Transcriptome profile of the early stages of breast cancer tumoral spheroids

Rosario Pacheco-Marín [a,e], Jorge Melendez-Zajgla [b], Gonzalo Castillo-Rojas [c], Edna Mandujano-Tinoco [b], Alfredo Garcia-Venzor [b], Salvador Uribe-Carvajal [d], Alfredo Cabrera-Orefice [d], Carolina Gonzalez-Torres [b], Javier Gaytan-Cervantes [b], Irma B Mitre-Aguilar [f] and Vilma Maldonado[a]*.

[a] Epigenetics and [b] Functional Genomics laboratories, National Institute of Genomic Medicine. Periférico Sur No.4809, Col Arenal Tepepan, Delegación Tlalpan, México, D.F., C.P 14610

[c] Microbial Molecular Immunology Program, Department of Microbiology and Parasitology. Faculty of Medicine, National Autonomous University of Mexico (UNAM), University City Avenue 3000 C.P. 04510, Coyoacan, Mexico City.

[d] Department of Molecular Genetics, Institute of Cellular Physiology (UNAM), University City Avenue 3000 C.P. 04510, Coyoacan, Mexico City.

[e] Posgraduate Program in Biological Sciences, Faculty of Medicine (UNAM), University City Avenue 3000 C.P. 04510, Coyoacan, Mexico City.

[f] Unit of Biochemistry, National Institute of Medical Sciences and Nutrition Salvador Zubirán (INCMNSZ), Av. Vasco de Quiroga Nº 15, Colonia Belisario Domínguez Sección XVI, Delegación Tlalpan. CP.14080, México D. F., México

* Correspondence to vilmaml@gmail.com

Supplementary Table S1. Primers of lncRNAs, mRNAs and endogenous control (TBP)

GEN	Sequence forward	Sequence reverse
lnc-CAMK1G-1:2	5´-CTCCCTGCTTCGGGTCAGA-3´	5´-GCCCCATAAAAGGCTGTTCTC-3´
lnc-PPP2R4-4:1	5´-CCCTGCACGTGGATATTAGAGTTT-3´	5´-GGGACCTTGCACCTACAAAATC-3´
lnc-AGBL1-4:1	5´-AATGGCAACCATCAAATCAAGA-3´	5´-GCAAAGCATCACAATTCATGGA-3´
lnc-KIAA1755-5:3	5´-TCTCGAAATCTGCAGACATCTTG-3´	5´-GTGAGAGTGGAGCCGACCAT-3´
lnc-IGSF5-1:1	5´-GGGCCACAACTCAACCCATA-3´	5´-GCTGGTCGTTGCTCAAGAAAC-3´
lnc-MLN-1:2	5´-TTCAGGCCCCGAGTTTCTC-3´	5´-GGGCAAGGGCAGAGTAGGA-3´
lnc-CPEB4-6:1	5´- ACCTCTCCTGCGTTTGATCTG-3´	5´-GGCCGTGGGCAAGTTACTT-3´
lnc-MYC-2:24, 2:5, 2:8 and 2:9 isoforms	5´-CTTCCAGTGGATTTCCTTGC-3´	5´-TTTCTCCCAGACACGGATTC-3´
lnc-GAPDH-2:1	5´-GGCTGTTGTGATTCAGTTGG-3´	5´-ATCGCCCACCAAGATTACAG-3´
lnc-HIST1H2AG-2:1	5´-ATGGTTTGCAGAGGCTCAAG-3´	5´-TGGGTGACAGGTTTGATGTC-3´
lnc-ANKRD30A-4:1	5´-TGCTGCAGGCAATTATGC-3´	5´-GCTTTTTCCAAAGGGGTTG-3´
MYC	5´-GTTTGCCATTACCGGTTCTC-3´	5´-TGCAGAAGGTCCGAAGAAAG-3´
GAPDH	5´-CAATGACCCCTTCATTGACC-3´	5´-ATGACAAGCTTCCCGTTCTC-3´
HIST1H2AG	5´-TGCCCAAAAAGACTGAGAGC-3´	5´-AAAGAGCCGTTGGTTTGGAC-3´
ANKRD30A	5´-AACACGGCTCTCCATTATGC-3´	5´-TAGCCTTGTTGTGCACTTCG-3´
COX8A	5´-TCCCCTGGATCATGTCATTC-3´	5´-CACCATGGAGACATCAAGAGG-3´
NDUFA1	5´-CGTGTGCTTGTTGATTCCAG-3´	5´-TGCGCCTATCTCTTTCCATC-3´
TP53	5´-GTCTTTGAACCCTTGCTTGC-3´	5´-CCACAACAAAACACCAGTGC-3´
ITGB6	5´-CGACTGTGACTGTGGTGAAT-3´	5´-CTTTGGTTCAGAAATGCAAG-3´
TBP	5´-CCACAGCTCTTCCACTCACA -3´	5´-CTCATGATTACCGCAGCAAA -3´

Supplementary Table S2. Cell index between monolayers and spheroids disaggregated, as well as MDA-MB- 231 monolayer cells (Positive control)

Culture condition	**Time**			
	24h		**30h**	
	Cell index	p-value	Cell index	p-value
Monolayer	0.02 ± 0.01	<0.0001	0.02 ± 0.01	<0.0001
Spheroids disaggregated 6 days	0.26 ± 0.11		0.42 ± 0.04	
MDA-MB-231	1.684 ± 0.11		1.797 ± 0.07	

Supplementary Table S3. Genes mRNAs top up/down related to breast cancer and metastasis

GEN	Name	References
CLDN4*	Claudin 4	1-4
FST*	Follistatin	5-8
MIF*	Macrophage migration inhibitory factor (glycosylation-inhibiting factor)	9,10
NPY1R*	Neuropeptide Y receptor Y1	11
PGR*	Progesterone receptor	12
PMP22*	Peripheral myelin protein 22	13-15
S100A9*	S100 calcium binding protein A9	16
TFF1*	Trefoil factor 1	17
RERG*	RAS-like, estrogen-regulated, growth inhibitor	18
COL12A1*	Collagen, type XII, alpha 1	19
COL4A5*	Collagen, type IV, alpha 5	20,21
S100A6	S100 calcium binding protein A6	22
RPS12	Ribosomal protein S12	23,24
NAALADL2	N-acetylated alpha-linked acidic dipeptidase-like 2	25
IFIT2	Interferon-induced protein with tetratricopeptide repeats 2	26,27
ATP5E	ATP synthase, H+ transporting, mitochondrial F1 complex	28
ANKRD1	Ankyrin repeat domain 1 (cardiac muscle)	29
RPS21	Ribosomal Protein S21	30
POF1B	Premature ovarian failure, 1B	31
RPL36AL	Ribosomal protein L36a-like	32

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* Related genes in breast cáncer metastasis and down-regulated are in bold letters. *See references on next page*

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Supplementary Table S4. Genes up and down regulated of top five canonical pathways (IPA)

Pathways	Up-regulated	Down-regulated	Total
1.EIF2 signaling	**AKT1, PPP1CA,** **EIF** (1/2AK1/2B2/2B5/3B/3C/3D/3G/3I/3K/3L/4A1-4A3/4G1) **RPL** (3-8/11-14/17-19/22-24/ 26-27/29-32/34-39/41/10A/ 13A/18A/23A/26L1/35A/36AL/ 37A/7A/P0-P2) **RPS** (2/3/5-9/11-16/18-21/23/25/27-30/15A/3A/4X/A)	**PIK3CA** and **RRAS2**	87
2.Mitochondrial dysfunction	*Include all genes the oxidative phosphorylation more:* **AIFM1**, **APH1A**, **FIS1**, **GPX4**, **HSD17B10**, **PARK7**, **PRDX** (3 and 5) and **TXN2**	**BACE2**, **BCL2**, **NDUFAF2** and **MAP2K4**	55
3.mTOR signaling	**AKT1**, **DDIT4**, **RND3**, **EIF** (3B/3C/3D/3G/3I/3K/3L/4A1-4A3/4EBP1/4G1) **PPP2** (R4 and R1A), **RAC1**, **RHO** (A and C), **VEGFA** and *include all the genes* **RPS** *of pathway* EIF2 *signaling*	**PIK3CA**, **RICTOR**, **RPS6KA3** and **RRAS2**	52
4.Regulation of EIF4 and p70S6K	**AKT1**, **PPP2** (R4 and R1A), **EIF4EBP1** more *include all the genes* **EIF** (*except* EIF2AK1) *and* **RPS** *of pathway* EIF2 *signaling*	**ITGA2**, **PIK3CA** and **RRAS2**	48
5.Oxidative phosphorylation	**ATP** (5B/5C1/5E/5G1/5H/5I/ 5J2/5L/5O) **NDUF** (A1/A2/A6/A9/A13/B1-B3/B7-B11/S2-S3/S5/S8/V1) **COX** (4I1/5A/5B/6A1/6B1/7A2/ 7B/7C/8A), **CYB5A**, **CYC1**, **SDHB** and **UQCR** (C1 and H)	**NDUFS4**	42

Supplementary Table S5. Canonical pathways related to cellular cycle (IPA)

Pathways	**Up-regulated**	**Down-regulated**	**Total**
PI3K/AKT signaling	AKT1, CDC37, CDKN1B, EIF4EBP1, HSP90AB1, HSP90B1, INPPL1, PPP2R4, PPP2R1A and TP53	BCL2, ITGA2, PIK3CA and RRAS2	14
PTEN signaling	AKT1, CDKN1B, INPPL1, MCRS1 and PIK3CA	BCL2, BMPR2, ITGA2, MAGI3, RAC1 and RRAS2	11
Cell cycle: G1/S Checkpoint regulation	CDK4, CDKN1B, E2F4, MAX, MYC, PA2G4, TP53, RPL5 and RPL11		9
HER-2 signaling in breast Cancer	AKT1, CDKN1B, ITGB5 and TP53	AREG, ITGB6, PARD3, PIK3CA and RRAS2	9
Cell cycle regulation by BTG family protein	CDK4, E2F4, PPP2R4, PPP2R1A and PRMT1		5
Estrogen-mediated S-phase entry	CDK4, CDKN1B, E2F4 and MYC		4

Supplementary Table S6. Relations between lncRNA and the a adjacent mRNA protein-coding gene differentially expressed.

Symbol mRNA	Gene name	Fold change	Symbol lncRNA	Fold change	Location lncRNA	Location mRNA	Chr
ABHD2	Abhydrolase domain containing 2	2.5	ABHD2-1:5	**	89584454-89626050	89631381-89745591	15
ADI1	Acireductone dioxygenase 1	3.0	ADI1-2:1	**	3302112-3305236	3501690-3523350	2
ANKRD30A	Ankyrin repeat domain 30A	*	ANKRD30A-4:1	*	37229037-37232860	37414785-37521495	10
ANKRD40	Ankyrin repeat domain 40	2.2	ANKRD40-1:1 ANKRD40-1:5	* *	48840212-48844918 48838535-48844877	48770551-48785270	17 17
ARL15	ADP-ribosylation factor-like 15	-4.6	ARL15-1:1	*	53072474-53115516	53180614-53606403	5
ARL4A	ADP-ribosylation factor-like 4A	-2.0	ARL4A-1:1	*	13141016-13142014	12726452-12730559	7
ARMCX3	Armadillo repeat containing, X-linked 3	2.1	ARMCX3-1:1	2.7	100882858-100883508	100878120-100882831	X
ATF7IP	Activating transcription factor 7 interacting protein	-3.2	ATF7IP-3:1	**	14369524-14374231	14518611-14651697	12
BOD1	Biorientation of chromosomes in cell division 1	2.7	BOD1-2:1	*	173217812-173236042	173034148-173043666	5
CCT5	Chaperonin containing TCP1, subunit 5 (epsilon)	2.2	CCT5-2:1	-2.5	10137250-10138477	10250282-10266501	5
CD63	CD63 molecule	2.1	CD63-1:1	6.0	56155334-56156412	56119230-56122910	12
CHD2	Chromodomain helicase DNA binding protein 2	-2.4	CHD2-4:2	-2.5	93855786-94098099	93443551-93571237	15
CHMP1A	Chromatin modifying protein 1A	2.6	CHMP1A-2:1	*	89458783-89478972	89710843-89724129	16
COMMD10	COMM domain containing 10	-2.4	COMMD10-3:1	**	115910178-115927094	115420727-115628978	5
COMMD3	COMM domain containing 3	2.1	COMMD3-1:1	*	22546715-22547477	22605312-22609246	10
CYB5A[4]	Cytochrome b5 type A (microsomal)	2.6	CYB5A-1:2	*	71878626-71879804	71920527-71959251	18
DAD1	Defender against cell death 1	2.4	DAD1-2:1 DAD1-2:6	** **	22849083-22951948 22850773-22901719	23033807-23058143	14
EEF1B2	Eukaryotic translation elongation factor 1 beta 2	5.4	EEF1B2-2:2	**	206950246-206950880	207024318-207027653	2
EIF3B[1]	Eukaryotic translation initiation factor 3, subunit B	3.1	EIF3B-1:1	**	2423756-2428232	2394474-2420380	7
EIF4A3[1]	Eukaryotic translation initiation factor 4A, isoform 3	2.3	EIF4A3-1:1	4.0	77965743-77966640	78109013-78120982	17
FAM83B	Family with sequence similarity 83, member B	-2.4	FAM83B-1:1	2.3	54807965-54809897	54711569-54806820	6
GAPDH	Glyceraldehyde-3-phosphate dehydrogenase-like 6	2.7	GAPDH-2:1	**	6619441-6619688	6643657-6647536	12
GGCT	Gamma-glutamyl cyclotransferase	3.2	GGCT-1:21	-7.0	30555925-30602441	30536237-30544457	7
GLCE	Glucuronic acid epimerase	-2.0	GLCE-1:1	**	69571014-69580903	69452973-69564544	15
H3F3A	H3 histone family 3A	-2.9	H3F3A-1:1	*	226274582-226277993	226250408-226259703	1
HIATL1	Hippocampus abundant transcript-like 1	-2.5	HIATL1-4:1	3.9	97320996-97330312	97136833-97223202	9
HIST1H2AG	Histone cluster 1, H2AG	4.5	HIST1H2AG-2:1	8.0	26988232-26991703	27100817-27101314	6
ITGA2[7]	Integrin, alpha 2 (CD49B, alpha 2 subunit of VLA-2 receptor)	-2.3	ITGA2-1:3	*	52405707-52410027	52285156-52390609	5
LEMD3	LEM domain containing 3	-2.2	LEMD3-1:1	**	65675437-65680508	65563351-65642141	12
LRBA	LPS-responsive vesicle trafficking, beach and anchor containing	-2.3	LRBA-2:1	-3.5	150509142-150817385	151185594-151936649	4
MYC[3]	v-myc myelocytomatosis viral oncogene homolog (avian)	4.4	MYC-2:24 MYC-2:5 MYC-2:8 MYC-2:9	* ** 4.5 2.2	129001463-129108824 128806806-128951855 128808059-128903041 128808066-128903206	128748315-128753680	8
NRIP1	Nuclear receptor interacting protein 1	-2.8	NRIP1-2:1	2.7	16742841-16774624	16333556-16437126	21
NT5DC1	5'-nucleotidase domain containing 1	-2.7	NT5DC1-1:1	4.0	116575370-116577906	116421999-116566853	6
PARK7[6]	Parkinson disease (autosomal recessive, early onset) 7	2.8	PARK7-1:1	*	8086798-8182762	8021714-8045342	1
PCBP1	Poly(rC) binding protein 1	3.0	PCBP1-1:1	**	70316853-70323232	70314585-70316335	2
PGR	Progesterone receptor	-14.0	PGR-1:1	**	100554893-100558686	100900355-101000544	11
PGRMC1	Progesterone receptor membrane component 1	2.6	PGRMC1-1:1	2.1	118425492-118469573	118370211-118378429	X
PLCB1	Phospholipase C, beta 1 (phosphoinositide-specific)	-4.6	PLCB1-1:1	**	8000549-8008603	8113296-8865547	20
PMP22	Peripheral myelin protein 22	18.4	PMP22-1:1	**	15170778-15175607	15133096-15168644	17

PPP2R4[5]	Protein phosphatase 2A activator, regulatory subunit 4	3.7	PPP2R4-4:1	-3.1	132083295-132087184	131873228-131911225	9
			PPP2R4-5:1	*	132094579-132101180		
			PPP2R4-5:3	-5.4	132099158-132109743		
			PPP2R4-5:4	-3.0	132104122-132121817		
PSME1	Proteasome (prosome, macropain) activator subunit 1 (PA28 alpha)	3.0	PSME1-1:2	3.1	24608654-24609653	24605378-24608176	14
RASA1	RAS p21 protein activator (GTPase activating protein) 1	-3.0	RASA1-3:4	4.0	87564841-87581884	86564151-86687733	5
			RASA1-3:9	3.8	87564864-87583473		
RPL24[2]	Ribosomal protein L24	4.0	RPL24-2:3	**	102382007-102392757	101399934-101405563	3
RPLP1[2]	Rbosomal protein, large, P1	4.4	RPLP1-1:1	**	69755365-69857576	69745159-69747884	15
			RPLP1-1:11	**	69854062-69863422		
			RPLP1-1:18	10.0	69857531-69863775		
			RPLP1-1:9	**	69850531-69863422		
RPS14[1]	Ribosomal protein S14	4.5	RPS14-1:2	**	149855094-149865531	149823792-149829319	5
RPS7[1]	Ribosomal protein S7	2.3	RPS7-1:10	2.3	3606168-3609321	3622853-3628509	2
SBDS	Shwachman-Bodian-Diamond syndrome pseudogene	2.5	SBDS-5:1	-4.0	65956996-65958553	66452690-66460588	7
			SBDS-5:2	-2.9	65956083-65958525		
SCD	Stearoyl-CoA desaturase (delta-9-desaturase)	7.0	SCD-1:10	2.0	102133372-102143125	102106772-102124588	10
SLC12A2	Solute carrier family 12 (sodium/potassium/chloride transporters), member 2	3.3	SLC12A2-1:2	**	127270935-127277199	127419483-127525380	5
SLC16A4	Solute carrier family 16, member 4 (monocarboxylic acid transporter 5)	-2.5	SLC16A4-1:1	6.0	110828997-110881793	110905473-110933704	1
SLC25A5	Solute carrier family 25 (mitochondrial carrier; adenine nucleotide translocator), member 5	3.2	SLC25A5-1:1	**	118556880-118557510	118602363-118605359	X
SLITRK6	SLIT and NTRK-like family, member 6	-2.1	SLITRK6-9:4	*	88096247-88323218	86366922-86373483	13
SSR3	Signal sequence receptor, gamma (translocon-associated protein gamma)	2.9	SSR3-3:1	**	156470852-156534851	156257929-156272973	3
			SSR3-3:2	2.2	156465135-156534823		
SUMF2	Sulfatase modifying factor 2	3.3	SUMF2-2:1	**	56560817-56564978	56131917-56148365	7
TCF12	Transcription factor 12	-2.7	TCF12-1:13	*	57592563-57599959	57210833-57580716	15
TMEM167B	Transmembrane protein 167B	2.5	TMEM167B-1:3	2.3	109642815-109643234	109633403-109639554	1
TOB1	Transducer of ERBB2, 1	2.1	TOB1-2:1	*	48986998-48987593	48939584-48941413	17
TRMT112	tRNA methyltransferase 11-2 homolog (S. cerevisiae); similar to CG12975	4.4	TRMT112-2:1	*	64161814-64163303	64084163-64085033	11
USP3	Ubiquitin specific peptidase 3	-2.5	USP3-1:2	2.4	63682429-63729347	63796810-63883663	15
WDR1	WD repeat domain 1	2.3	WDR1-1:1	5.0	10069713-10074643	10075963-10118573	4
WDR74	WD repeat domain 74	2.6	WDR74-1:1	9.3	62619730-62623386	62600383-62607628	11
			WDR74-1:12	6.0	62621840-62622960		
			WDR74-1:14	2.5	62620826-62622960		
			WDR74-1:22	2.5	62619460-62622708		
			WDR74-1:25	-2.3	62619520-62621168		
			WDR74-1:4	-2.3	62619460-62623339		
ZNF259	Zinc finger protein 259	2.2	ZNF259-1:1	-2.5	116645826-116646592	116649276-116658739	11
ZNF740	Zinc finger protein 740	2.0	ZNF740-1:1	**	53553370-53554784	53574535-53584654	12

* Only expressed in monolayers

** only expressed in spheroids

[1, 2] Genes of EIF2 signaling

[1] Genes of mTOR signaling

[1, 5, 7] Genes of regulation of EIF4 and p70S6K pathways

[3] Gen of cell cycle: G1/S Checkpoint and estrogen-mediated S-phase entry

[4, 6] Genes of mitochondrial dysfunction and Oxidative phosphorylation pathways

[5] Genes Cell cycle by BTG,

[5, 7] Genes PI3K/AKT pathways

[7] Genes of PTEN signaling

Supplementary Table S7. qRT-PCR validation of lncRNAs and mRNAs genes.

GEN	**Relative expression**		**p-value**	**Validation method**
	qRT-PCR	**RNA-seq**	**qRT-PCR**	
lnc-AGBL1-4:1	-19.69 ± 1.68	-5.1	0.001	Relative standard curve
lnc-CAMK1G-1:2	-2.77 ± 0.65	-11.3	0.001	Relative standard curve
lnc-PPP2R4-4:1	-1.57 ± 0.51	-3.1	0.042	Relative standard curve
lnc-KIAA1755-5:3	0.57 ± 0.33	-4.0	**0.265**	Relative standard curve
lnc-MLN-1:2	3.24 ± 1.12	23	0.022	Relative standard curve
lnc-IGSF5-1:1	1.75 ± 0.70	22	0.015	Relative standard curve
lnc-CPEB4-6:1	2.44 ± 1.02	19	0.030	Relative standard curve
ITGB6	0.15554072 ± 0.30	-10.52	0.00147	Comparative ΔΔ C_T
ANKRD30A	0.144032353 ± 0.23	*	0.00001	Comparative ΔΔ C_T
MYC	0.84319542 ± 0.22	4.4	0.0007	Comparative ΔΔ C_T
GAPDH	1.292590582 ± 0.15	2.65	**0.406**	Comparative ΔΔ C_T
p53	1.555923172 ± 0.06	2.30	0.0015	Comparative ΔΔ C_T
HIST1H2AG	1.076666667 ± 0.17	4.49	**0.06**	Comparative ΔΔ C_T
NDUFA1	3.494077936 ± 0.15	5.8	0.0008	Comparative ΔΔ C_T
COX8A	9.030698223 ± 0.16	5.74	0.013	Comparative ΔΔ C_T
lnc-GAPDH-2:1	3.07182457 ± 0.19	**	0.028	Comparative ΔΔ C_T
lnc-HIST1H2AG-2:1	3.83134634 ± 0.08	8.0	0.041	Comparative ΔΔ C_T
lnc-MYC-2:24 **2:5** **2.8** **2:9 isoforms**	0.23693361 ± 0.22	* ** 4.5 2.2	0.000004	Comparative ΔΔ C_T

Genes * only monolayer or ** only spheroids this was expressed

Supplementary Table S8. lncRNAs dysregulated with know function.

ID lncRNA LNCipedia (Ensembl)	**Disease**	**Description**	**Ref.**
lnc-C22orf32-1	Nasopharyngeal carcinoma (NPC)	High levels of lnc-C22orf32-1 was found in the primary NPC tissues. This increase was associated with advanced tumor stages. High levels of lnc-C22orf32-1 were associated with the male patients *The differential expression of lncRNA observed in NPC might be attributed to epigenetic regulation including microRNA, DNA methylation, and histone modification	1, 2
lnc-MYC-2 (PVT1)	Breast, Ovarian Pancreas Renal and Gastric cáncer Diabetes and other	e.g., Amplification of PVT1 contributes to the pathophysiology of ovarian and breast cancer. *The lncRNA PVT1 interacts with MYC in the nucleus.	1, 3*
lnc-CPN2-1 (LINC00887)	Renal cell carcinoma (RCC)	lnc-CPN2-1 overexpression in RCC Small interfering RNA (siRNA)-mediated knockdown of lnc-CPN2-1 did not influence cell proliferation	4
lnc-PPP2R4-5 (LINC01503) lnc-FAM46A-1 (LINC01526) lnc-MBL2-4 (LINC01468)	Tongue squamous cell carcinoma (SCC)	These lncRNAs were upregulated in the microdissected tongue SCC tissues. High level of lnc-MBL2-4:3 was associated with the node positive tongue cancer patients In addition, reduced expression of lnc-MBL2-4:3 in the cisplatin resistant HN21B cells	5
lnc-HSCB-1 (TTC28-AS1)	HSCs (hematopoietic stem cell) and limphoid progenitor cells	This lnc-RNA showed a strong co-expression with the BCR protein-conding gene. *BCR can be considered as a candidate tumor suppressor gene which may be involved in meningioma pathogenesis	6,7*
lnc-SERPINC1-1 (GAS5)	Breast,Renal cancer B-cell neoplasms *Kidney,Lymphoma Melanoma, Prostata Systemic lupus erythaematosus and autoimmune disease	e.g., GAS5 promoted the apoptosis of triple-negative and oestrogen receptor-positive cells but only dual PI3K/mTOR inhibition was able to enhance GAS5 levels in all cell types	1, 8*
Lnc-DDX27-1 (ZFAS1)	Breast and Ductal carcinoma	ZFAS1 show distinct patterns of expression during post-pubertal mammary Development. ZFAS1 is a putative tumor suppressor gene.	1, 9
lnc-C11orf89-2 (H19)	Breast, bladder, cervical, adrenocortical cancer and other	Estrogen signaling increased the expression of *H19* and ERα. H19 expression *also is* important for estrogen-induced luminal progenitor expansion. *c-Myc and H19 expression shows strong association in primary breast and lung carcinomas	1*, 10
lnc-SCYL1-1 (MALAT1)	Breast, cervical, bladder hepatocelluar	The MALAT1 level was significantly lower in tumor tissue compared to adjacent non-cancerous tissue in breast cancer. Downregulation of MALAT1 induced EMT.	11

	carcinoma and other	Induction of EMT by MALAT1 downregulation may be mediated by the PI3K-AKT pathway	
lnc-DLK1-4 (MEG3)	Breast, bladder cancer, Glioma	MEG3a exhibited a strong ability to inhibit cell growth and proliferation in several human cancer cell lines, such HeLa, MCF-7, and H4.	12
lnc-NAP1L2-1 (XIST1)	Breast, ovarian, bladder, testicular cáncer and other	XIST typically is expressed by all female somatic cells, XIST expression has been found to be lost in female breast, ovarian, and cervical cancer cell lines.	13
lnc-TMEM30B-4:1 (HIF1A-AS1)	*Non-small cell lung cancer (NSCLC), Kidney, Renal Oesophageal denocarcinoma and Cardiovascular disease	*The expression of XIST and HIF1A-AS1 in tumor tissues and serum of NSCLC was higher than that of the control group (nontumourous)	1, 14
lnc-ATXN7-8 (ADAMTS9-AS2)	Glioma	ADAMTS9-AS2 was downregulated in glioma tissue. Co-relationship of ADAMTS9-AS2 with ADAMTS9 and DNMT1. Overexpression of ADAMTS9-AS2 inhibited migration and invasion abilities of the T98G cells While knockdown opposite effect in U87 cells	15
lnc-TMEM60-1 (APTR)	Glioblastoma	*APTR* represses *p21 t*ranscription by recruiting the PRC2 complex to the *p21* promoter. *APTR* is important for G1-S transition most likely due to a requirement for the activity of CyclinE/CDK2 kinase	16
lnc-METTL15-4 (BDNF-AS)	Huntington's disease (HD) Obesity, Schizophrenia Other	*BDNF*-AS and *BDNF* transcripts are Co-expressed in many tissues, which suggest *BDNF*-AS potential for regulation of *BDNF* mRNA in monkey and mouse tissues *BDNF-AS inhibits BDNF transcription by recruiting EZH2 to the BDNF promoter region and plays an important role in the development of HD	1, 17
lnc-EMX2-4 (CASC2)	Glioma, endometrial, colorectal cáncer snd Melanoma, renal carcinoma	CASC2 was lowly expressed in glioma tissues. Overexpression of CAS2 inhibited proliferation, migration, and invasion, and promoted cell apoptosis. Up-regulated CASC2 decreased the expression of miR-2.	1,18
lnc-RPLP1-1 (DRAIC)	Prostate cancer	Prostate cancer is accompanied by a decrease of FOXA1 and NKX3-1, which leads to the decrease of tumor-suppressive lncRNAs, DRAIC and PCAT29.	19
lnc-CHIC1-3 (JPX)	Embryonic stem cell (ES)	*Jpx* expression increases during ES cell differentiation and remained elevated in somatic cells *Xist* is controlled by two parallel RNA switches – Tsix for Xa and Jpx for Xi. Whereas Tsix represses *Xist* on Xa, Jpx activates *Xist* on Xi	20
lnc-LPAR6-1 (LINC00441)	Lung carcinoma cell	A549 cells shows a predominantly nuclear localization of ncRNA-RB1 (LINC00441). This lncRNA no effects on RB1 transcript or protein levels, suggesting that the two transcripts do not regulate each other.Transcriptional regulation of CALR expression by ncRNA-RB1	21
lnc-MAF-2 (MAFTRR)	T and B lymphocyte	High expression of linc-MAF-4 correlated with a low amount of *MAF* transcript in $CD4^+$ T_H1 cells, conversely T_H2 cells had low expression of linc-MAF-4 and abundant *MAF* transcripts.	22

lnc-TRAF5-1 (LINC00467)	Neuroblastoma	Linc00467 suppressed the expression of its downstream protein-coding gene RD3, and induced neuroblastoma cell survival by reducing the expression of the tumour suppressor gene DKK1 Down-regulation of N-Myc expression resulted in increased linc00467 expression.	23
lnc-YBEY-2 (MCM3AP-AS1)	Human B-cell	This lncRNA were enriched in the nucleus and were upregulated in high MYC expression. MYC binding was enriched at the promoter of these lncRNA	24
lnc-NEDD1-4 (RMST)	Brain *Rhabdomyosarcoma	*RMST* associates with SOX2, one of the most important transcription factors controlling neural stem cell (NSC) fate Knockdown of *RMST* prevented neuronal differentiation	1*, 25
lnc-ABCB4-1 (TP53TG1)	Colon cancer	Under conditions of cellular stress (ultraviolet irradiation or exposure to bleomycin or cisplatin), expression of TP53TG1 was induced in a wild-type TP53-dependent manner	26
lnc-OSBP2-2 (TUG1)	MCF-7 and HeLa cells B-cell neoplasms bladder cáncer non-small cell lung cancer Huntington's disease	*Under genotoxic stress-induced apoptosis in HeLa and MCF-7 cells TUG1, UCA1, and PANDA not affected. Cell proliferation inhibition and apoptosis induction were observed in TUG1 siRNA-transfected bladder urothelial carcinoma T24 and 5637 cells.	1, 27
lnc-HEATR6-1 (WFDC21P)	Dendritic cells	lnc-D (WFDC21P), was exclusively expressed in human conventional dendritic cells (DCs). Knockdown of lnc-DC impaired DC differentiation from human monocytes and mouese bone marrow cells and reduced capacity of DCs to stimulate T cell activation. Lnc-DC mediated these effects by activating the transcription factor STAT3.	28
lnc-TGFBR2-1 (RP11-1024P17.1)	Vascular disease.	This lncRNA could to be implicated in proliferation and migration of vascular smooth muscle cells (VSMCs)	29

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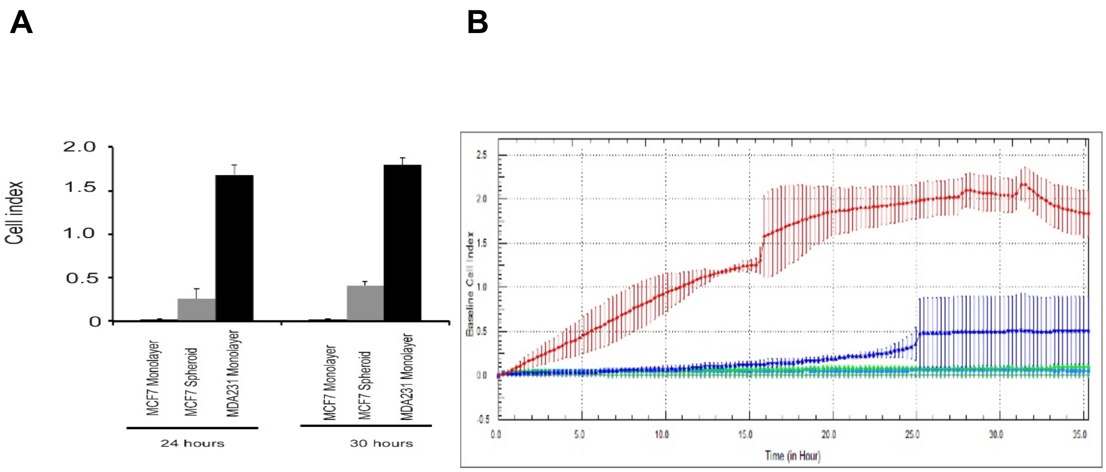
Supplementary Table S9. qPCR quantification for transfection of lncRNAs and the relationship of lncRNA overexpressed with its nearby coding gene (mRNA).

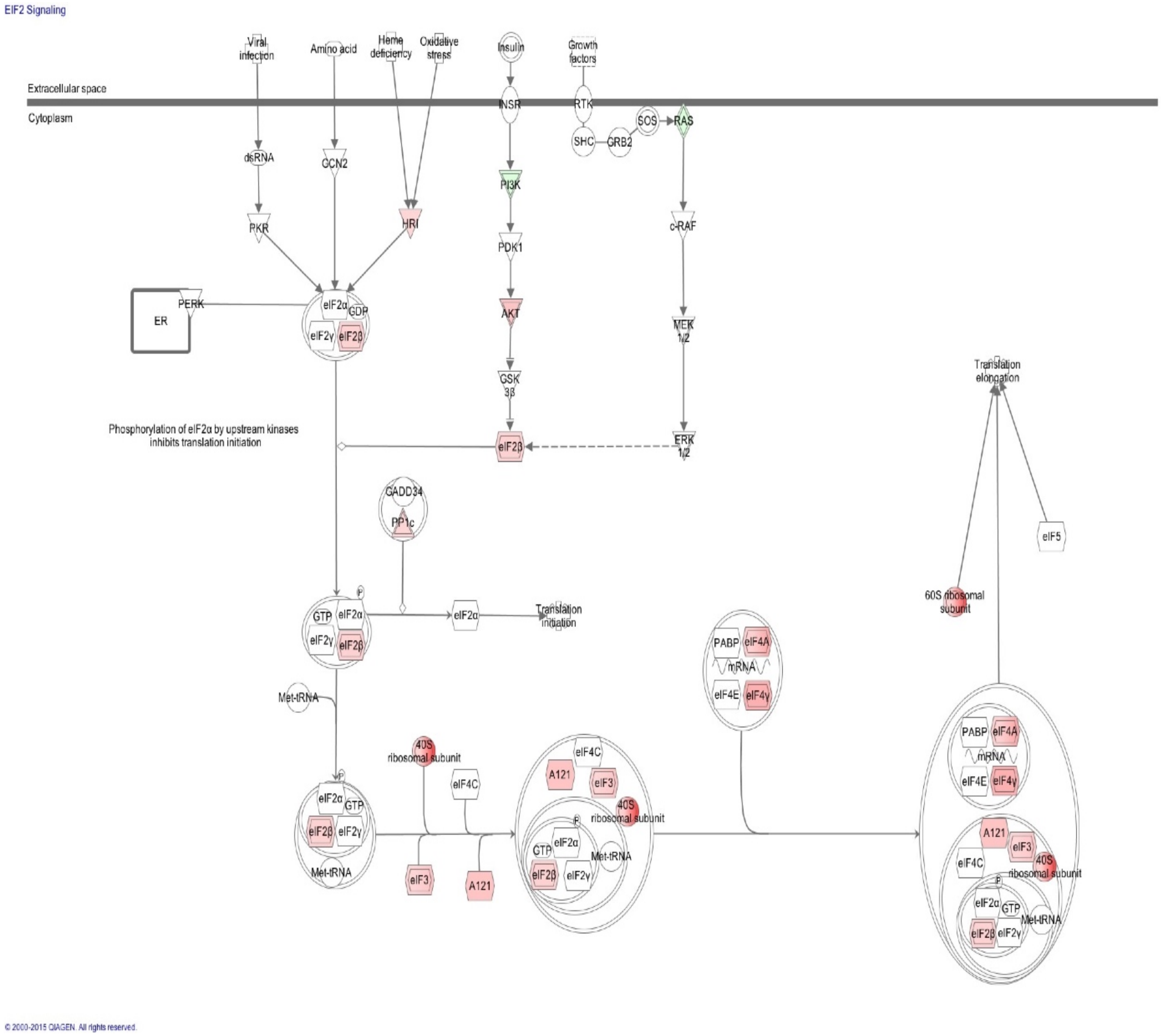
lncRNAs neighbor mRNA	lncRNAs **Fold difference (log$_2$)**		p-value	mRNAs **Fold difference (log$_2$)**		p-value
lncRNA / mRNA	*VEC -*	*VEC+*		*VEC -*	*VEC +*	
lnc-GAPDH-2:1/ mRNA GADPH	-5.75 ± 0.23	5.82 ± 0.16	0.00006	-1.77 ± 0.19	1.78 ± 0.17	0.000007
lnc-HIST1H2AG-2:1/ mRNA HIST1H2AG	-14.68 ± 0.30	15.00 ± 0.22	0.00007	-1.90 ± 0.22	2.05 ± 0.16	0.00026
lnc-MYC-2:24, 2:5, 2.8,2:9 isoforms/ mRNA MYC	-13.44 ± 0.21	13.38 ± 0.36	0.00002	-1.08 ± 0.26	2.70 ± 0.24	0.00045

VEC- Empty vector and VEC + plasmid transfection with lncRNAs



Supplementary Figure S1. Real time MCF-7 monolayers and spheroids disaggregated migration data. **(A)** Results from an RTCA migration assay conducted with FBS in the bottom chamber of a CIM plate well. MCF-7 monolayers cell and spheroids disaggregated cells migration is compared to MDA-MB-231 (Positive control). Results are shown as mean ± SD Cell Index from triplicate wells at the 24 and 30 h time point **(B)** xCELLigence plots representative. MDA-MB-231 in red, spheroids disaggregated in blue and monolayers cell in green, Light colored lines representing conducted without SBF.

A


EIF2 Signaling
Viral infection
Amino acid
Heme deficiency
Oxidative stress
Insulin
Growth factors
Extracellular space
Cytoplasm
INSR
RTK
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RAS
SHC
GRB2
dsRNA
GCN2
PI3K
PKR
HRI
PDK1
c-RAF
PERK
ER
eIF2α
GDP
eIF2γ
eIF2β
AKT
MEK 1/2
GSK 3β
Translation elongation
Phosphorylation of eIF2α by upstream kinases inhibits translation initiation
eIF2β
ERK 1/2
GADD34
PP1c
eIF5
60S ribosomal subunit
P
GTP
eIF2α
eIF2γ
eIF2β
eIF2α
Translation initiation
PABP
eIF4A
mRNA
eIF4E
eIF4γ
Met-tRNA
40S ribosomal subunit
eIF4C
A121
eIF3
eIF4C
40S ribosomal subunit
eIF2α
GTP
eIF2β
eIF2γ
Met-tRNA
eIF3
A121
GTP
eIF2α
Met-tRNA
eIF2β
eIF2γ
PABP
eIF4A
mRNA
eIF4E
eIF4γ
A121
eIF3
eIF4C
40S ribosomal subunit
eIF2α
GTP
Met-tRNA
eIF2β
eIF2γ
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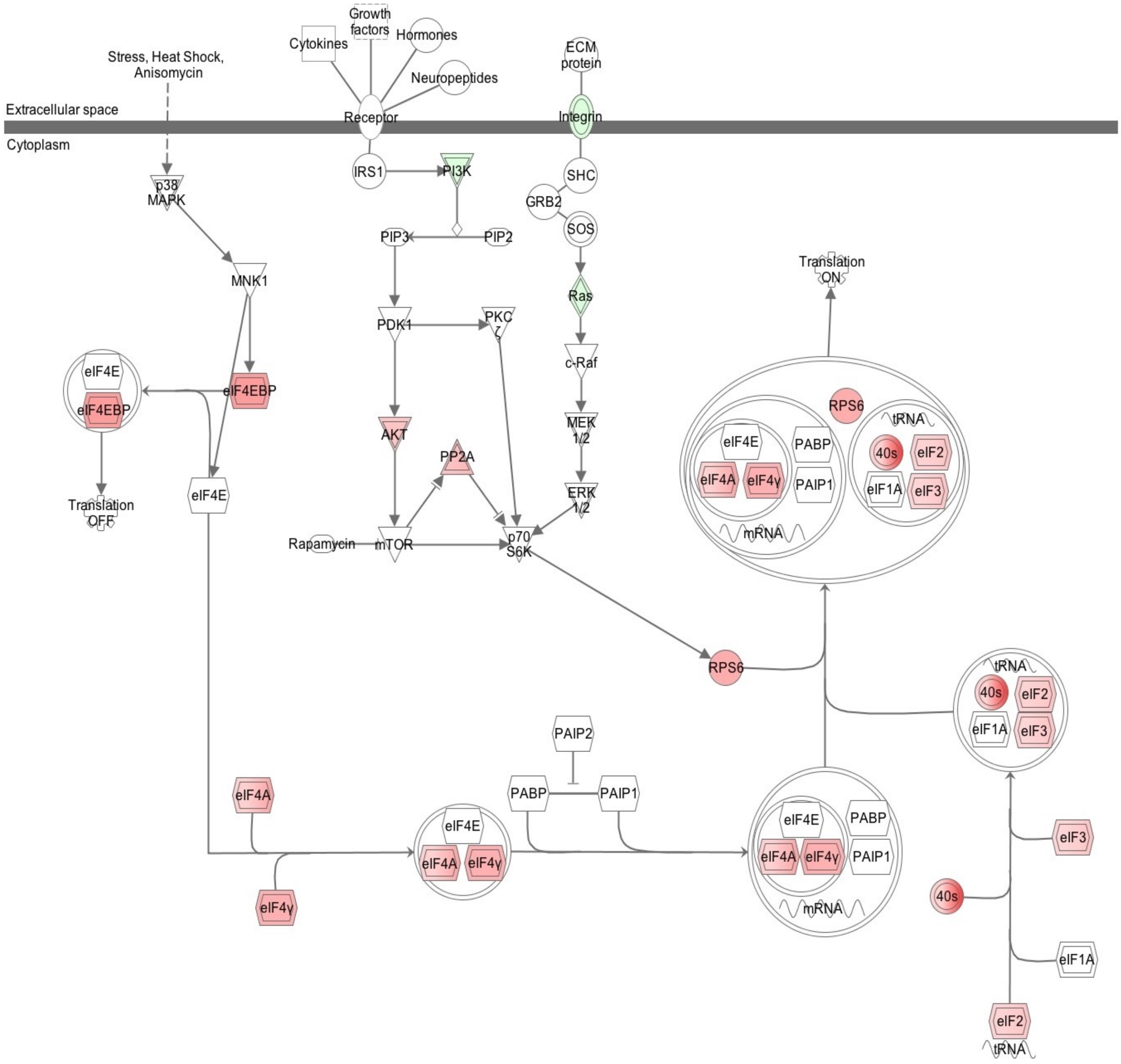
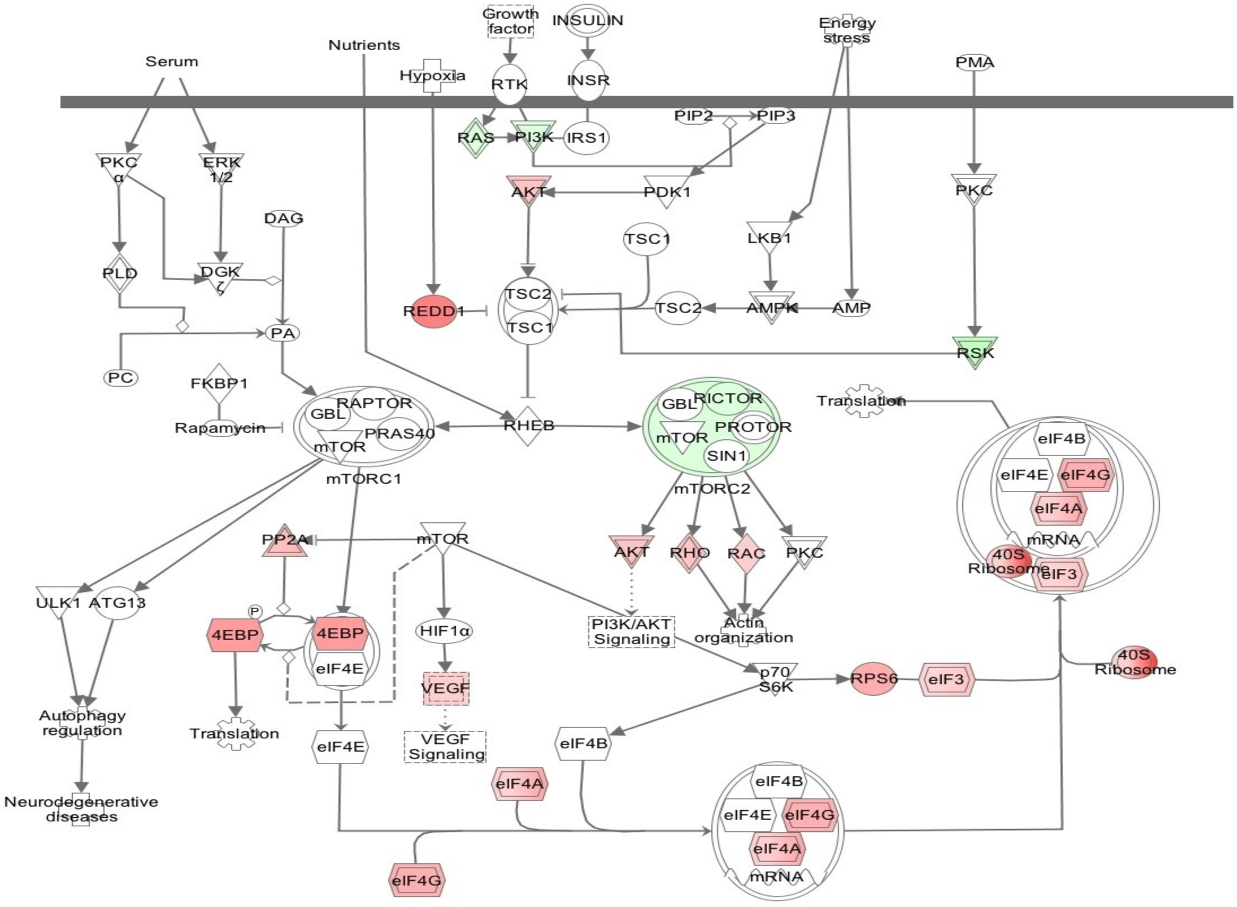
B

Regulation of eIF4 and p70S6K Signaling



C

mTOR Signaling



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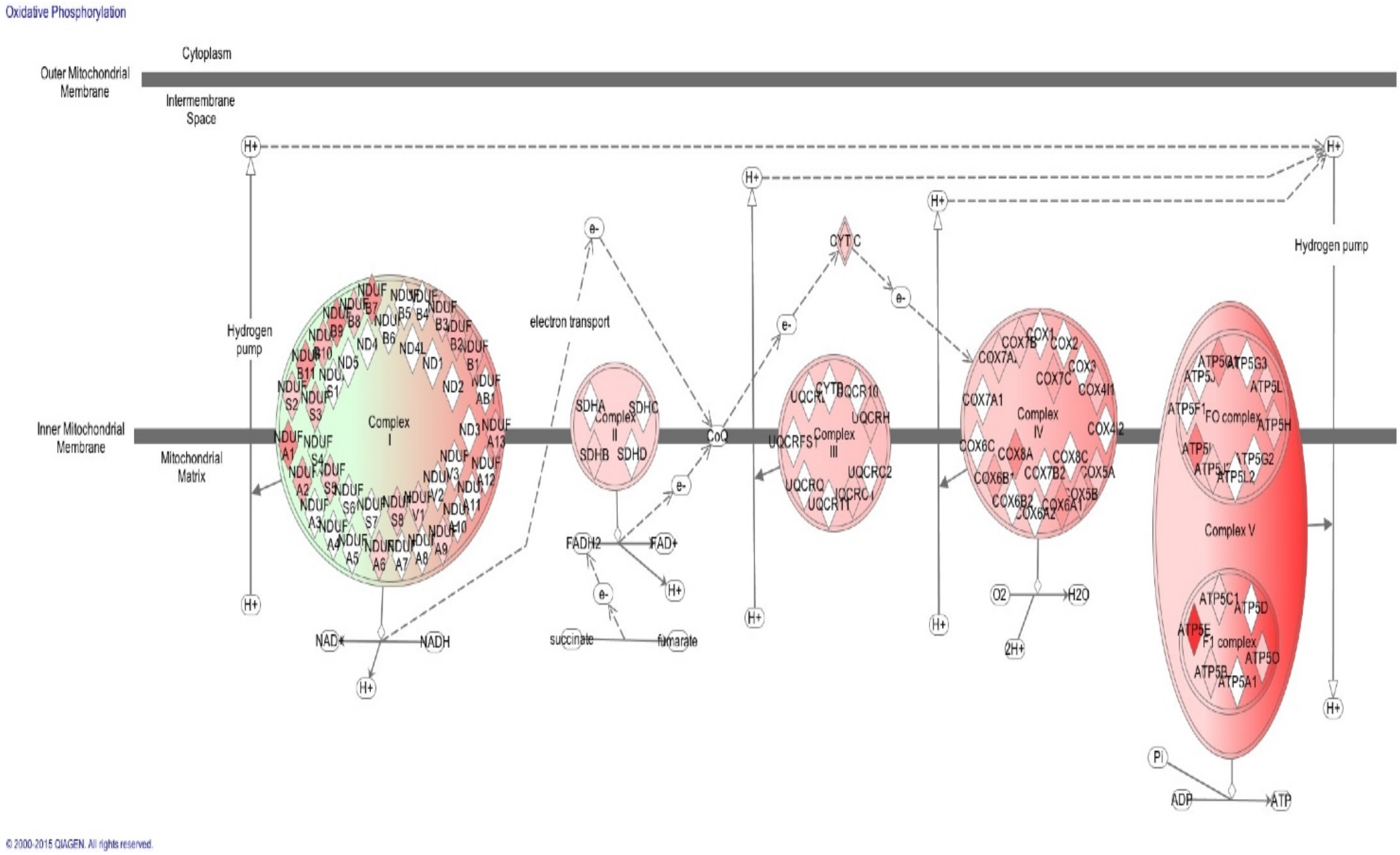
Oxidative Phosphorylation
Cytoplasm
Outer Mitochondrial Membrane
Intermembrane Space
Inner Mitochondrial Membrane
Mitochondrial Matrix
Hydrogen pump
H+
e-
electron transport
CYT C
CoQ
Complex I
Complex II
Complex III
Complex IV
Complex V
FO complex
F1 complex
SDHA
SDHB
SDHC
SDHD
FADH2
FAD+
succinate
fumarate
NAD+
NADH
O2
H2O
2H+
Pi
ADP
ATP
ND1
ND2
ND3
ND4
ND4L
ND5
UQCRFS1
UQCRQ
UQCR10
UQCRH
UQCRC2
UQCRC1
UQCR11
CYTB
COX1
COX2
COX3
COX4I1
COX4I2
COX5A
COX5B
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ATP5H
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ATP5E
ATP5O
ATP5B
ATP5A1
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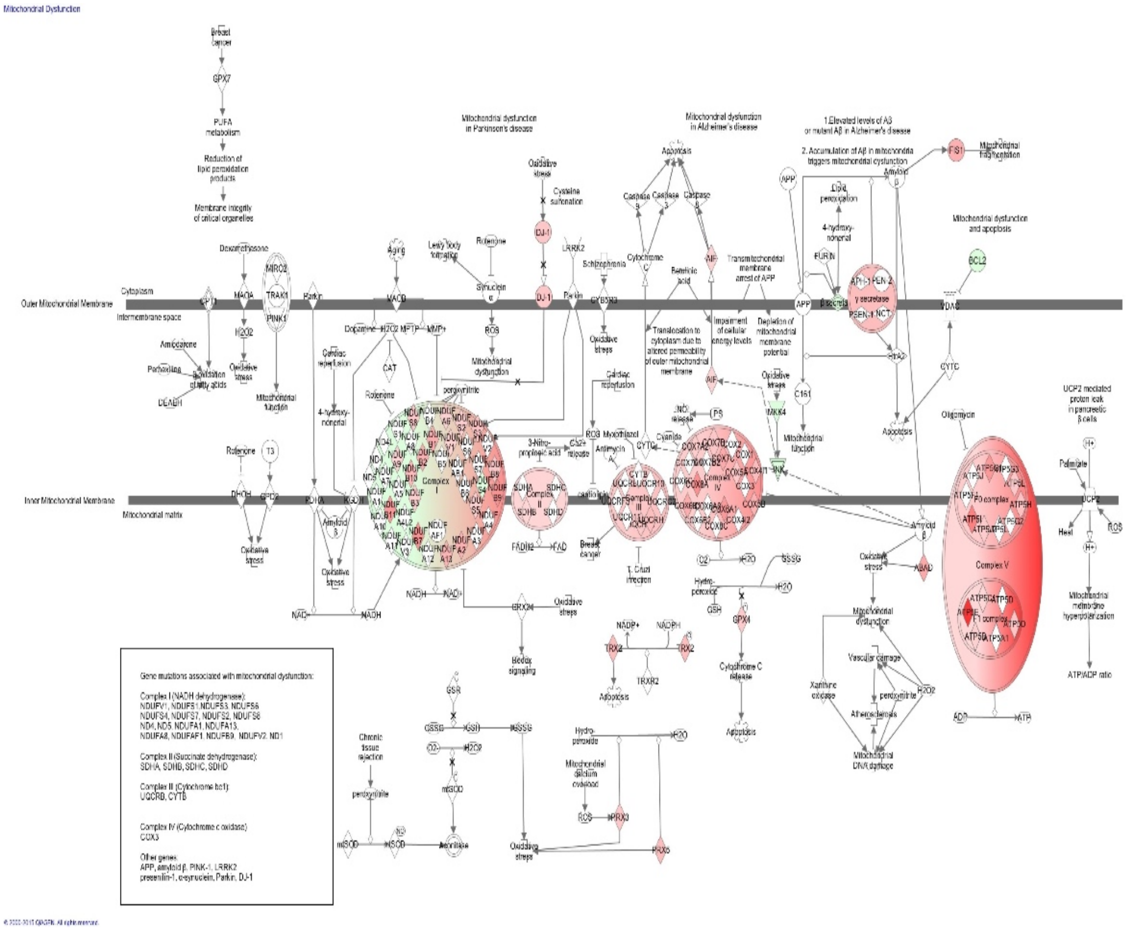
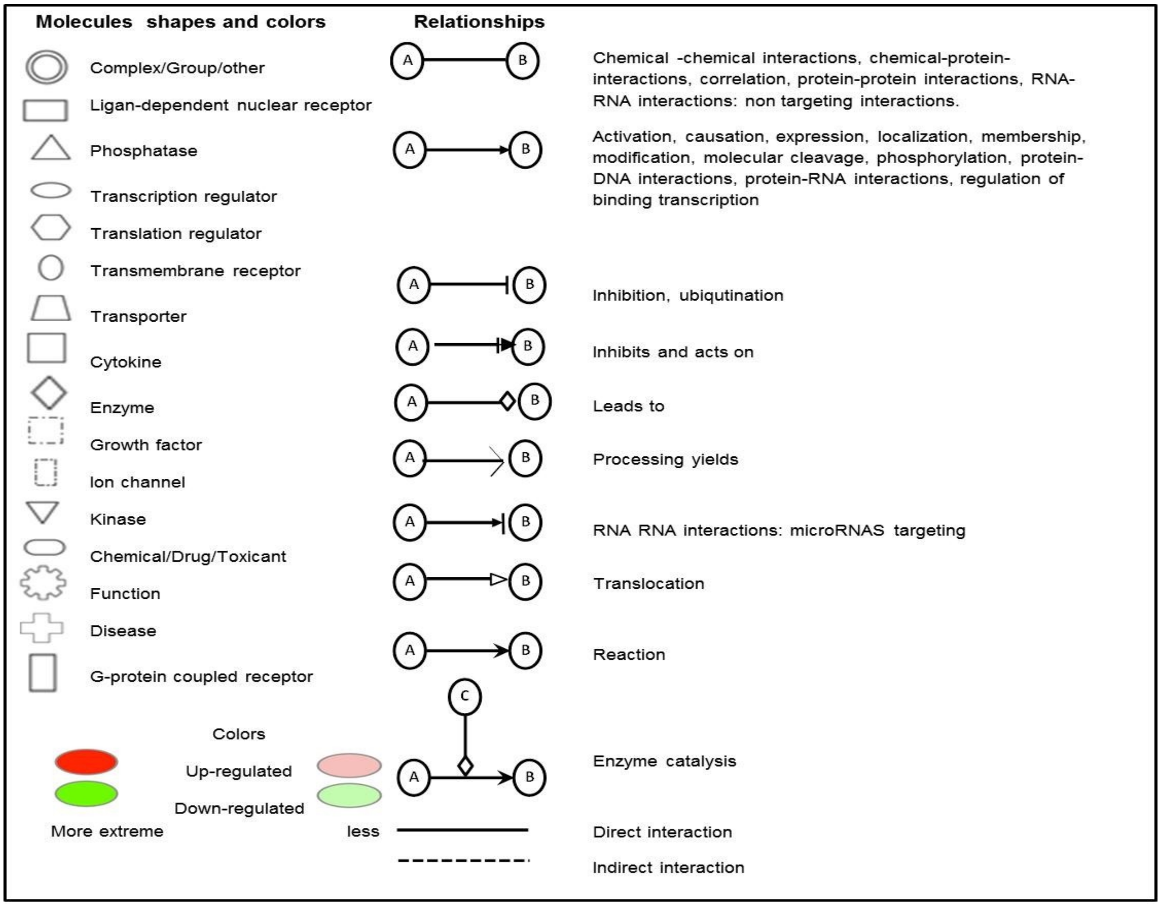
E



Supplementary Figure S2. The top five dysregulated canonical pathways (IPA). (A) EIF2 Signaling, (B) Regulation of elF4 and p70S6K Signaling, (C) mTOR Signaling, (D) Oxidative Phosphorylation and (E) Mitochondrial Dysfunction. *Relationships, molecules shapes and genes with synonyms or group genes in pathways see chart following*

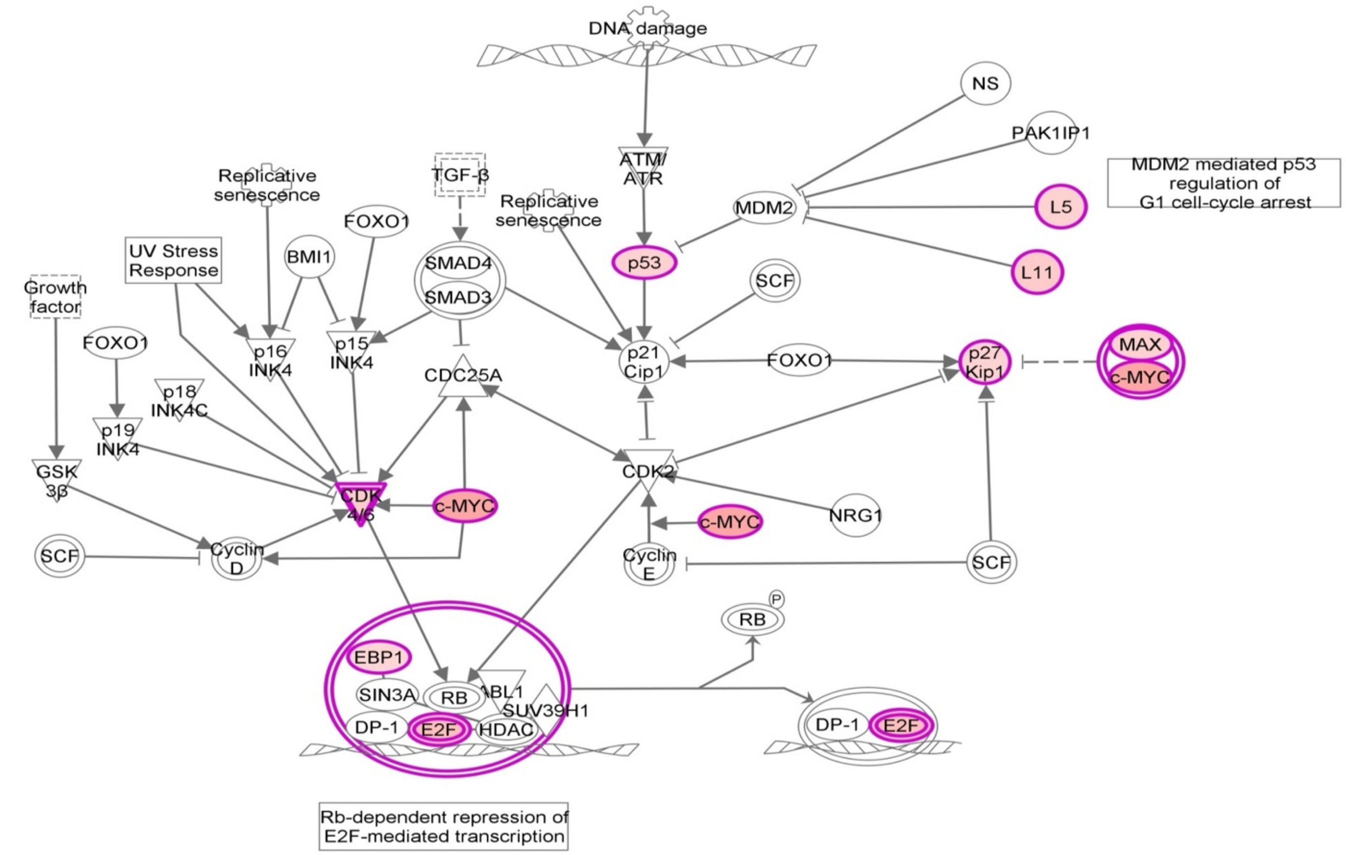

Molecules shapes and colors
Complex/Group/other
Ligan-dependent nuclear receptor
Phosphatase
Transcription regulator
Translation regulator
Transmembrane receptor
Transporter
Cytokine
Enzyme
Growth factor
Ion channel
Kinase
Chemical/Drug/Toxicant
Function
Disease
G-protein coupled receptor
Colors
Up-regulated
Down-regulated
More extreme
less
Relationships
A
B
C
Chemical -chemical interactions, chemical-protein-interactions, correlation, protein-protein interactions, RNA-RNA interactions: non targeting interactions.
Activation, causation, expression, localization, membership, modification, molecular cleavage, phosphorylation, protein-DNA interactions, protein-RNA interactions, regulation of binding transcription
Inhibition, ubiqutination
Inhibits and acts on
Leads to
Processing yields
RNA RNA interactions: microRNAS targeting
Translocation
Reaction
Enzyme catalysis
Direct interaction
Indirect interaction

Genes with synonyms or group genes in pathways IPA

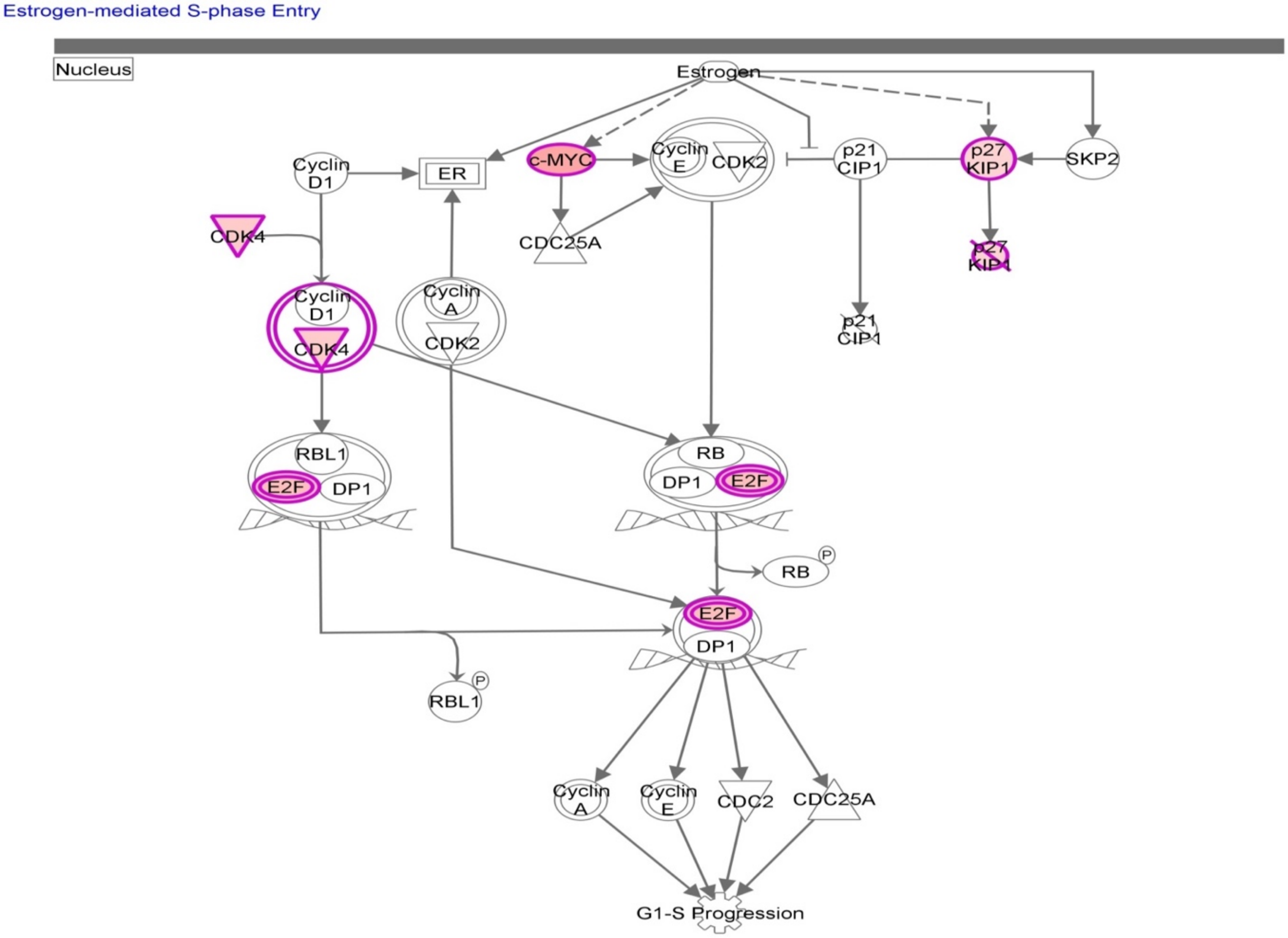
Name gen	Name gen image pathways IPA
All RPS	40S ribosomal subunit
All RPL	60S ribosomal subunit
APH1A	γ-secretase
BACE2	β-secretase
BMPR2	GFR
CDKN1B	p27Kip1
CYC1	CYT C or complex III
DDIT4	REDD1
EIF1	A121
E2F4	E2F
EIF1, EIF2B2 and EIF2B5	eIF2
EIF2AK1	HRI
EIF2B2 and EIF2B5	eIF2β
EIF3B, EIF3C, EIF3D, EIF3G, EIF3I, EIF3K and EIF3L	eIF3
EIF4A1, EIF4A2 and EIF4A3	eIF4A
EIF4EBP1	4EBP or 4E-BP1 or eIF4BP
EIF4G1	eIF4γ and eIF4G
FAU	RPS30 of 40S ribosomal subunit
INPPL1	SHIP
ITGA2	Integrin
ITGB6 and ITGB5	β-Integrin
LMNA	Lamin A
MAP2K4	MKK4 or JNK
MCRS1	MSP58
PA2G4	EBP1
PARK7	DJ-1
PIK3CA	PI3K
PPP2R4 and PPP2R1A	PP2A
RND3	RHO
RPS6KA3	RSK
TXN2	TRX2

A

Cell Cycle: G1/S Checkpoint Regulation

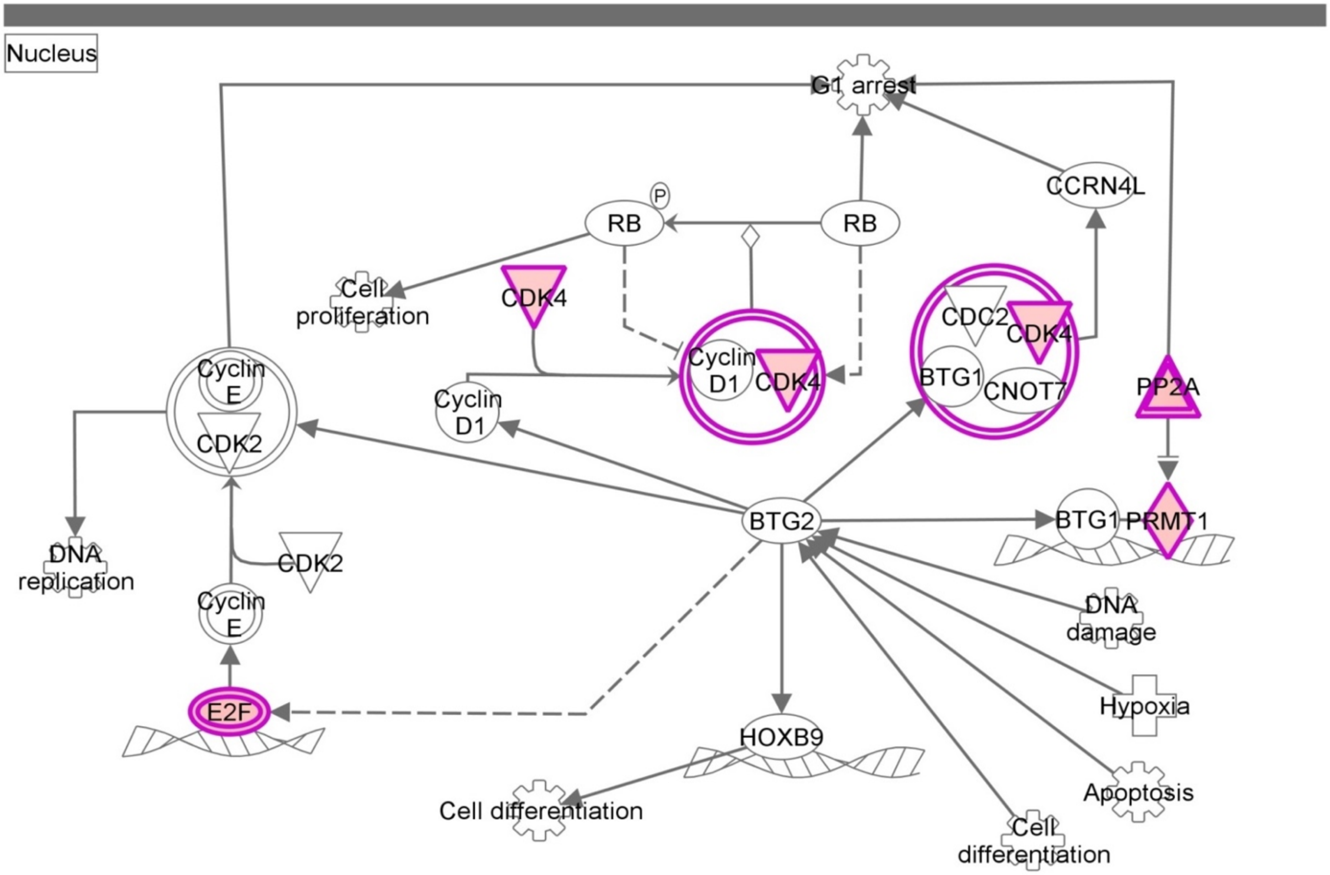


B


Estrogen-mediated S-phase Entry
Nucleus
Estrogen
Cyclin D1
ER
c-MYC
Cyclin E
CDK2
p21 CIP1
p27 KIP1
SKP2
CDK4
CDC25A
p27 KIP1
Cyclin D1
CDK4
Cyclin A
CDK2
p21 CIP1
RBL1
E2F
DP1
RB
DP1
E2F
P
RB
E2F
DP1
P
RBL1
Cyclin A
Cyclin E
CDC2
CDC25A
G1-S Progression

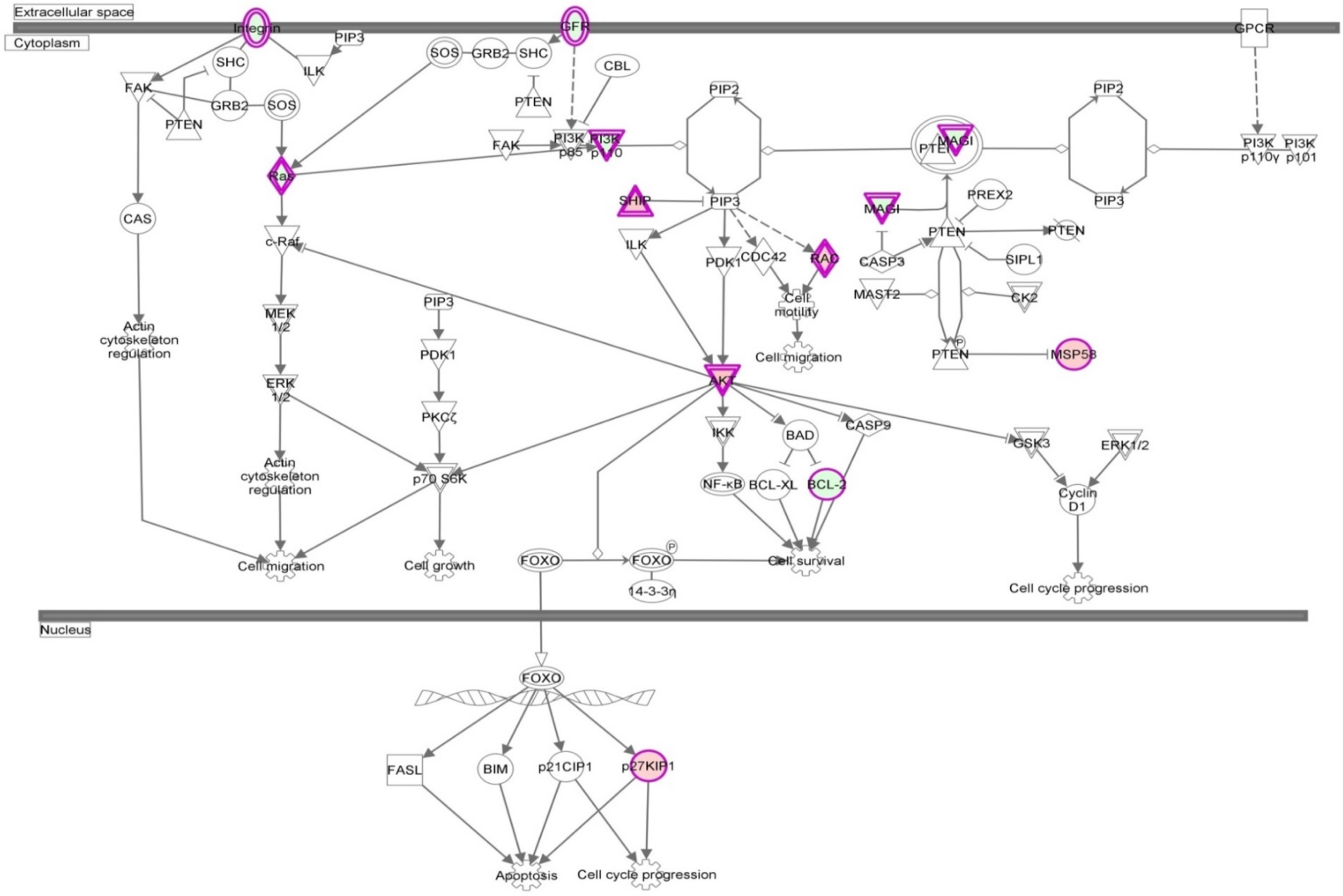
C

Cell Cycle Regulation by BTG Family Proteins



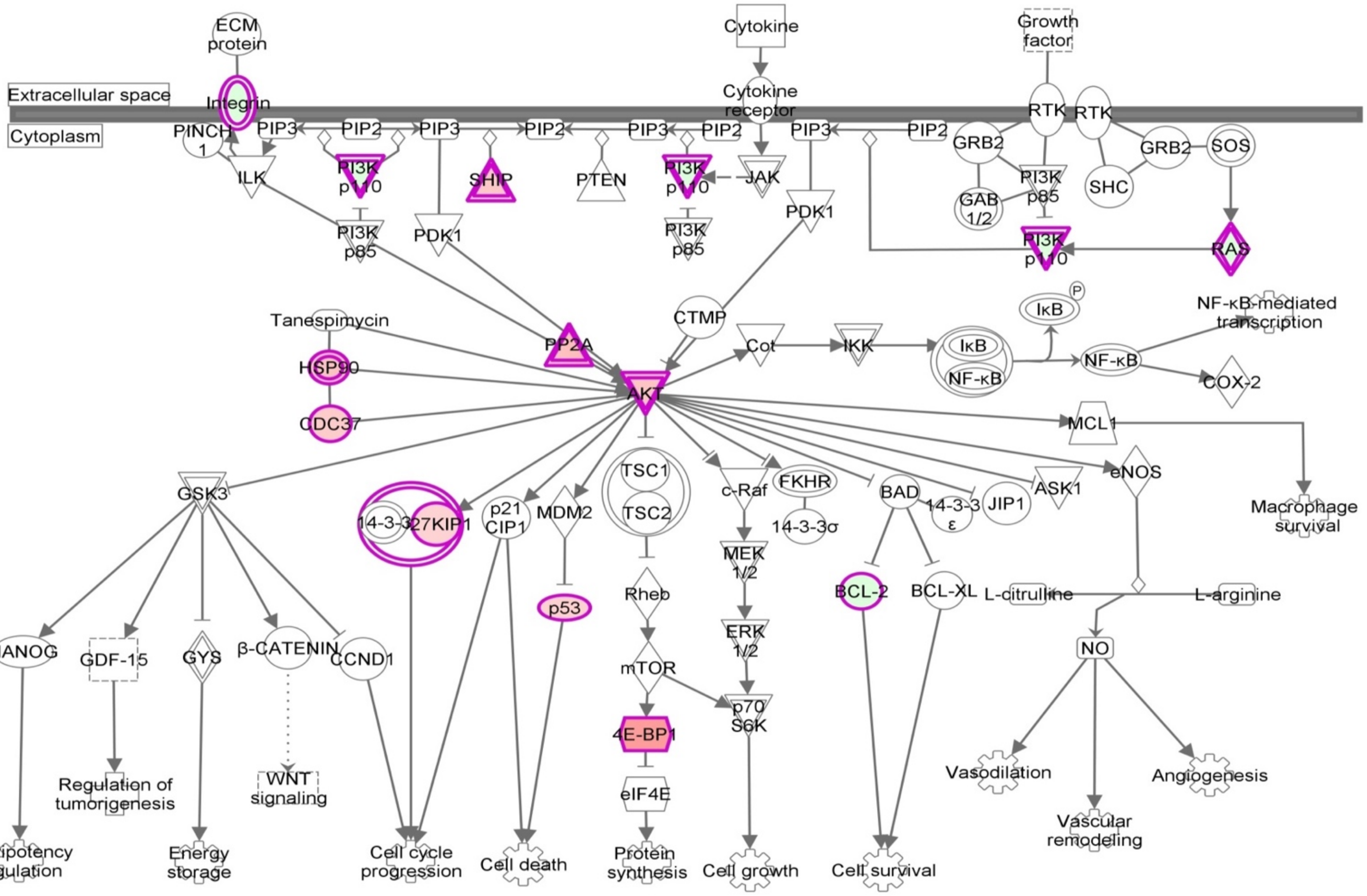
D

PTEN Signaling

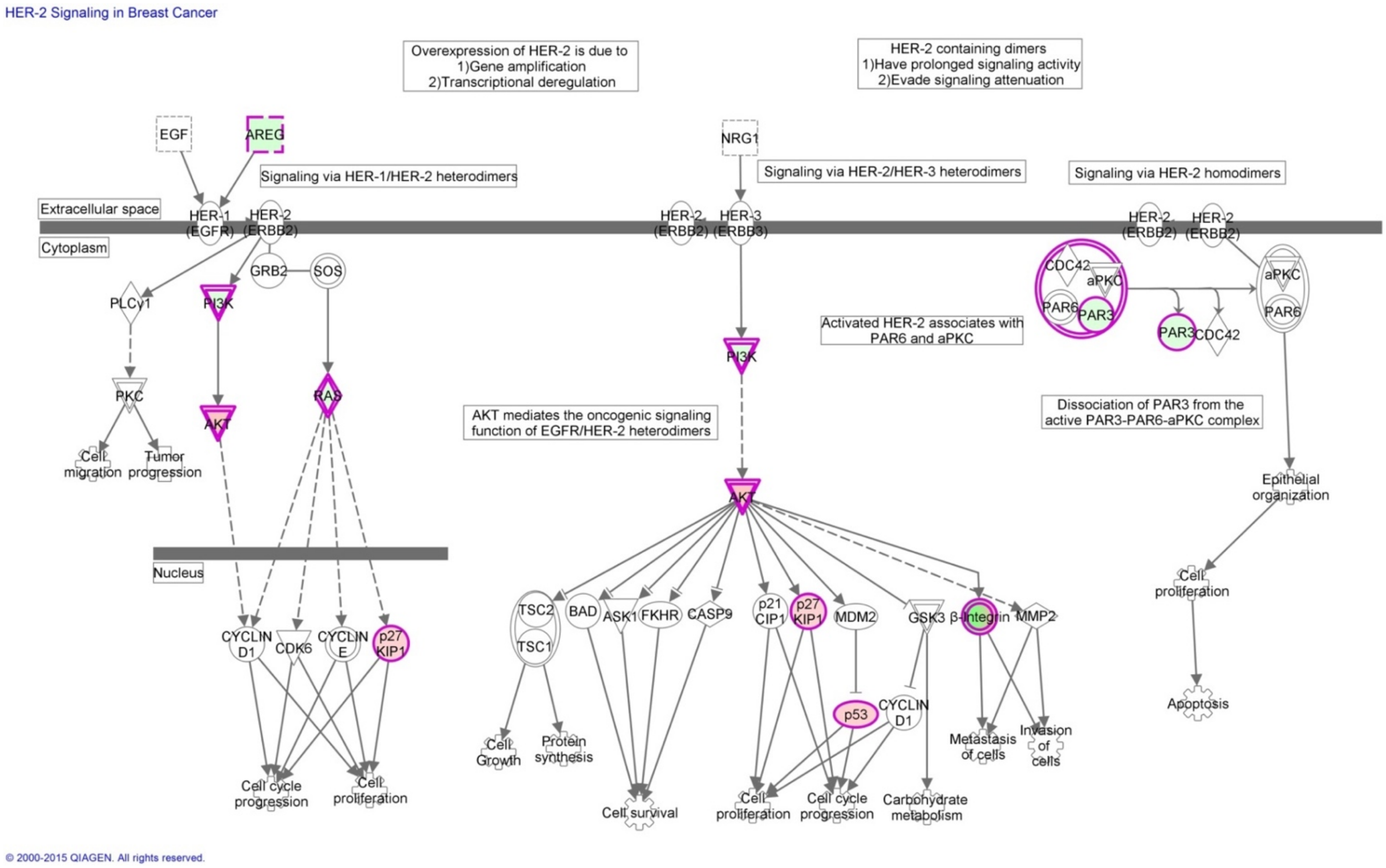


E

PI3K/AKT Signaling



F



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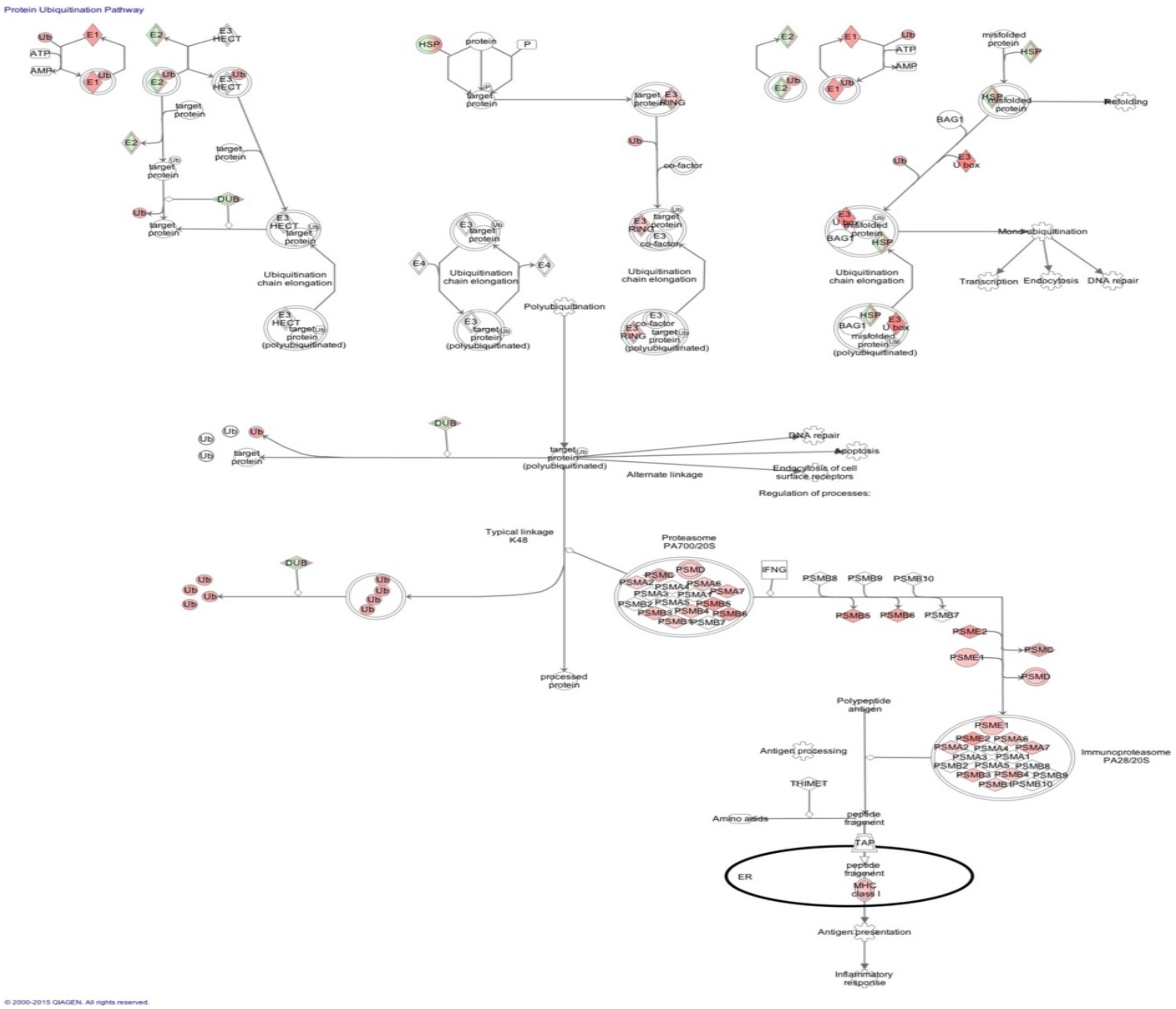
Supplementary Figure S3. Some interesting pathways associated with the cell cycle (IPA). (A) cell cycle G1/S checkpoint regulation, (B) Estrogen-mediated S-phase entry, (C) cell cycle regulation by BTG family protein, (D) PTEN signaling, (E) PI3K/AKT signaling and (F) HER-2 signaling in breast cancer

A


Protein Ubiquitination Pathway
Ub
E1
ATP
AMP
E1 Ub
E2
E3 HECT
E2 Ub
E3 HECT Ub
target protein
E2
target protein
target protein Ub
DUB
Ub
target protein
E3 HECT target protein Ub
Ubiquitination chain elongation
E3 HECT target protein (polyubiquitinated) Ub
HSP
protein
P
target protein Ub
target protein E3 RING
Ub
co-factor
E3 RING target protein Ub E3 co-factor
Ubiquitination chain elongation
E3 RING E3 co-factor target protein (polyubiquitinated) Ub
E4
E3 target protein Ub
Ubiquitination chain elongation
E4
E3 target protein (polyubiquitinated) Ub
Polyubiquitination
E2
E2 Ub
E1
Ub
ATP
AMP
E1 Ub
misfolded protein
HSP
HSP misfolded protein
Refolding
BAG1
Ub
E3 U box
E3 U box BAG1 misfolded protein HSP Ub
Mono-ubiquitination
Ubiquitination chain elongation
Transcription
Endocytosis
DNA repair
HSP BAG1 E3 U box misfolded protein (polyubiquitinated) Ub
Ub
Ub
Ub
Ub
target protein
DUB
target protein (polyubiquitinated) Ub
Alternate linkage
DNA repair
Apoptosis
Endocytosis of cell surface receptors
Regulation of processes:
Typical linkage K48
Proteasome PA700/20S
DUB
Ub
Ub
Ub
Ub
Ub
Ub
Ub
PSMC PSMD PSMA2 PSMA6 PSMA4 PSMA7 PSMA3 PSMA1 PSMB2 PSMA5 PSMB5 PSMB3 PSMB4 PSMB6 PSMB1 PSMB7
IFNG
PSMB8
PSMB9
PSMB10
PSMB5
PSMB6
PSMB7
PSME2
PSME1
PSMC
PSMD
processed protein
Polypeptide antigen
Antigen processing
PSME1 PSME2 PSMA6 PSMA2 PSMA4 PSMA7 PSMA3 PSMA1 PSMB2 PSMA5 PSMB8 PSMB3 PSMB4 PSMB9 PSMB1 PSMB10
Immunoproteasome PA28/20S
THIMET
Amino acids
peptide fragment
TAP
ER
peptide fragment
MHC class I
Antigen presentation
Inflammatory response
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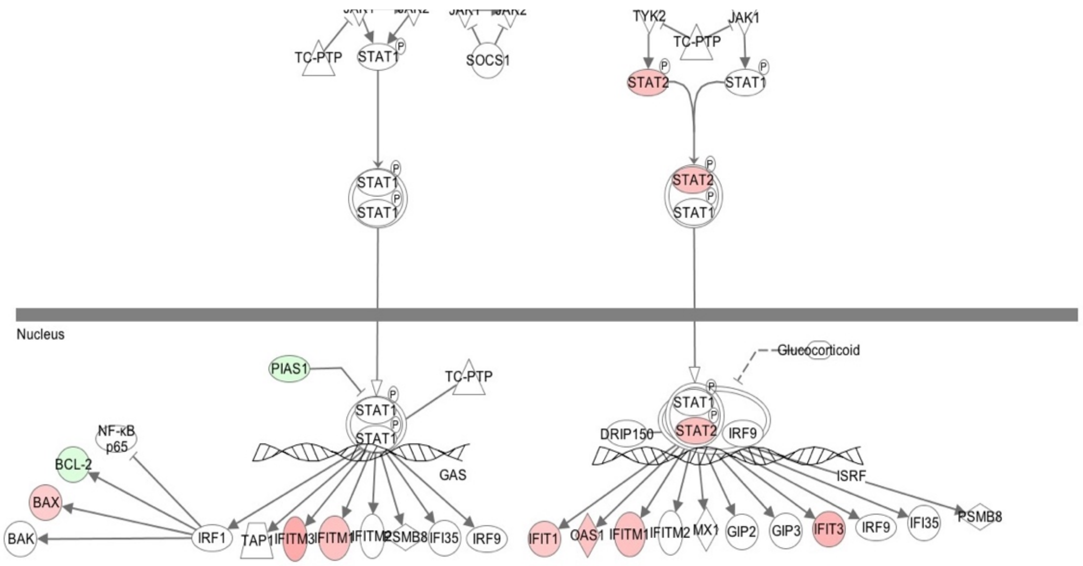
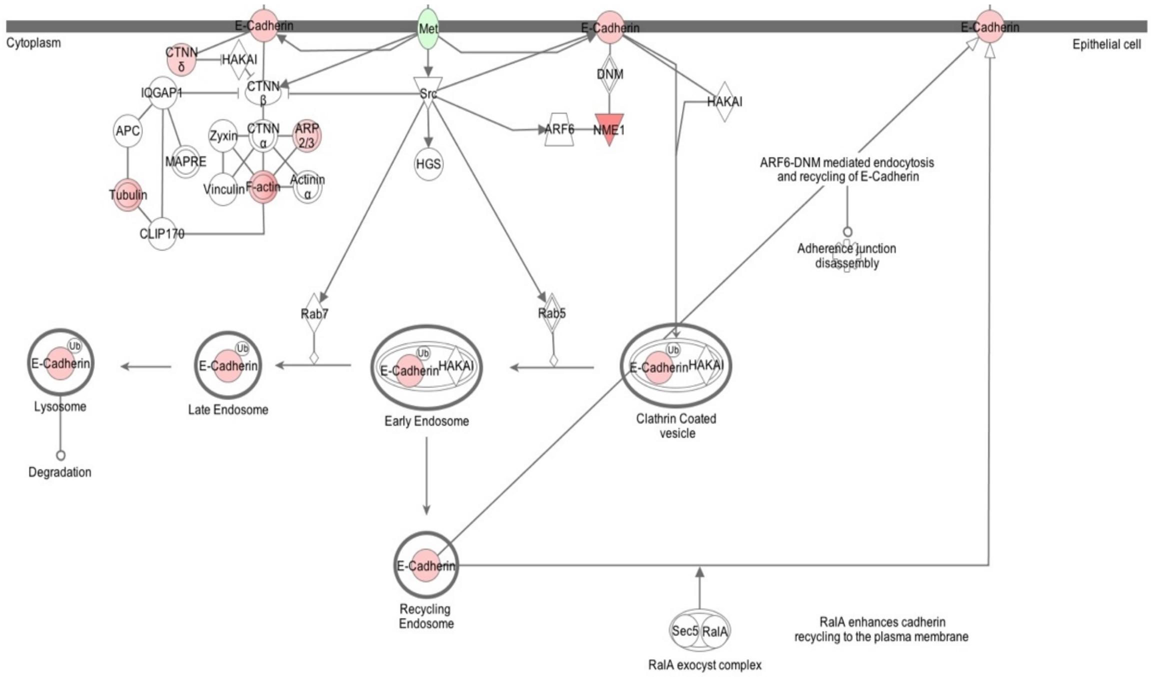
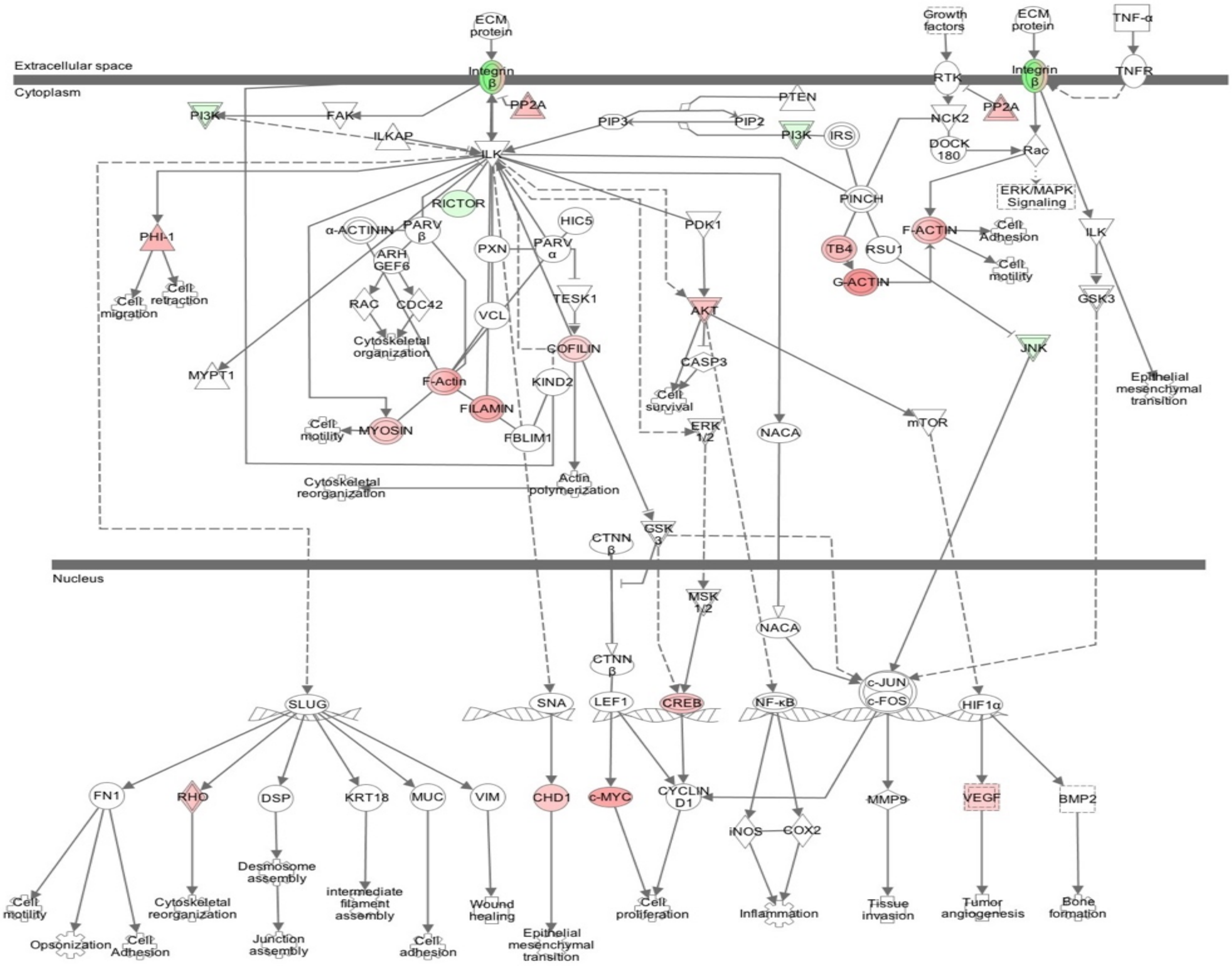
B


TC-PTP
STAT1
P
SOCS1
TYK2
TC-PTP
JAK1
STAT2
STAT1
STAT1
STAT1
STAT2
STAT1
Nucleus
PIAS1
TC-PTP
STAT1
STAT1
GAS
NF-κB
p65
BCL-2
BAX
BAK
IRF1
TAP1
IFITM3
IFITM1
IFITM2
PSMB8
IFI35
IRF9
Glucocorticoid
STAT1
DRIP150
STAT2
IRF9
ISRF
IFIT1
OAS1
IFITM1
IFITM2
MX1
GIP2
GIP3
IFIT3
IRF9
IFI35
PSMB8

C


Cytoplasm
E-Cadherin
Met
E-Cadherin
E-Cadherin
Epithelial cell
CTNN δ
HAKAI
CTNN β
IQGAP1
Src
DNM
HAKAI
APC
Zyxin
CTNN α
ARP 2/3
ARF6
NME1
MAPRE
HGS
Vinculin
F-actin
Actinin α
Tubulin
CLIP170
ARF6-DNM mediated endocytosis and recycling of E-Cadherin
Adherence junction disassembly
Rab7
Rab5
Ub
E-Cadherin
Lysosome
Degradation
Ub
E-Cadherin
Late Endosome
Ub
E-Cadherin
HAKAI
Early Endosome
Ub
E-Cadherin
HAKAI
Clathrin Coated vesicle
E-Cadherin
Recycling Endosome
Sec5
RalA
RalA exocyst complex
RalA enhances cadherin recycling to the plasma membrane

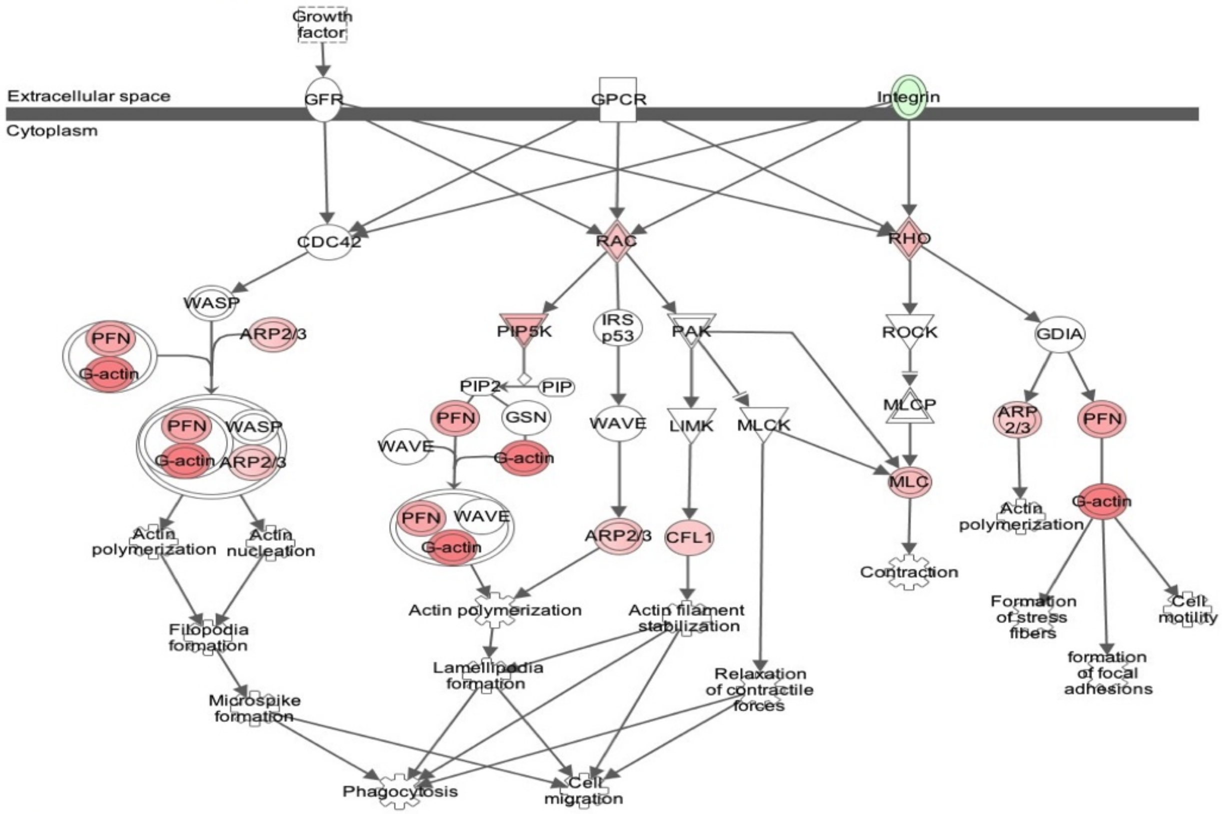
D



ILK Signaling

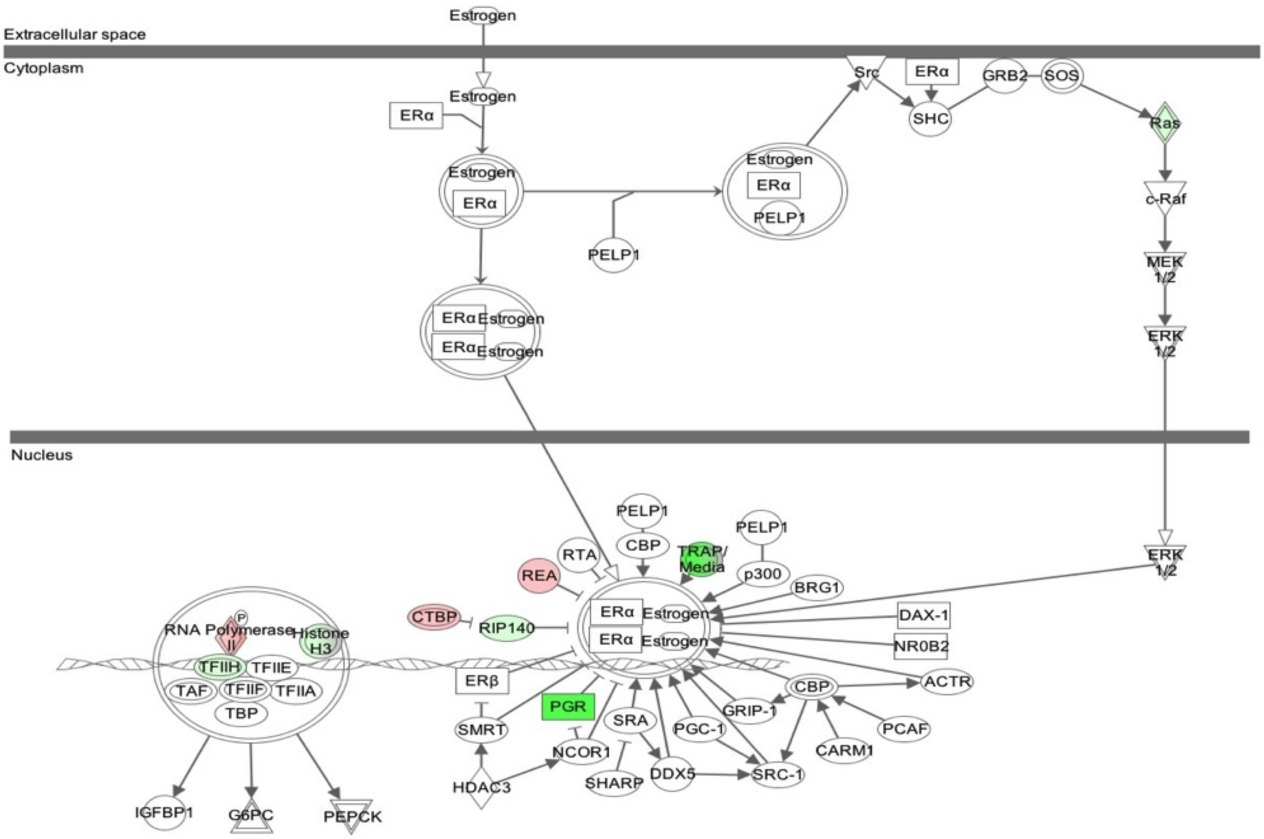
E

Regulation of Actin-based Motility by Rho



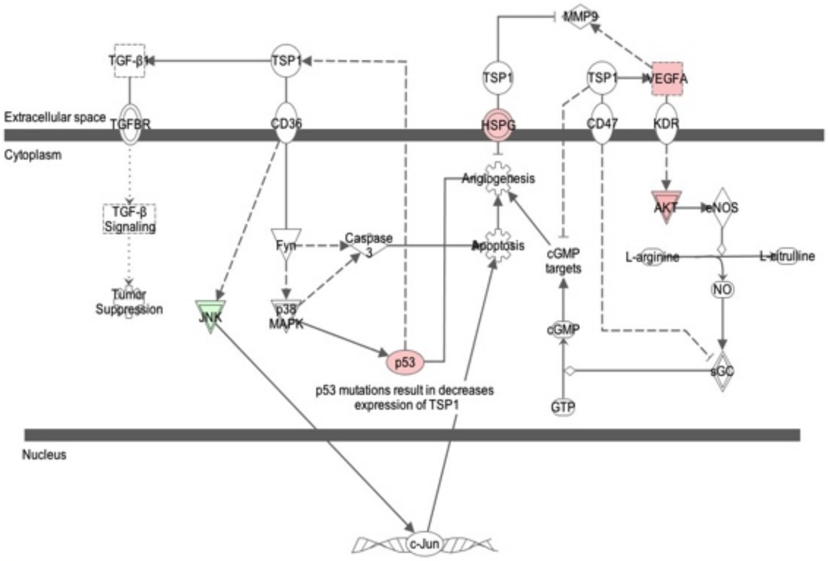
F

Estrogen Receptor Signaling

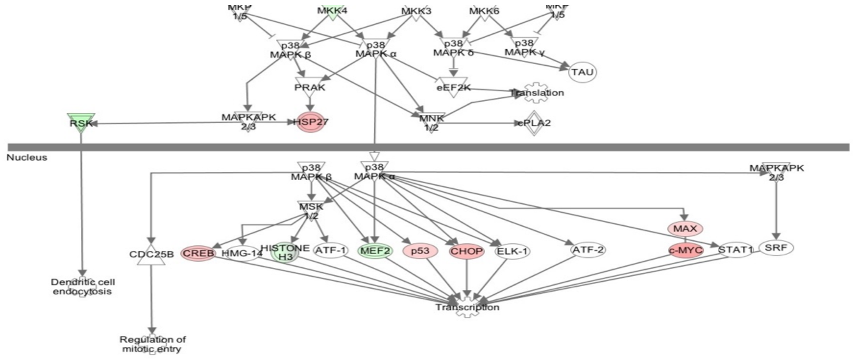


G

Inhibition of Angiogenesis by TSP1

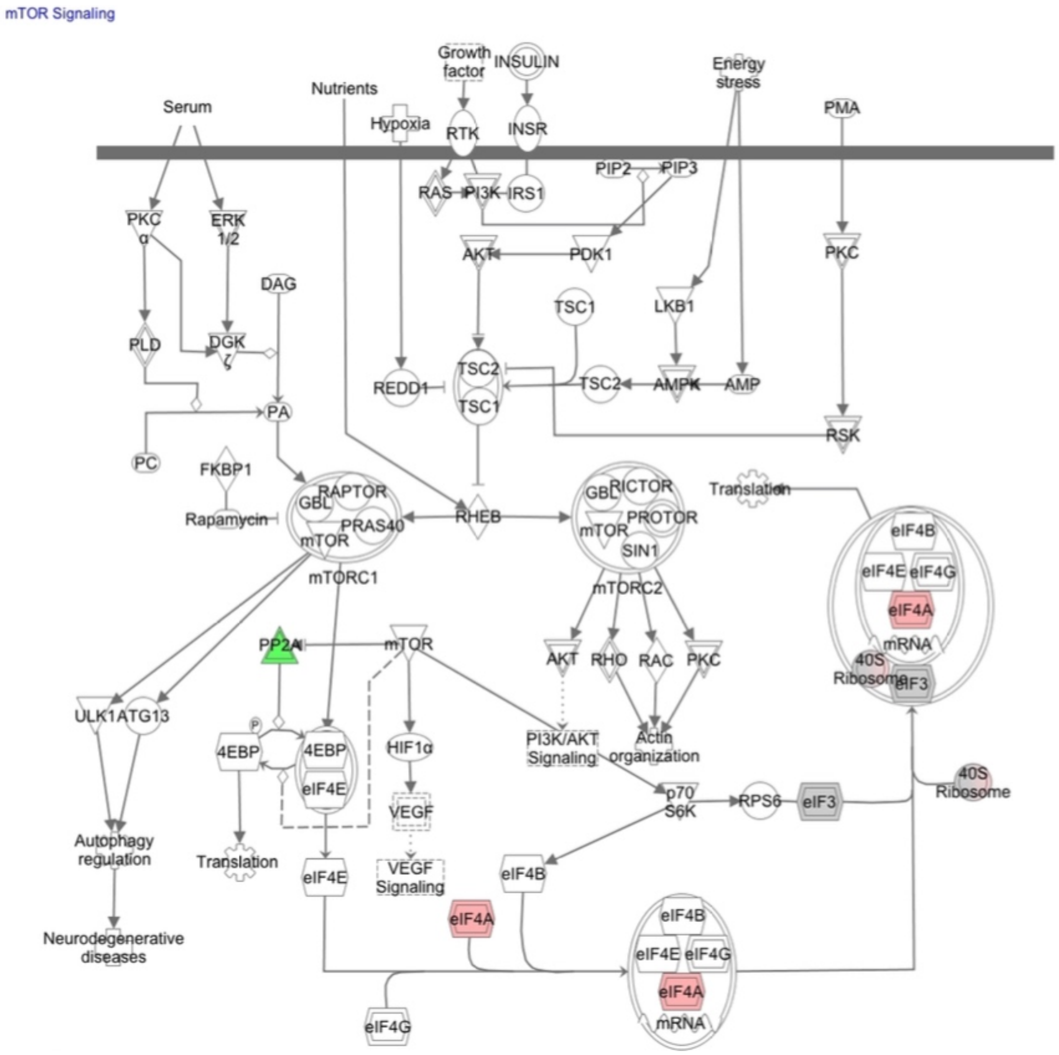


H

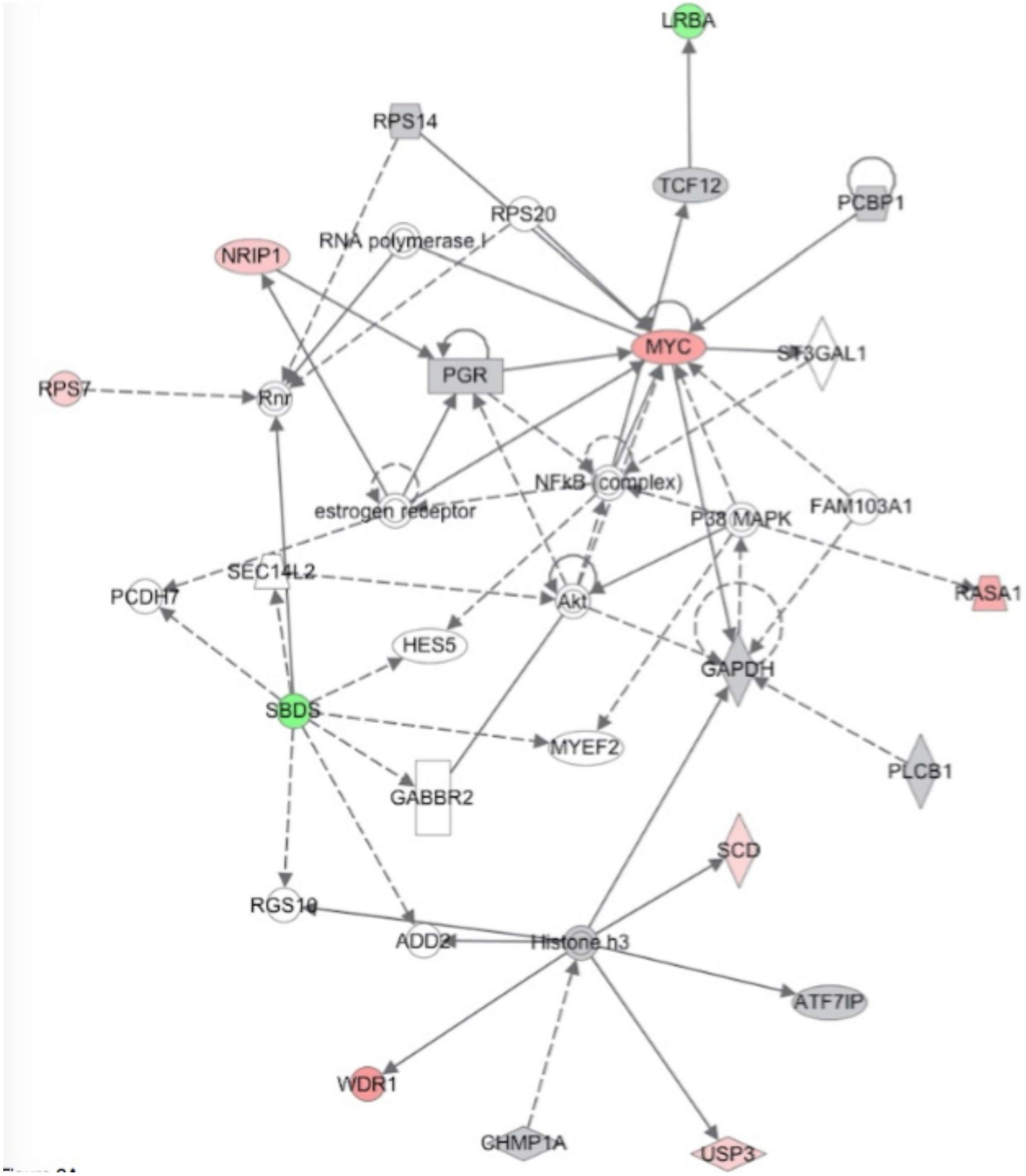


Supplementary Figure S4. Some canonical pathways associated with other cellular processes are also; (A) protein ubiquitination, (B) Interferon signaling, (C) Remodeling of epithelial adherents junctions, (D) ILK signaling, (E) Integrin signaling, (F)) Estrogen receptor signaling, (G)Inhibition of angiogenesis by TSP1 and (H) p38 MAPK signaling.

A


mTOR Signaling

B


LRBA
RPS14
TCF12
PCBP1
RPS20
RNA polymerase I
NRIP1
MYC
ST3GAL1
RPS7
Rnr
PGR
NFkB (complex)
estrogen receptor
FAM103A1
P38 MAPK
RASA1
SEC14L2
PCDH7
Akt
HES5
GAPDH
SBDS
MYEF2
PLCB1
GABBR2
SCD
RGS10
ADD2
Histone h3
ATF7IP
WDR1
CHMP1A
USP3

C

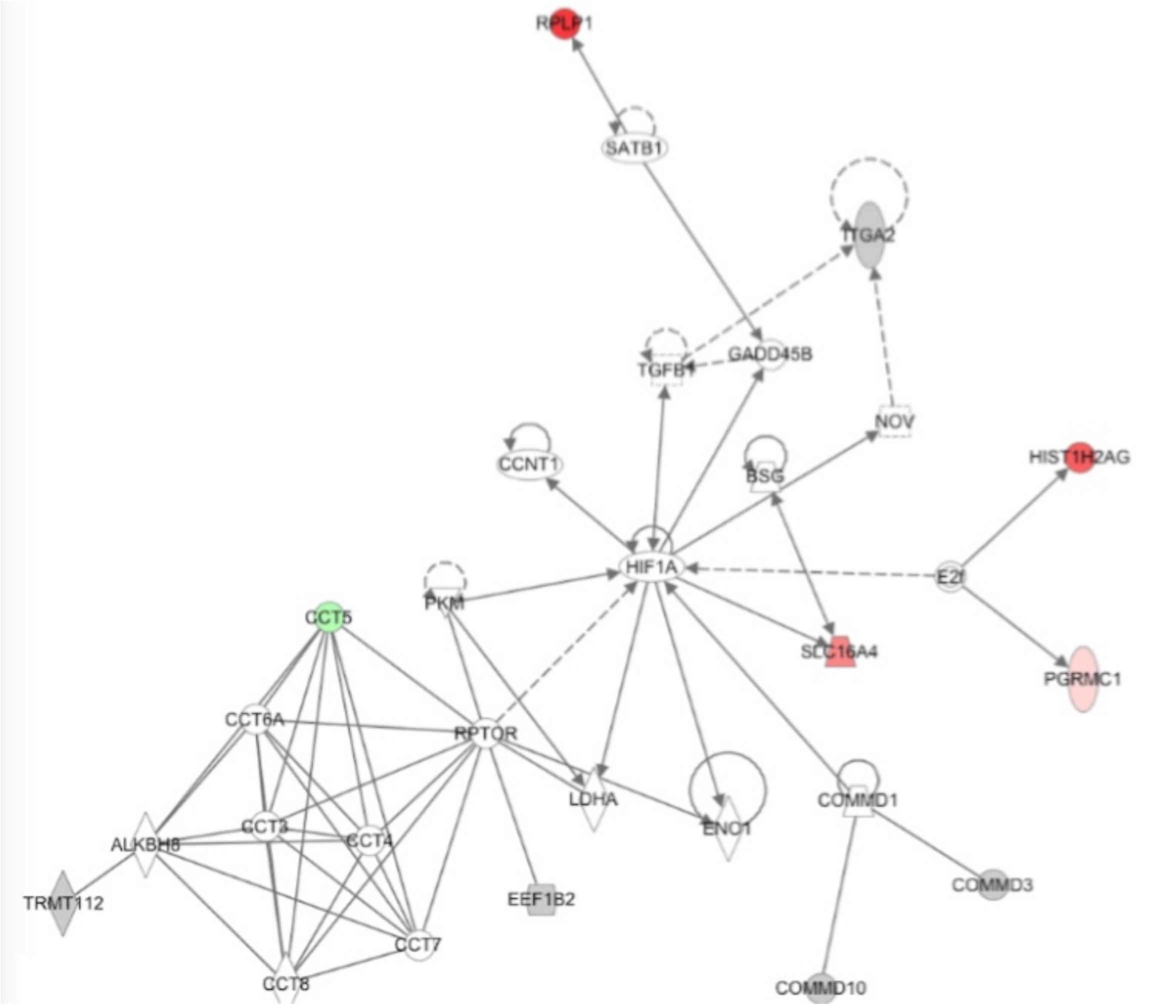
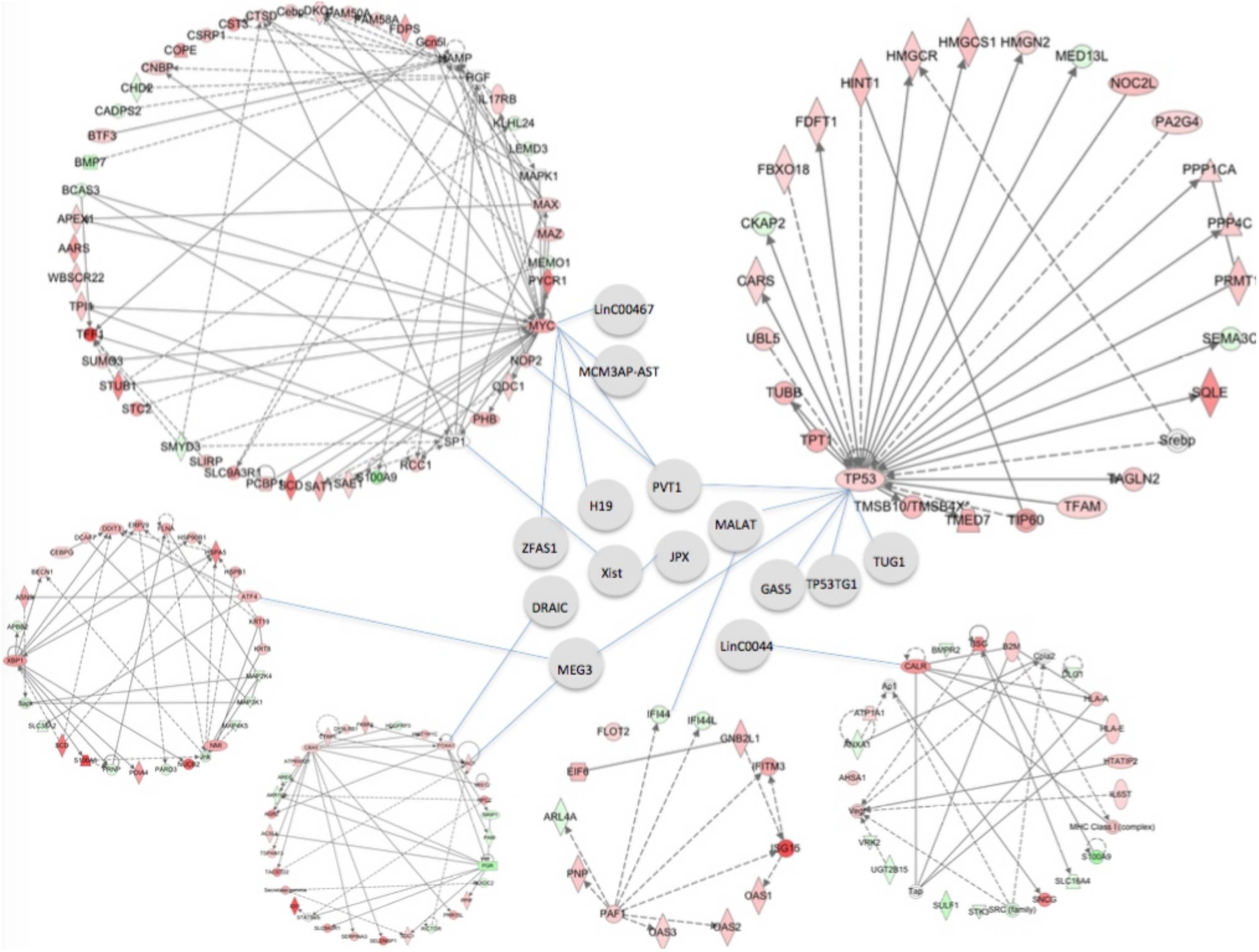


Fig S5. Dysregulated pathways analysis using 63 lncRNA that present his neighbor RNAm dysregulated. A) mTOR network. B) MYC Network and C) HIF network.



Supplementary Figure S6. Correlation between mRNA/lncRNAs networks. Green and red circles represent the mRNAs and gray circles the lncRNAs .