U59877	down	down		down			3
AI189226		down		down			
D14889	up		up				3
D14889	up		up	up			3
M28212	up				up		3
AJ133534	up		up				3
X98001			up				
D25274			up				
U41654			up				2, 3
M35416		up					
M31469	up	up	up	up	up		
M31469	up	up		up	up		
AF054183	up	up	up	up	up		3
X63465	up		up				
X63465			up				
S80343	up						3
D79990		down		down		down	
U28686	down	down		down	down	down	
U89505	up		up				

TABLE 4-continued

Summary

Genbank Accession No.	AnCg BP	AnCg MD	DLPFC BP	DLPFC MD	CB BP	CB MD	flags
U23946	up						
M11433				down			3
X00129	up	up		up			3
N92548		down		down			
AW044624	up						3
U27768				up			2, 3
U78166		up	up				3
D26129	up		up	down		down	
AF037204			up			down	3
X13973	up		up				
M63488	up			up	up		
D87735		up					2, 3
X55954	up	up					3
AI708983		up	up				3
X57958			up				3
Y00281	up		up				2, 3
AL031659	up		up				
AA977163	up	up	up				3
M13932		up					3
AI526078		up					3
D14530	up						3
S79522		up					3
X55715	up		up				3
M84711		up	up			up	2, 3
Y11651			up				3
L10333	up	up	up				2, 3
AB020693			up		up		
M84820				up			
AL049940	up		up			down	
AJ001515				down			
AB020658			up				
X91257	up	up	up	up			3
M55580		down		down			
AF051323	up	up		up			
D12676	up				up		3
M25756			up		down	down	2
AF070614			up				3
L10338	down						3
AF049498	up	up		up		up	2, 3
AB011178		down		down		down	
AB007937				down	down		
AB015345	up	up		up	up		
X97064			up				3
AJ131245			up				3
AF055006	up						3
AF054184		up	up				2, 3
U73167		down		down			
AB000220			up				
AB002438		down		down		down	
Z11793				down			
D86957		down		down		down	3
AI743134		down		down		down	
Z81326			up				3
D28423	up				up		
AL031681	up					down	
L41887			up			down	
Y00757		up	up				2
AF036268		up	up	up			3
AB007960		down		down			
U33760	up	up	up		up		
W26700		up		up	up		
U08989				up			3
U01824		down		down			3
W28850		down					3
D26443		down		down		down	3
H10201				down			
X60036			up				3
M20681	up	up					3
AF007216		down		down		down	2, 3
AF011390		down			up		2, 3
AF015926		down		down			3

TABLE 4-continued

Summary

Genbank Accession No.	AnCg BP	AnCg MD	DLPFC BP	DLPFC MD	CB BP	CB MD	flags
D86959			up			down	
U96094	up				up	up	3
D80000			up			down	2, 3
X59960	up		up				3
AF053136		up					2
AL049650	up	up		up			
AA733050	up	up					
U40571				down			3
AF034546			up				3
X02317			up				3
X63753			up			down	3
AJ001183				down			2
Z46629		down		down		down	
AB011088	down			down			
J03040				down		down	
J04765	up		up				
AF052124	up		up			down	
AF039843		down		down		down	
Y08685			up			down	3
D78130			up				3
M32313				up			2
M32886		down		down			3
U88666			up				3
J00306	up						2
AI636761	up						2, 3
AB011107		down		down		down	
L78440		up		up			
U04735	up		up	up	up		
M86752				up	up		
X99325	up	up		up	up		2, 3
AF099989			up			down	3
M31303			up				2
X85116		down		down	down	down	
AL035306			up				3
U77942						down	2
D63506				down		down	
U34804		up					
U40215	up	up		up	up		3
AF039945		down					2
U93305		up		up			
X68194		down	up	down		down	3
D38522			up				
U18062	up		up				3
M95787	down		down				
AF010400	up		up				
AL050265			up				3
AL050107				down		down	
D50495	up						2
M80627		down		down		down	
D15050		down		down		down	
U19969	down					down	
X52882		up					3
U49188		up	up				
L24804			up				
X75861			up				3
W28869	up				up		3
L06139		down		down		down	
X93512				up	up		
S95936	up		up	down		down	3
M55153			down				3
L12350				down			
L12350		down		down			
AJ133115		down		down			
M24748	up			up			
X97544			up				3
X97544			up				3
L27476				down		down	
AB028950				down			
AI688299		down	up	down		down	3
R16035	up						3
U81006			up				3

TABLE 4-continued

Summary

Genbank Accession No.	AnCg BP	AnCg MD	DLPFC BP	DLPFC MD	CB BP	CB MD	flags
M92383		up					
D38305				down		down	
D13641		up					3
U09477	up						3
M12125			down				3
M12125			down	down			3
U12595	up						3
X00437	down		down				
AB011089			up				3
AF084260			up				3
AJ133769				up	up		
X89066	up						3
AF042181				up	down	up	
AF001294			up				
AF035283		down		down			
X06956	up	up		up	up		
X06956	up	up	up				2
AF005392	up	up	up				
X01703	up		up				
AF035316			up	down		down	3
X02344	up					down	
U47634	up						3
X00734	up				up		
S75463	up						3
D17517			up			down	
U18934			up				
AI310002			up				2
AF075599		up		up	up	up	
U67122			up				3
X04741	up	up	up				3
U27460		up	up				
U30930		down	up	down			3
T79616		up					3
J04973		up	up				2, 3
L32977			up				
AA526497		up					3
U30888				up			
M36200				up	up		
AL050223			up				3
U56833			up				3
L06132		up					3
AJ002428	up	up		up	up		
L08666			up				3
AF024710		down		down			
AF022375		down		down			
M63978		down			up		
X51521				down			
Z19554		down		down		down	3
AF060902		up		up			
D26068	down						2
AB011113		up				down	
W27944		down		down			
W26496		down		down		down	
D14661			up				
Y08614			up				3
J04977	up		up	up			3
M30938	up				up		
U89436	up	up					3
X56468			up				3
X56468	up		up				3
M92843		down		down		down	2, 3
U07802				down		down	
X78992		down		down		down	
AL050276		down		down			
L11672	down						
AD000092	up	up					
V00599			up			down	
X55989			down				
S81916	up	up	up		up		
J00153	down		down	down			
AL049423		down		down			

TABLE 4-continued

Summary

Genbank Accession No.	AnCg BP	AnCg MD	DLPFC BP	DLPFC MD	CB BP	CB MD	flags
AF052148	down		down				
AL022101	up		up		up		
AL118582				down		down	
AI095508					up		2
W28612		down		down		down	
AL049378	down		down				
AI700633		down	up	down			
AF070536	down		down				
AF052119		up	up	up			
AL080113			up				
AL049265		down					
AL049390				down		down	
AF070577		up	up				2
AI827895				down		down	
X95677				up	up		
AL080093			up				
AL049969		down		down		down	
AF052141		up	up	up			
L27560				down			
W27522			up				
AL022718	up					down	
AJ005694			down				3
AL120687				up	up		
AL046322	up			up			
AW043812			down				
H12054				up			
AC003007		up		up			
J03071	down			down			
M57417	down				down	down	
M33764	up						
M58028	up						
X74262	up						
U19796	up						
U22028	down						
X79568			down				
M55914	up						
M21154	up						
M10905				down			
U33429				up			
AB014539				down			
X63432			up		up		
X56841				down			
Y00067			up				
AF007140	up						
X13839			down	down			
AF023268			up				
AF053356	down						
U37122		down		down			
AB000450	up						
AI126004			up				
AF002668	up						
X54304				down			
U57843	down						
X02344	up						
X04098	up		up			up	
U96074	up						
D32053	up		up				
U59632			down				
X14346	down						
Z98046	up						
AL096737				down			
AB014598				down			
U17886			up				
AI986201			up				
AL080181				down			
AB014514	up						

TABLE 4-continued

	Summary						
Genbank Accession No.	AnCg BP	AnCg MD	DLPFC BP	DLPFC MD	CB BP	CB MD	flags
R92331				down			
U24183			up				
D00860	up						
U09510			up				
AI635895			up				
U66033				down			
U51334				down			
AF020762			up				
U24105				up			
M36820				down			
U59912				up			
X63368	up						
AF047863				up			
U11861	up						
AL080122			up				
M14648				down			
Y14153				up			
X81637	down						
M88108			up				
AF042384	up						
AA704137				up			
AB011156	up						
AI862521	up						
AF047469				up			
AF025887			up				
AF091085	up						
AL035494			up				
AI540925	down						
D32129				down			
AB028972	up						
AF091071				up			
AL040137				down			
X15187	up						
U48730	down						
L08488	up						
K03460	up						
AF005361				up			
M95585				up			
M91670	up						

TABLE 5

	RT-PCR Confirmation				
Gene Bank Acc. No.	Gene Name	AnCg BP	AnCg MD	DLPFC BP	DLPFC MD
AB020629	ABCA8				down
U37122	ADD3				down
X63575	ATP2B2			down	
X71490	ATP6V0D1		up		
U66879	BAD	down			
J04046	CALM3			up	
AF112471	CAMK2B		up		
D50310	CCNI	up			
AL080061	CLIC4				down
S74445	CRABP1		up		up
U37143	CYP2J2		down		down
M34309	ERBB3				down
M80634	FGFR2				down
(SEQ ID NO: 1)					
M64347	FGFR3				down
L08485	GABRA5	up			
AF016917	GABRD	down		down	
AC004131	GPRC5B		down		down

TABLE 5-continued

	RT-PCR Confirmation				
Gene Bank Acc. No.	Gene Name	AnCg BP	AnCg MD	DLPFC BP	DLPFC MD
AB006626	HDAC4		up		
L19182	IGFBP7	down		down	
X57206	ITPKB				down
X77196	LAMP2				down
M29273	MAG		down		down
X76220	MAL		down		down
Z24725	MIG2				down
AB018342	MYO10				down
M12267	OAT		down		down
Y10275	PSPH				down
AJ001612	PSPHL	down		down	
AB007943	RAP1GA1				down
AF060877	RGS20		down		down
AL049538	RIN2				down
AB018305	SPON1		down		down
U58334	TP53BP2		down		down
U28964	YWHAZ	up			

TABLE 6

Summary of Anti-Depressant Treatment Data

Genbank Accession No.	Gene Name	SSvDS	SSvFS	SNvDN	SNvFN	SNvSS
M80899	AHNAK			down		down
K03000	ALDH1A1	down			down	
X97074	AP2S1	down				
U34846	AQP4	down		down	up	
D87468	ARC	up		down	down	down
L04510	ARFD1	down				
AF006087	ARPC4					down
J05096	ATP1A2	down				
M37457	ATP1A3	down		down		up
J04027	ATP2B1	up				
AJ010953	ATP2C1	down				
AA877795	ATP6V1D	down				
X79888	AUH	down				
AF001383	BIN1	down				
U68485	BIN1	down				
U72649	BTG2	up				
X95592	C1D	down				
M76559	CACNA2D1	up				
AF068862	CALB1	down		down		
AF112471	CAMK2B	down				
U02390	CAP2	down		down	up	
U20325	CART				up	
X66364	CDK5	down				up
U03749	CHGA					down
U07223	CHN2				down	
AB020709	CNK2	down				
S80562	CNN3	down				
S74445	CRABP1	up				
S74445	CRABP1	up				
M27691	CREB1					up
M33146	CSRP1	down				
AI810807	DNCI1	down				
D86550	DYRK1A	down				
U49436	EIF5	down				
X51956	ENO2			up		up
L35594	ENPP2					down
L35594	ENPP2					down
D45421	ENPP2					down
AA977580	FACL3	down				
D14838	FGF9			up		up
L08485	GABRA5	down				
AB020645	GLS	down	up			down
X71973	GPX4			up		
X77748	GRM3	down		down		
J05459	GSTM3	down				
X92814	HRASLS3	down				
L12723	HSPA4	down	down			up
L15189	HSPA9B	up				
AI912041	HSPE1	down				
X57830	HTR2A	down				down
AB017563	IGSF4	up				
U96876	INSIG1	down				
X07979	ITGB1	down				
AL050126	LAP1B	down		down		
AF087693	LIN7A	down		down		
AL039458	LRIG1					up
M29273	MAG			up		
X76220	MAL	down			down	
X14474	MAPT	up				
M16279	MIC2	down				down
D14812	MRGX	down				
AI547258	MT2A	down			down	down
AF013570	MYH11	down		down	down	down
D10667	MYH11	down		down	down	down
D10667	MYH11	down		down	down	down
AF001548	MYH11	down		down	down	down
AF001548	MYH11	down		down	down	down
X77548	NCOA4	up		up		
U35139	NDN					down
AA203354	NDUFB3	down				
X73066	NME1	down				
X17620	NME1	down				

TABLE 6-continued

Summary of Anti-Depressant Treatment Data

Genbank Accession No.	Gene Name	SSvDS	SSvFS	SNvDN	SNvFN	SNvSS
AL038662	NME1	down				
X58965	NME2	down				
M86707	NMT1	down				
U61849	NPTX1	down		down		
AI198311	NPY					down
U55258	NRCAM	down				
AB002341	NRCAM	down				
U55258	NRCAM	down				
U03985	NSF			down		
X75958	NTRK2			down		
L13385	PAFAH1B1	down				
U24152	PAK1	down				
U24152	PAK1	down				
U40370	PDE1A	up				
AB002345	PER2		down	down	down	
V00572	PGK1	down				
M83088	PGM1				up	
AF017786	PPAP2B	down				
M29551	PPP3CB	down		down		
X67951	PRDX1	down				
U29185	PRNP					up
D00760	PSMA2	down				
D26598	PSMB3	down				
D29011	PSMB5		up			
D44466	PSMD1			up		
AB009398	PSMD13	down				
M57399	PTN	down				
M57399	PTN	down				
Z48541	PTPRO		up			
M93426	PTPRZ1	down				
X63578	PVALB	down				up
U41654	RAGA	down				
U27768	RGS4	down		down		
D87735	RPL14	down		down		
Y00281	RPN1	down			up	
M84711	RPS3A	down				down
L10333	RTN1	down	up			
M25756	SCG2	down			up	
AF049498	SCN2B			down		
AF054184	SEC61G			down		
Y00757	SGNE1	down				
AF007216	SLC4A4	down		down		
AF011390	SLC4A4	down		down		
D80000	SMC1L1			down		
AF053136	SNCB			up		
AJ001183	SOX10					up
AB018305	SPON1	down				
M32313	SRD5A1	up				
AI636761	SST	down	up			down
J00306	SST	down	up			down
X99325	STK25			down		
M31303	STMN1	down				
U77942	STX7	down	down			up
AF039945	SYNJ2	down				
D50495	TCEA2	up		up		
X06956	TUBA1					down
AI310002	UBE2D2	down				
J04973	UQCRC2	down				
D26068	WBSCR1	down				
M92843	ZFP36					up
AI095508		down		down		
AF070577		down				

TABLE 7a

AnCg BP Genetic Ontology

Genbank Accession No.	Gene Name	Description
		26S proteasome
D00762	PSMA3	proteasome (prosome, macropain) subunit, alpha type, 3
D44466	PSMD1	proteasome (prosome, macropain) 26S subunit, non-ATPase, 1
D00761	PSMB1	proteasome (prosome, macropain) subunit, beta type, 1
D38048	PSMB7	proteasome (prosome, macropain) subunit, beta type, 7
D78151	PSMD2	proteasome (prosome, macropain) 26S subunit, non-ATPase, 2
D29012	PSMB6	proteasome (prosome, macropain) subunit, beta type, 6
D38047	PSMD8	proteasome (prosome, macropain) 26S subunit, non-ATPase, 8
D26598	PSMB3	proteasome (prosome, macropain) subunit, beta type, 3
D26600	PSMB4	proteasome (prosome, macropain) subunit, beta type, 4
		synaptic transmission
X82068	GRIA3	glutamate receptor, ionotrophic, AMPA 3
D11428	PMP22	peripheral myelin protein 22
AI636761	SST	somatostatin
AA126505	NCAM1	neural cell adhesion molecule 1
L10338	SCN1B	sodium channel, voltage-gated, type I, beta polypeptide
X81438	AMPH	amphiphysin (Stiff-Man syndrome with breast cancer 128 kDa autoantigen)
M19650	CNP	2',3'-cyclic nucleotide 3' phosphodiesterase
L19058	GRIK1	glutamate receptor, ionotropic, kainate 1
AI198311	NPY	neuropeptide Y
U68485	BIN1	bridging integrator 1
M81886	GRIA1	glutamate receptor, ionotropic, AMPA 1
Z11695	MAPK1	mitogen-activated protein kinase 1
X77748	GRM3	glutamate receptor, metabotropic 3
AF052113	RAB14	RAB14, member RAS oncogene family
U40215	SYN2	synapsin II
U61849	NPTX1	neuronal pentraxin I
		Chaperone
J04988	HSPCB	heat shock 90 kDa protein 1, beta
U12595	TRAP1	heat shock protein 75
L12723	HSPA4	heat shock 70 kDa protein 4
L08069	DNAJA1	DnaJ (Hsp40) homolog, subfamily A, member 1
AL038340	CRYAB	crystallin, alpha B
AL038340	CRYAB	crystallin, alpha B
X02344	TUBB2	tubulin, beta, 2
AF026166	CCT2	chaperonin containing TCP1, subunit 2 (beta)
M63959	LRPAP1	low density lipoprotein receptor-related protein associated protein 1
L26336	HSPA2	heat shock 70 kDa protein 2
AF026292	CCT7	chaperonin containing TCP1, subunit 7 (eta)
L08069	DNAJA1	DnaJ (Hsp40) homolog, subfamily A, member 1
AA149486	COX17	COX17 homolog, cytochrome c oxidase assembly protein (yeast)
Y00371	HSPA8	heat shock 70 kDa protein 8
X74801	CCT3	chaperonin containing TCP1, subunit 3 (gamma)
X56468	YWHAQ	tyrosine 3-monooxygenase/tryptophan 5-monooxygenase activation protein, theta polypeptide
U41816	PFDN4	prefoldin 4
L15189	HSPA9B	heat shock 70 kDa protein 9B (mortalin-2)
L26336	HSPA2	heat shock 70 kDa protein 2

TABLE 7b

AnCg MD: Genetic Ontology

Genbank Accession No.	Gene Name	Description
		transporter activity
T79616	UQCRB	ubiquinol-cytochrome c reductase binding protein
AI526089	COX5B	cytochrome c oxidase subunit Vb
AL049929	ATP6IP2	ATPase, H+ transporting, lysosomal interacting protein 2
AF006621	C4orf1	chromosome 4 open reading frame 1
L09235	ATP6V1A1	ATPase, H+ transporting, lysosomal 70 kDa, VI subunit A, isoform 1
M22760	COX5A	cytochrome c oxidase subunit Va
U01824	SLC1A2	solute carrier family 1 (glial high affinity glutamate transporter), member 2
N50520	COX7B	cytochrome c oxidase subunit VIIb

TABLE 7b-continued

AnCg MD: Genetic Ontology

Genbank Accession No.	Gene Name	Description
		transporter activity
AF007216	SLC4A4	solute carrier family 4, sodium bicarbonate cotransporter, member 4
AF011390	SLC4A4	solute carrier family 4, sodium bicarbonate cotransporter, member 4
AA526497	UQCRH	ubiquinol-cytochrome c reductase hinge protein
AA845575	ATP5J	ATP synthase, H+ transporting, mitochondrial F0 complex, subunit F6
X52947	GJA1	gap junction protein, alpha 1, 43 kDa (connexin 43)
D26443	SLC1A3	solute carrier family 1 (glial high affinity glutamate transporter), member 3
AF053070	NDUFV1	NADH dehydrogenase (ubiquinone) flavoprotein 1, 51 kDa
X76228	ATP6V1E1	ATPase, H+ transporting, lysosomal 31 kDa, VI subunit E isoform I
X63575	ATP2B2	ATPase, Ca++ transporting, plasma membrane 2

TABLE 7c

DLPFC BP Genetic Ontology

Genbank Accession No.	Gene Name	Description
		hydrogen ion transporter activity
AA917672	ATP5L	ATP synthase, H+ transporting, mitochondrial F0 complex, subunit g
AI526089	COX5B	cytochrome c oxidase subunit Vb
J04973	UQCRC2	ubiquinol-cytochrome c reductase core protein II
AF050640	NDUFS2	NADH dehydrogenase (ubiquinone) Fe—S protein 2, 49 kDa (NADH-coenzyme Q reductase)
AL049929	ATP6IP2	ATPase, H+ transporting, lysosomal interacting protein 2
AF047181	NDUFB5	NADH dehydrogenase (ubiquinone) 1 beta subcomplex, 5, 16 kDa
AF047436	ATP5J2	ATP synthase, H+ transporting, mitochondrial F0 complex, subunit f, isoform 2
D14710	ATP5A1	ATP synthase, H+ transporting, mitochondrial F1 complex, alpha subunit, isoform 1
U09813	ATP5G3	ATP synthase, H+ transporting, mitochondrial F0 complex, subunit c (subunit 9) isoform 3
N50520	COX7B	cytochrome c oxidase subunit VIIb
AF087135	ATP5H	ATP synthase, H+ transporting, mitochondrial F0 complex, subunit d
D89052	ATP6V0B	ATPase, H+ transporting, lysosomal 21 kDa, V0 subunit c''
X76228	ATP6V1E1	ATPase, H+ transporting, lysosomal 31 kDa, V1 subunit E isoform 1
		Chaperone
U56833	VBP1	von Hippel-Lindau binding protein 1
L08069	DNAJA1	DnaJ (Hsp40) homolog, subfamily A, member 1
AL038340	CRYAB	crystallin, alpha B
AL038340	CRYAB	crystallin, alpha B
X15183	HSPCA	heat shock 90 kDa protein 1, alpha
X56468	YWHAQ	tyrosine 3-monooxygenase/tryptophan 5-monooxygenase activation protein, theta polypeptide
L24804	TEBP	unactive progesterone receptor, 23 kD
W28616	HSPCB	heat shock 90 kDa protein 1, beta
D49738	CKAP1	cytoskeleton-associated protein 1
AF026166	CCT2	chaperonin containing TCP1, subunit 2 (beta)
M63959	LRPAP1	low density lipoprotein receptor-related protein associated protein 1
X87949	HSPA5	heat shock 70 kDa protein 5 (glucose-regulated protein, 78 kDa)
L26336	HSPA2	heat shock 70 kDa protein 2
M22382	HSPD1	heat shock 60 kDa protein 1 (chaperonin)
L08069	DNAJA1	DnaJ (Hsp40) homolog, subfamily A, member 1
AF035316	TUBB	tubulin, beta polypeptide
AI912041	HSPE1	heat shock 10 kDa protein 1 (chaperonin 10)
Y00371	HSPA8	heat shock 70 kDa protein 8
X74801	CCT3	chaperonin containing TCP1, subunit 3 (gamma)
X56468	YWHAQ	tyrosine 3-monooxygenase/tryptophan 5-monooxygenase activation protein, theta polypeptide
W29042	CIA30	CGI-65 protein
L15189	HSPA9B	heat shock 70 kDa protein 9B (mortalin-2)
		OXPHOS
X71490	ATP6V0D1	ATPase, H+ transporting, lysosomal 38 kDa, V0 subunit d isoform 1
D14710	ATP5A1	ATP synthase, H+ transporting, mitochondrial F1 complex, alpha subunit, isoform 1, cardiac muscle
U09813	ATP5G3	ATP synthase, H+ transporting, mitochondrial F0 complex, subunit c (subunit 9) isoform 3
AF087135	ATP5H	ATP synthase, H+ transporting, mitochondrial F0 complex, subunit d
AA845575	ATP5J	ATP synthase, H+ transporting, mitochondrial F0 complex, subunit F6
AF047436	ATP5J2	ATP synthase, H+ transporting, mitochondrial F0 complex, subunit f, isoform 2
AA917672	ATP5L	ATP synthase, H+ transporting, mitochondrial F0 complex, subunit g
X83218	ATP5O	ATP synthase, H+ transporting, mitochondrial F1 complex, O subunit
D89052	ATP6V0B	ATPase, H+ transporting, lysosomal 21 kDa, V0 subunit c''

TABLE 7c-continued

DLPFC BP Genetic Ontology

Genbank Accession No.	Gene Name	Description
AA056747	ATP6V1A1	ATPase, H+ transporting, lysosomal 70 kDa, V1 subunit A
X76228	ATP6V1E1	ATPase, H+ transporting, lysosomal 31 kDa, V1 subunit E isoform 1
AI526089	COX5B	cytochrome c oxidase subunit Vb
N50520	COX7B	cytochrome c oxidase subunit VIIb
AC002400	NDUFAB1	NADH dehydrogenase (ubiquinone) 1, alpha/beta subcomplex, 1, 8 kDa
AA203354	NDUFB3	NADH dehydrogenase (ubiquinone) 1 beta subcomplex, 3, 12 kDa
AF047181	NDUFB5	NADH dehydrogenase (ubiquinone) 1 beta subcomplex, 5, 16 kDa
AF050640	NDUFS2	NADH dehydrogenase (ubiquinone) Fe—S protein 2, 49 kDa (NADH-coenzyme Q reductase)
J04973	UQCRC2	ubiquinol-cytochrome c reductase core protein II
L32977	UQCRFS1	ubiquinol-cytochrome c reductase, Rieske iron-sulfur polypeptide 1
U17886	SDHB	succinate dehydrogenase complex, subunit B, iron sulfur (Ip)

TABLE 7d

DLPFC MD Genetic Ontology

Genbank Accession No.	Gene Name	Description
		transmission of nerve impulse
D11428	PMP22	peripheral myelin protein 22
AF049498	SCN2B	sodium channel, voltage-gated, type II, beta polypeptide
M82919	GABRB3	gamma-aminobutyric acid (GABA) A receptor, beta 3
X59834	GLUL	glutamate-ammonia ligase (glutamine synthase)
X81438	AMPH	amphiphysin (Stiff-Man syndrome with breast cancer 128 kDa autoantigen)
M54927	PLP1	proteolipid protein 1 (Pelizaeus-Merzbacher disease, spastic paraplegia 2, uncomplicated)
Z11695	MAPK1	mitogen-activated protein kinase 1
U01824	SLC1A2	solute carrier family 1 (glial high affinity glutamate transporter), member 2
M32886	SRI	sorcin
U40215	SYN2	synapsin II
X15376	GABRG2	gamma-aminobutyric acid (GABA) A receptor, gamma 2
D26443	SLC1A3	solute carrier family 1 (glial high affinity glutamate transporter), member 3
X68194	SYPL	synaptophysin-like protein
U61849	NPTX1	neuronal pentraxin I
		neurogenesis
D11428	PMP22	peripheral myelin protein 22
U30930	UGT8	UDP glycosyltransferase 8 (UDP-galactose ceramide galactosyltransferase)
W28770	NP25	neuronal protein
M54927	PLP1	proteolipid protein 1 (Pelizaeus-Merzbacher disease, spastic paraplegia 2, uncomplicated)
D83017	NELL1	NEL-like 1 (chicken)
U34846	AQP4	aquaporin 4
Z70276	FGF12	fibroblast growth factor 12
M80899	AHNAK	AHNAK nucleoprotein (desmoyokin)
M57399	PTN	pleiotrophin (heparin binding growth factor 8, neurite growth-promoting factor 1)
AF016004	GPM6B	glycoprotein M6B
X70326	MLP	MARCKS-like protein
AF036268	SH3GL2	SH3-domain GRB2-like 2
M93426	PTPRZ1	protein tyrosine phosphatase, receptor-type, Z polypeptide 1
U61849	NPTX1	neuronal pentraxin I
M93650	PAX6	paired box gene 6 (aniridia, keratitis)
		phosphoric ester hydralase activity
X55740	NT5E	5'-nucleotidase, ecto (CD73)
AF001601	PON2	paraoxonase 2
X68277	DUSP1	dual specificity phosphatase 1
L35594	ENPP2	ectonucleotide pyrophosphatase/phosphodiesterase 2 (autotaxin)
AB013382	DUSP6	dual specificity phosphatase 6
L05147	DUSP3	dual specificity phosphatase 3 (vaccinia virus phosphatase VH1-related)
Z48541	PTPRO	protein tyrosine phosphatase, receptor type, O
N36638	PPP1R3C	protein phosphatase 1, regulatory (inhibitor) subunit 3C
AJ001612	PSPHL	phosphoserine phosphatase-like

TABLE 7d-continued

DLPFC MD Genetic Ontology

Genbank Accession No.	Gene Name	Description
AF017786	PPAP2B	phosphatidic acid phosphatase type 2B
U60644	PLD3	phospholipase D3
AF056490	PDE8A	phosphodiesterase 8A
M93426	PTPRZ1	protein tyrosine phosphatase, receptor-type, Z polypeptide 1

TABLE 8

Selected Potential Druggable Targets

Genbank Accession No.	Gene Name	Target category	Description
AB020629	ABCA8	transporter	ATP-binding cassette, sub-family A (ABC1), member 8
X63575	ATP2B2	transporter	ATPase, Ca++ transporting, plasma membrane 2
X71490	ATP6V0D1	transporter	ATPase, H+ transporting, lysosomal 38 kDa, V0 subunit d isoform 1
S74445	CRABP1	transporter	cellular retinoic acid binding protein 1
M34309	ERBB3	tyrosine kinase receptor	v-erb-b2 erythroblastic leukemia viral oncogene homolog 3 (avian)
M80634 (SEQ ID NO: 1)	FGFR2	tyrosine kinase receptor	fibroblast growth factor receptor 2 (bacteria-expressed kinase, keratinocyte growth factor receptor, craniofacial dysostosis 1, Crouzon syndrome, Pfeiffer syndrome, Jackson-Weiss syndrome)
M64347	FGFR3	tyrosine kinase receptor	fibroblast growth factor receptor 3 (achondroplasia, thanatophoric dwarfism)
AC004131	GPRC5B	GPCR	G protein-coupled receptor, family C, group 5, member B
X77196	LAMP2	ligand for cell adhesion molecule	lysosomal-associated membrane protein 2
U37122	ADD3	regulator of kinase	adducin 3 (gamma)
U66879	BAD	regulator of protease	BCL2-antagonist of cell death
AB007943	RAP1GA1	regulator of kinase	RAP1, GTPase activating protein 1
AF060877	RGS20	regulator of GTPase	regulator of G-protein signalling 20
AL049538	RIN2	regulator of GTPase	Ras and Rab interactor 2
U58334	TP53BP2	reuglator of protein degradation	tumor protein p53 binding protein, 2
U28964	YWHAZ	regulator of enzyme	tyrosine 3-monooxygenase/tryptophan 5-monooxygenase activation protein, zeta polypeptide
J04046	CALM3	kinase	calmodulin 3 (phosphorylase kinase, delta)
AF112471	CAMK2B	kinase	calcium/calmodulin-dependent protein kinase (CaM kinase) II beta
D50310	CCNI	regulator of kinase	cyclin I
U37143	CYP2J2	monooxygenase	cytochrome P450, family 2, subfamily J, polypeptide 2
AB006626	HDAC4	enzyme	histone deacetylase 4
X57206	ITPKB	kinase	inositol 1,4,5-trisphosphate 3-kinase B
M12267	OAT	enzyme	ornithine aminotransferase (gyrate atrophy)
Y10275	PSPH	enzyme	phosphoserine phosphatase
AJ001612	PSPHL	enzyme	phosphoserine phosphatase-like
AL080061	CLIC4	channel	chloride intracellular channel 4
L08485	GABRAS	channel	gamma-aminobutyric acid (GABA) A receptor, alpha 5
AF016917	GABRD	channel	gamma-aminobutyric acid (GABA) A receptor, delta
L19182	IGFBP7	regulator of receptor ligand	insulin-like growth factor binding protein 7
M29273	MAG	myelination	myelin associated glycoprotein
X76220	MAL	myelination	mal, T-cell differentiation protein
Z24725	MIG2	signal transduction	mitogen inducible 2

TABLE 8-continued

Selected Potential Druggable Targets

Genbank Accession No.	Gene Name	Target category	Description
AB018342	MYO10	partner for calmodulin-like protein	myosin X
AB018305	SPON1	axon growth guidance	spondin 1, (f-spondin) extracellular matrix protein

SEQUENCE LISTING

<160> NUMBER OF SEQ ID NOS: 1

<210> SEQ ID NO 1
<211> LENGTH: 3106
<212> TYPE: DNA
<213> ORGANISM: Homo sapiens
<220> FEATURE:
<223> OTHER INFORMATION: human fibroblast growth factor receptor 2 (FGFR2) (keratinocyte growth factor receptor) cDNA
<220> FEATURE:
<221> NAME/KEY: CDS
<222> LOCATION: (419)..(2887)
<223> OTHER INFORMATION: human FGFR2

<400> SEQUENCE: 1

cccgcgagca aagtttggtg gaggcaacgc aagcctgagt cctttcttcc tctcgttccc 60

caaatccgag ggcagcccgc gggcgtcatg gcgctcctcc gcagcctggg gtacgcgtga 120

agcccgggag gcttggcgcc ggcgaagacc caaggaccac tcttctgcgt ttggagttgc 180

tccccgcaac cccgggctcg tcgctttctc catcccgacc cacgcggggc cggggacaac 240

acaggtcgcg gaggagcgtt gccattcaag tgactgcagc agcagcgcag cgcctcggtt 300

cctgagccca ccgcagctga aggcattgcg cgtagtccat gcccgtagag gaagtgtgca 360

gatgggatta acgtccacat ggagatatgg aagaggaccg gggattggta ccgtaaccat 420

ggtcagctgg ggtcgtttca tctgcctggt cgtggtcacc atggcaacct tgtccctggc 480

ccggccctcc ttcagtttag ttgaggatac cacattagag ccagaagagc caccaaccaa 540

ataccaaatc tctcaaccag aagtgtacgt ggctgcgcca ggggagtcgc tagaggtgcg 600

ctgcctgttg aaagatgccg ccgtgatcag ttggactaag gatggggtgc acttggggcc 660

caacaatagg acagtgctta ttggggagta cttgcagata aagggcgcca cacctagaga 720

ctccggcctc tatgcttgta ctgccagtag gactgtagac agtgaaactt ggtacttcat 780

ggtgaatgtc acagatgcca tctcatccgg agatgatgag gatgacaccg atggtgcgga 840

agattttgtc agtgagaaca gtaacaacaa gagagcacca tactggacca acacagaaaa 900

gatggaaaag cggctccatg ctgtgcctgc ggccaacact gtcaagtttc gctgcccagc 960

cgggggggaac ccaatgccaa ccatgcggtg gctgaaaaac gggaaggagt ttaagcagga 1020

gcatcgcatt ggaggctaca aggtacgaaa ccagcactgg agcctcatta tggaaagtgt 1080

-continued

ggtcccatct gacaagggaa attatacctg tgtagtggag aatgaatacg ggtccatcaa 1140

tcacacgtac cacctggatg ttgtggagcg atcgcctcac cggcccatcc tccaagccgg 1200

actgccggca aatgcctcca cagtggtcgg aggagacgta gagtttgtct gcaaggttta 1260

cagtgatgcc cagccccaca tccagtggat caagcacgtg gaaaagaacg gcagtaaata 1320

cgggcccgac gggctgccct acctcaaggt tctcaagcac tcggggataa atagttccaa 1380

tgcagaagtg ctggctctgt tcaatgtgac cgaggcggat gctggggaat atatatgtaa 1440

ggtctccaat tatatagggc aggccaacca gtctgcctgg ctcactgtcc tgccaaaaca 1500

gcaagcgcct ggaagagaaa aggagattac agcttcccca gactacctgg agatagccat 1560

ttactgcata ggggtcttct taatcgcctg tatggtggta acagtcatcc tgtgccgaat 1620

gaagaacacg accaagaagc cagacttcag cagccagccg gctgtgcaca agctgaccaa 1680

acgtatcccc ctgcggagac aggtaacagt ttcggctgag tccagctcct ccatgaactc 1740

caacaccccg ctggtgagga taacaacacg cctctcttca acggcagaca cccccatgct 1800

ggcaggggtc tccgagtatg aacttccaga ggacccaaaa tgggagtttc caagagataa 1860

gctgacactg ggcaagcccc tgggagaagg ttgctttggg caagtggtca tggcggaagc 1920

agtgggaatt gacaaagaca agcccaagga ggcggtcacc gtggccgtga agatgttgaa 1980

agatgatgcc acagagaaag acctttctga tctggtgtca gagatggaga tgatgaagat 2040

gattgggaaa cacaagaata tcataaatct tcttggagcc tgcacacagg atgggcctct 2100

ctatgtcata gttgagtatg cctctaaagg caacctccga gaatacctcc gagcccggag 2160

gccacccggg atggagtact cctatgacat taaccgtgtt cctgaggagc agatgacctt 2220

caaggacttg gtgtcatgca cctaccagct ggccagacgg atggagtact tggcttccca 2280

aaaatgtatt catcgagatt tagcagccag aaatgttttg gtaacagaaa acaatgtgat 2340

gaaaatagca gactttggac tcgccagaga tatcaacaat atagactatt acaaaaagac 2400

caccaatggg cggcttccag tcaagtggat ggctccagaa gccctgtttg atagagtata 2460

cactcatcag agtgatgtct ggtccttcgg ggtgttaatg tgggagatct tcactttagg 2520

gggctcgccc tacccaggga ttcccgtgga ggaacttttt aagctgctga aggaaggaca 2580

cagaatggat aagccagcca actgcaccaa cgaactgtac atgatgatga gggactgttg 2640

gcatgcagtg ccctcccaga gaccaacgtt caagcagttg gtagaagact tggatcgaat 2700

tctcactctc acaaccaatg aggaatactt ggacctcagc caacctctcg aacagtattc 2760

acctagttac cctgacacaa gaagttcttg ttcttcagga gatgattctg ttttttctcc 2820

agaccccatg ccttacgaac catgccttcc tcagtatcca cacataaacg gcagtgttaa 2880

aacatgaatg actgtgtctg cctgtcccca aacaggacag cactgggaac ctagctacac 2940

tgagcaggga gaccatgcct cccagagctt gttgtctcca cttgtatata tggatcagag 3000

gagtaaataa ttggaaaagt aatcagcata tgtgtaaaga tttatacagt tgaaaacttg 3060

taatcttccc caggaggaga agaaggtttc tggagcagtg gactgc 3106

1. A method for determining whether a subject is predisposed for major depression disorder, the method comprising the steps of:

(i) isolating a subject's brain tissue, wherein the brain tissue is dorsolateral prefrontal cortex tissue;

(ii) contacting the subject's isolated brain tissue with a nucleic acid reagent that selectively associates with a polynucleotide with 95% identity to SEQ ID NO. 1;

(iii) detecting the level of reagent that selectively associates with the said polynucleotide; and

(iv) comparing the detected level of selectively associated reagent with a control, whereby if the detected level is significantly less than the control, an increased likelihood that the subject has or is predisposed for major depression disorder is determined; and whereby, if the detected level is not significantly less than the control, an increase in said likelihood is not determined by the method.

2.-**29**. (canceled)

30. The method of claim **1**, wherein the subject is deceased.

* * * * *