The use of network analyses for elucidating mechanisms in cardiovascular disease

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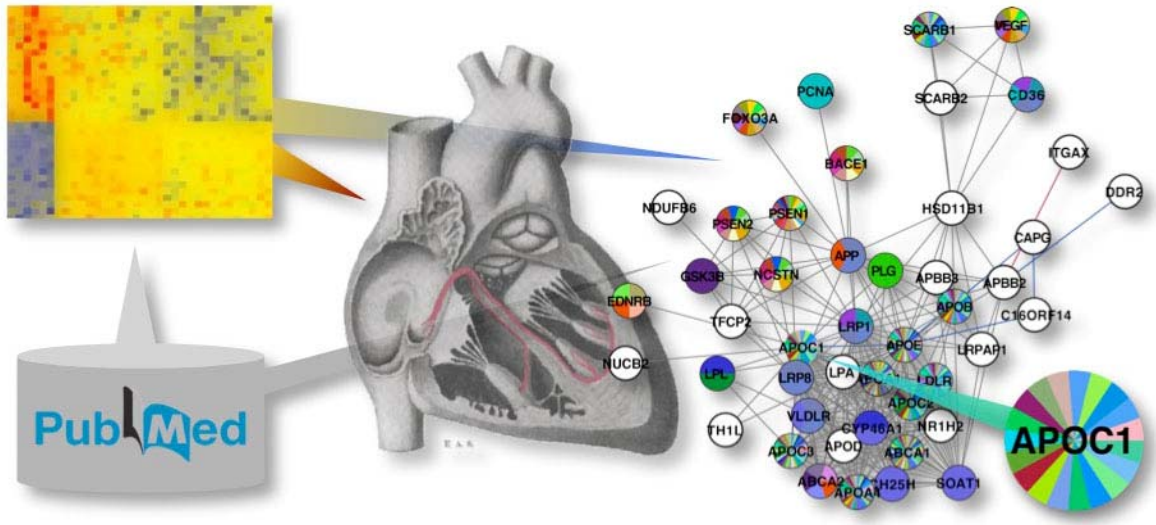
This paper provides a basic introduction to networks and then examines the use of network analysis to investigate mechanisms in cardiovascular disease by combining results from transcriptomics and literature mining.



Supplemental information

This supplementary information section provides extended information on a number of components of the manuscript. Specifically, Figure S1 displays the results of the multivariate statistical analysis (OPLS). The list of 43 differentially expressed (DE) genes and probes are provided in Tables S1 and S2, respectively (complete with hyperlinks). The results of the manual curation of the *AgilentLiteratureSearch* plugin for APOC1 are presented in Table S3 and Figure S2 and the individual papers of interest are discussed (hyperlinks are provided to the PubMed citations). Figure S3 displays the network of gene ontology (GO) terms obtained from the analysis of the sub-network shown in Figure 7 with *ClueGO*, using all terms in GO Biological Process. Finally, Figure S4 shows the coloring of the entire sub-network from Figure 7 following analysis with *ClueGO* using all evidence codes and the Benjamini and Hochberg correction.

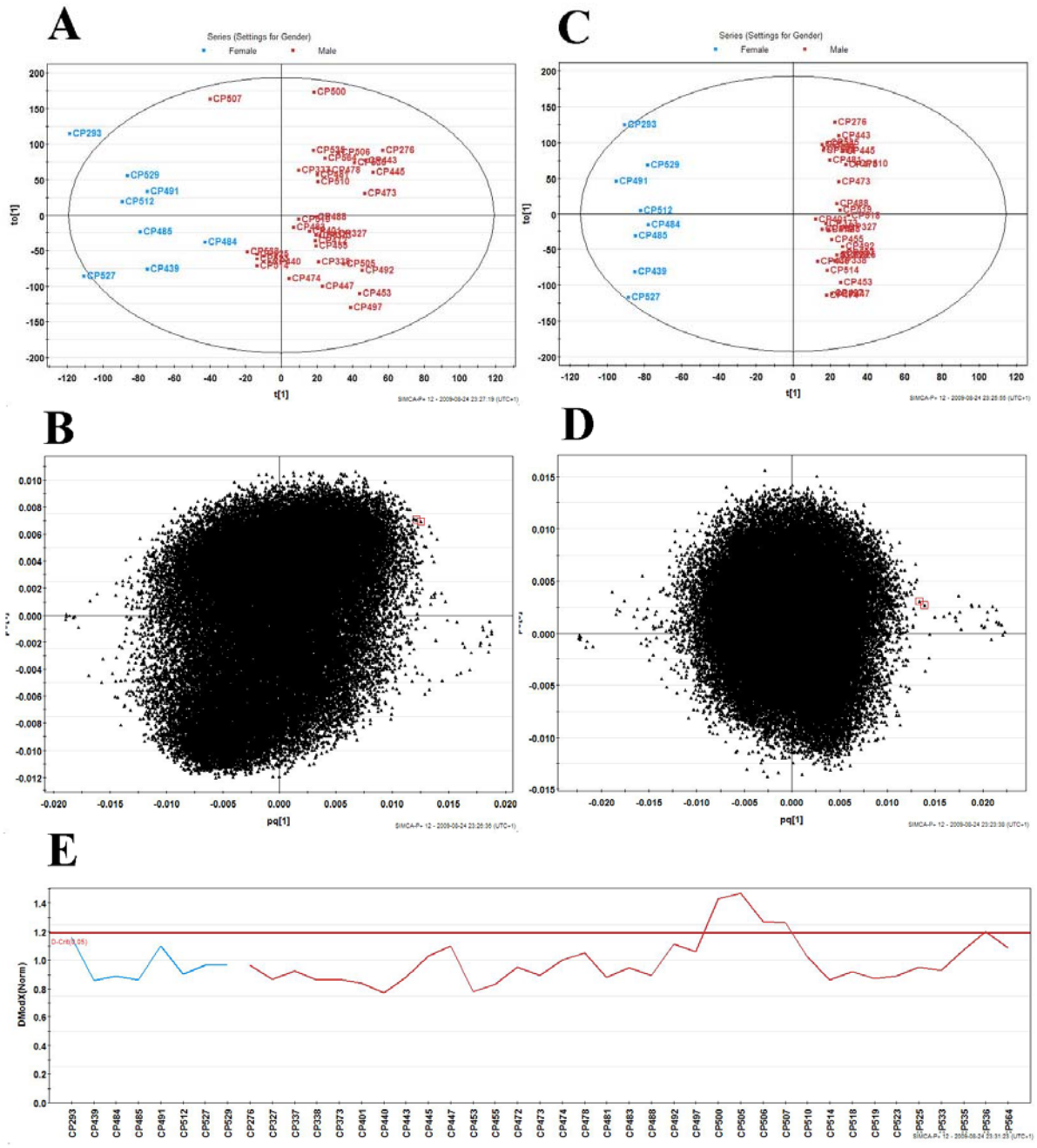


Figure S1. Orthogonal partial least squares of latent structures (OPLS) analysis including all 49 observations in the X block with "gender" as the sole Y-component resulted in a reasonably good model for the current data set (R2Y(cum) = 0.78), but with poor predictive power (Q2=0.008). The scores plot and loadings plots are displayed in panels **A** and **B**, respectively. The corresponding DModX (distance to model X) plot displayed in panel **E** indicates that four of the male subjects (CP500-CP507) are poorly described by the model. The same four subjects were identified as weak outliers in the quality control preceding the limma analysis due to the high level of noise present in these four data sets. To evaluate their influence on the OPLS model, a second OPLS analysis excluding named subjects (CP500, CP505, CP506 and CP507) was performed (panels **C**-**D**). The result was a drastic improvement in the separation between groups (panel **C**; (R2Y(cum) = 0.99), as well as a slight improvement in the predictive power of the model (Q2=0.12). The orthogonal variance

in the X block (i.e., the variation in the data set which is not relevant to gender) could be fitted to three significant orthogonal vectors, resulting in a good separation of the male and female group as visualized in the scores plot (panel **C**). Furthermore in the all inclusive model, the variation in expression levels of the two APOC1 probe sets (marked with red boxes in the loading plots) contributed equally to the gender-related (X-axis) and the gender-unrelated (Y-axis) variation, as evidenced by their location in the upper right corner of the loadings plot (panel **B**). Following removal of the four weak outliers, the orthogonal, gender-unrelated noise was removed, as evidenced by the proximity of the two APOC1 probes to the X-axis in the loadings plot (panel **D**). As such, it appears that the quality issues associated with these four microarray data sets have a confounding effect on the importance of the APOC1 gene in the observed gender differences in gene expression of atherosclerotic plaques.

Table S1. List of 43 differentially expressed (DE) genes

Entrez Gene	Symbol	Description	Chr[a]	P_T[b]	P_{DE}[c]	M (log2 FC)[d]	Avg. Expr.[e]
6192	RPS4Y1	ribosomal protein S4, Y-linked 1	Y	1	1	5.49	9.1
8284	JARID1D	jumonji, AT rich interactive domain 1D	Y	1	1	3.99	6.4
8653	DDX3Y	DEAD (Asp-Glu-Ala-Asp) box polypeptide 3, Y-linked	Y	4	2	3.72	6.2
9086	EIF1AY	eukaryotic translation initiation factor 1A, Y-linked	Y	2	2	3.07	5.2
8287	USP9Y	ubiquitin specific peptidase 9, Y-linked (fat facets-like, Drosophila)	Y	2	2	2.60	4.6
84663	CYorf15B	chromosome Y open reading frame 15B	Y	3	3	2.53	5.1
246126	CYorf15A	chromosome Y open reading frame 15A	Y	2	2	1.74	4.1
7544	ZFY	zinc finger protein, Y-linked	Y	3	1	1.51	3.4
55410	CYorf14	chromosome Y open reading frame 14	Y	1	1	1.36	4.6
9956	HS3ST2	heparan sulfate (glucosamine) 3-O-sulfotransferase 2	16	1	1	1.33	8.5
84830	C6orf105	chromosome 6 open reading frame 105	6	2	1	1.22	5.9
22829	NLGN4Y	neuroligin 4, Y-linked	Y	2	1	1.12	3.6
10993	SDS	serine dehydratase	12	2	1	1.06	7.1
341	APOC1	apolipoprotein C-I	19	2	1	1.04	9.6
5616	PRKY	protein kinase, Y-linked	Y	1	1	0.87	4.4
100133941	CD24	CD24 molecule	6	6	3	0.82	5.3
2170	FABP3	fatty acid binding protein 3, muscle and heart (mammary-derived growth inhibitor)	1	2	1	0.75	6.3
93010	B3GNT7	UDP-GlcNAc:betaGal beta-1,3-N-acetylglucosaminyltransferase 7	2	4	2	0.63	6.1
64595	TTTY15	testis-specific transcript, Y-linked 15	Y	1	1	0.57	3.9
7404	UTY	ubiquitously transcribed tetratricopeptide repeat gene	Y	3	2	0.48	3.7
2491	CENPI	centromere protein I	X	4	1	0.34	3.5
59342	SCPEP1	serine carboxypeptidase 1	17	2	1	0.32	8.4
1968	EIF2S3	eukaryotic translation initiation factor 2, subunit 3 gamma, 52kDa	X	3	1	-0.37	8.4
1654	DDX3X	DEAD (Asp-Glu-Ala-Asp) box polypeptide 3, X-linked	X	6	1	-0.38	10.0
65250	C5orf42	chromosome 5 open reading frame 42	5	2	1	-0.43	2.9
6191	RPS4X	ribosomal protein S4, X-linked	X	2	2	-0.43	12.2
8233	ZRSR2	zinc finger (CCCH type), RNA-binding motif and serine/arginine rich 2	X	2	2	-0.53	7.0
25960	GPR124	G protein-coupled receptor 124	8	2	2	-0.57	6.2
412	STS	steroid sulfatase (microsomal), isozyme S	X	4	1	-0.57	5.6
63934	ZNF667	zinc finger protein 667	19	2	1	-0.58	4.4
11238	CA5B	carbonic anhydrase VB, mitochondrial	X	2	1	-0.58	4.7
100128140	RPS4P17	ribosomal protein S4X pseudogene 17	17	1	1	-0.60	10.2
23263	MCF2L	MCF.2 cell line derived transforming sequence-like	13	8	1	-0.63	5.2
554203	LOC554203	alanyl-tRNA synthetase domain containing 1 pseudogene	X	3	3	-0.67	3.7
7403	UTX	ubiquitously transcribed tetratricopeptide repeat	X	4	3	-0.67	4.1
7543	ZFX	zinc finger protein, X-linked	X	4	2	-0.69	5.6
55787	CXorf15	chromosome X open reading frame 15	X	3	1	-0.72	5.6
10846	PDE10A	phosphodiesterase 10A	6	3	1	-0.72	3.6
8242	JARID1C	jumonji, AT rich interactive domain 1C	X	2	1	-0.73	4.0
1964	EIF1AX	eukaryotic translation initiation factor 1A, X-linked	X	4	3	-0.76	7.4
25924	MYRIP	myosin VIIA and Rab interacting protein	3	1	1	-0.76	2.7
7503	XIST	X (inactive)-specific transcript (non-protein coding)	X	9	9	-5.12	3.8

[a]Chromosome location of gene
[b]Total number of probesets for the indicated gene (i.e,. number of probesets that map to that gene)
[c]Number of probeset found differentially expressed ($P_{DE} \leq P_T$)
[d]M is the average over all probesets found DE for that gene, which is multiplied by the fold-change (FC).
[e]Average expression for all probeset A values for a given gene, where the A value is the average of all arrays.

Table S2. List of differentially expressed (DE) probes

Affy ID	Genbank	Entrez Gene	Unigene	Symbol	Description	Chr	M (log2 FC)	Pvalue	Qvalue	Avg. Expr.
224588_at	AA167449	7503	Hs.529901	XIST	X (inactive)-specific transcript (non-protein coding)	X	-8.81	6.90E-45	3.70E-40	4.79
227671_at	AV646597	7503	Hs.529901	XIST	X (inactive)-specific transcript (non-protein coding)	X	-7.70	2.40E-44	6.40E-40	3.94
221728_x_at	AA628440	7503	Hs.529901	XIST	X (inactive)-specific transcript (non-protein coding)	X	-6.75	3.40E-42	6.20E-38	4.13
214218_s_at	AV699347	7503	Hs.529901	XIST	X (inactive)-specific transcript (non-protein coding)	X	-6.38	8.40E-42	1.10E-37	3.49
224590_at	BE644917	7503	Hs.529901	XIST	X (inactive)-specific transcript (non-protein coding)	X	-6.07	5.90E-37	4.60E-33	3.24
235446_at	AW856618	7503	Hs.529901	XIST	X (inactive)-specific transcript (non-protein coding)	X	-3.88	2.20E-18	7.40E-15	3.93
224589_at	BF223193	7503	Hs.529901	XIST	X (inactive)-specific transcript (non-protein coding)	X	-3.75	3.50E-25	1.80E-21	2.97
231592_at	AV646335	7503	Hs.529901	XIST	X (inactive)-specific transcript (non-protein coding)	X	-1.97	2.10E-23	9.60E-20	3.11
203991_s_at	NM_021140	7403	Hs.522616	UTX	ubiquitously transcribed tetratricopeptide repeat	X	-0.95	8.80E-09	1.70E-05	4.17
1554447_at	BC029480	554203	Hs.648316	LOC554203	alanyl-tRNA synthetase domain containing 1 pseudogene	X	-0.94	1.50E-07	2.00E-04	4.06
229022_at	AI745209	7543	Hs.336681	ZFX	zinc finger protein, X-linked	X	-0.93	1.10E-11	2.30E-08	5.68
201018_at	AL079283	1964	Hs.522590	EIF1AX	eukaryotic translation initiation factor 1A, X-linked	X	-0.89	1.20E-06	1.30E-03	7.71
227498_at	AI480314						-0.88	2.80E-05	2.50E-02	3.35
229315_at	AI912175						-0.88	1.30E-08	2.30E-05	4.07
243712_at	AA022679	7503	Hs.529901	XIST	X (inactive)-specific transcript (non-protein coding)	X	-0.78	8.40E-07	1.00E-03	4.22
1569311_at	BC038589	554203	Hs.648316	LOC554203	alanyl-tRNA synthetase domain containing 1 pseudogene	X	-0.78	6.50E-07	8.00E-04	3.53
214156_at	AL050090	25924	Hs.594535	MYRIP	myosin VIIA and Rab interacting protein	3	-0.76	4.10E-06	4.40E-03	2.72
239207_at	BE503653	8242	Hs.631768	JARID1C	jumonji, AT rich interactive domain 1C	X	-0.73	6.00E-08	8.90E-05	4.02
205501_at	AI143879	10846	Hs.348762	PDE10A	phosphodiesterase 10A	6	-0.72	6.20E-05	4.30E-02	3.63
227520_at	AI885312	55787	Hs.555961	CXorf15	chromosome X open reading frame 15	X	-0.72	3.70E-05	3.00E-02	5.58
201019_s_at	NM_001412	1964	Hs.522590	EIF1AX	eukaryotic translation initiation factor 1A, X-linked	X	-0.70	1.20E-07	1.70E-04	8.47
201016_at	BE542684	1964	Hs.522590	EIF1AX	eukaryotic translation initiation factor 1A, X-linked	X	-0.68	4.70E-06	4.90E-03	6.11
235898_at	AW105010						-0.65	4.70E-05	3.50E-02	4.37
35147_at	AB002360	23263		MCF2L	MCF.2 cell line derived transforming sequence-like	13	-0.63	1.70E-06	1.90E-03	5.23
203992_s_at	AF000992	7403	Hs.522616	UTX	ubiquitously transcribed tetratricopeptide repeat	X	-0.62	3.90E-05	3.10E-02	5.31
216342_x_at	AL121916	100128140		RPS4P17	ribosomal protein S4X pseudogene 17	17	-0.60	1.50E-10	2.90E-07	10.21
65718_at	AI655903	25960	Hs.274136	GPR124	G protein-coupled receptor 124	8	-0.58	2.60E-08	4.20E-05	5.74
214082_at	AW003516	11238	Hs.653287	CA5B	carbonic anhydrase VB, mitochondrial	X	-0.58	1.00E-05	9.70E-03	4.68
236635_at	AI332774	63934	Hs.712574	ZNF667	zinc finger protein 667	19	-0.58	4.60E-05	3.50E-02	4.38
203769_s_at	NM_000351	412		STS	steroid sulfatase (microsomal), isozyme S	X	-0.57	6.90E-06	7.10E-03	5.62
221814_at	BF511315	25960	Hs.274136	GPR124	G protein-coupled receptor 124	8	-0.56	1.30E-05	1.20E-02	6.55
208174_x_at	NM_005089	8233	Hs.171909	ZRSR2	zinc finger (CCCH type), RNA-binding motif	X	-0.55	4.90E-07	6.40E-04	6.95
213876_x_at	AW089584	8233	Hs.171909	ZRSR2	zinc finger (CCCH type), RNA-binding motif	X	-0.50	4.20E-07	5.60E-04	6.99
217019_at	AL137162						-0.48	2.50E-06	2.80E-03	4.3
214678_x_at	R51161	7543	Hs.336681	ZFX	zinc finger protein, X-linked	X	-0.46	1.20E-05	1.10E-02	5.44
238220_at	BE670257	7403	Hs.522616	UTX	ubiquitously transcribed tetratricopeptide repeat	X	-0.45	1.40E-06	1.60E-03	2.78
213347_x_at	AW132023	6191	Hs.118076	RPS4X	ribosomal protein S4, X-linked	X	-0.44	1.40E-08	2.40E-05	12.09
227524_at	H06187						-0.43	6.50E-05	4.50E-02	3.67
219381_at	NM_023073	65250	Hs.643420	C5orf42	chromosome 5 open reading frame 42	5	-0.43	9.80E-06	9.70E-03	2.94
243172_at	AI079944						-0.43	3.20E-05	2.70E-02	3.02

200933_x_at	NM_001007	6191	Hs.118076	RPS4X	ribosomal protein S4, X-linked	X	-0.41	1.30E-08	2.30E-05	12.36
201210_at	NM_001356	1654	Hs.380774	DDX3X	DEAD (Asp-Glu-Ala-Asp) box polypeptide 3, X-linked	X	-0.38	3.10E-05	2.60E-02	9.97
224936_at	BE252813	1968	Hs.539684	EIF2S3	eukaryotic translation initiation factor 2, subunit 3 gamma	X	-0.37	2.10E-05	1.90E-02	8.44
1554448_at	BC029480	554203	Hs.648316	LOC554203	alanyl-tRNA synthetase domain containing 1 pseudogene	X	-0.29	6.60E-05	4.50E-02	3.57
218217_at	NM_021626	59342	Hs.514950	SCPEP1	serine carboxypeptidase 1	17	0.32	4.50E-05	3.50E-02	8.37
214804_at	BF793446	2491		CENPI	centromere protein I	X	0.34	7.10E-05	4.80E-02	3.5
208651_x_at	M58664	100133941		CD24	CD24 molecule	6	0.34	4.10E-05	3.20E-02	5.06
210322_x_at	AF000995	7404	Hs.115277	UTY	ubiquitously transcribed tetratricopeptide repeat gene, Y-linked	Y	0.48	4.70E-05	3.50E-02	4.05
211149_at	AF000994	7404	Hs.115277	UTY	ubiquitously transcribed tetratricopeptide repeat gene, Y-linked	Y	0.48	6.30E-07	8.00E-04	3.34
214983_at	AL080135	64595	Hs.433656	TTTY15	testis-specific transcript, Y-linked 15	Y	0.57	6.80E-07	8.20E-04	3.94
1555963_x_at	CA503291	93010	Hs.299329	B3GNT7	UDP-GlcNAc:betaGal beta-1,3-N-acetylglucosaminyltransfer 7	2	0.58	1.80E-05	1.60E-02	5.53
229150_at	AI810764						0.67	3.10E-05	2.60E-02	4.68
1555962_at	CA503291	93010	Hs.299329	B3GNT7	UDP-GlcNAc:betaGal beta-1,3-N-acetylglucosaminyltransfer 7	2	0.68	7.70E-06	7.80E-03	6.64
205738_s_at	NM_004102	2170	Hs.657242	FABP3	fatty acid binding protein 3, muscle and heart	1	0.75	1.70E-05	1.60E-02	6.25
1556656_at	BF477401						0.75	5.00E-05	3.60E-02	6.72
1560395_at	BC022384						0.80	5.80E-08	8.90E-05	3.44
206279_at	NM_002760	5616	Hs.632287	PRKY	protein kinase, Y-linked	Y	0.87	6.50E-12	1.40E-08	4.43
213553_x_at	W79394	341	Hs.110675	APOC1	apolipoprotein C-I	19	1.04	5.90E-05	4.20E-02	9.62
216379_x_at	AK000168	100133941		CD24	CD24 molecule	6	1.05	2.90E-08	4.60E-05	5.36
205695_at	NM_006843	10993	Hs.439023	SDS	serine dehydratase	12	1.06	5.90E-05	4.20E-02	7.11
209771_x_at	AA761181	100133941		CD24	CD24 molecule	6	1.07	7.00E-08	1.00E-04	5.44
207703_at	NM_014893	22829	Hs.439199	NLGN4Y	neuroligin 4, Y-linked	Y	1.12	1.40E-08	2.30E-05	3.55
229070_at	AA470369	84830	Hs.126409	C6orf105	chromosome 6 open reading frame 105	6	1.22	1.20E-05	1.10E-02	5.93
244482_at	AI753104						1.26	4.90E-11	1.00E-07	3.42
219697_at	NM_006043	9956		HS3ST2	heparan sulfate (glucosamine) 3-O-sulfotransferase 2	16	1.33	3.90E-05	3.10E-02	8.54
207063_at	NM_018542	55410	Hs.138453	CYorf14	chromosome Y open reading frame 14	Y	1.36	9.80E-14	2.50E-10	4.58
1560800_at	AL713714						1.44	3.10E-12	7.10E-09	4.17
230760_at	BF592062	7544	Hs.522845	ZFY	zinc finger protein, Y-linked	Y	1.51	4.40E-15	1.30E-11	3.41
232618_at	AF332224	246126	Hs.522863	CYorf15A	chromosome Y open reading frame 15A	Y	1.66	3.30E-14	9.10E-11	4.25
236694_at	AW468885	246126	Hs.522863	CYorf15A	chromosome Y open reading frame 15A	Y	1.82	1.30E-21	5.00E-18	3.88
223645_s_at	BF062193	84663	Hs.592254	CYorf15B	chromosome Y open reading frame 15B	Y	1.91	3.10E-17	1.00E-13	4.77
205001_s_at	AF000985	8653	Hs.99120	DDX3Y	DEAD (Asp-Glu-Ala-Asp) box polypeptide 3, Y-linked	Y	2.06	2.00E-15	6.20E-12	5.26
204410_at	NM_004681	9086	Hs.461178	EIF1AY	eukaryotic translation initiation factor 1A, Y-linked	Y	2.18	1.00E-13	2.50E-10	4.98
206624_at	NM_004654	8287	Hs.598540	USP9Y	ubiquitin specific peptidase 9, Y-linked)	Y	2.26	4.90E-13	1.20E-09	4.1
223646_s_at	AF332225	84663	Hs.592254	CYorf15B	chromosome Y open reading frame 15B	Y	2.49	4.40E-20	1.60E-16	5.14
228492_at	AV681765	8287	Hs.598540	USP9Y	ubiquitin specific peptidase 9, Y-linked	Y	2.93	1.30E-25	7.00E-22	5.13
214131_at	AL049280	84663	Hs.592254	CYorf15B	chromosome Y open reading frame 15B	Y	3.18	3.90E-23	1.60E-19	5.41
204409_s_at	BC005248	9086	Hs.461178	EIF1AY	eukaryotic translation initiation factor 1A, Y-linked	Y	3.96	1.20E-28	7.00E-25	5.42
206700_s_at	NM_004653	8284	Hs.80358	JARID1D	jumonji, AT rich interactive domain 1D	Y	3.99	5.90E-30	4.00E-26	6.38
205000_at	NM_004660	8653	Hs.99120	DDX3Y	DEAD (Asp-Glu-Ala-Asp) box polypeptide 3, Y-linked	Y	5.37	3.60E-37	3.30E-33	7.11
201909_at	NM_001008	6192	Hs.282376	RPS4Y1	ribosomal protein S4, Y-linked 1	Y	5.49	3.10E-38	3.40E-34	9.09

Manual curation of the APOC1 association network

Associations extracted from the literature were manually curated for the 24 nodes linked directly to APOC1 (Figure S2). Of the 24, we found 5 direct associations (dark green; 20.8%), 14 primary associations (light green; 58.3%), two secondary associations (light red; 8.3%), and 3 incorrect associations (dark red; 12.5%). Direct associations are, e.g. activation, inhibition or similar. In this network APOC1 appears to be directly associated to NUCB2, LPL, APOE, LDLR and VLDLR. For example, hyperlipidimic APOC1 mice show reduced NEFA (NUCB2) and increased glucose metabolism [1]. APOC1 dose-dependently inhibits LPL-dependent lipolysis [2]. APOC1 inhibits the APOE-mediated binding of triacylglycerol-rich lipoprotein particles to the VLDLR, whereas APOC1-enriched lipoproteins can still bind to the LDLR [3].

Primary associations indicate genes involved in the same biological processes or pathways. For example, the light green genes are related to cardiovascular disease: cholesterol binding lipoproteins (APOA1, APOA4, APOC1, APOC2, APOC3, APOD, APOE and LPA), cholesterol transporters (ABCA1 and ABCA2), lipoprotein receptors (LDLR, LRP1, LRP8 and VLDLR), and cholesterol metabolizing enzymes (CYP46A1 and CH25H) [4]. Secondary associations indicate genes that are directly associated to genes involved in the same biological process (e.g., downstream enzymes in a metabolic pathway). For example, SOAT1 and NR1H2 are downstream enzymes in the metabolism of cholesterol that metabolize the oxysterol products of CYP46A1 and CH25H [4]. Two gene names are incorrectly detected: NDUFB6 and TH1L. TH1L (also called NELFD) was incorrectly detected in a publication where a specific DNA variation was also named Th1l [5]. NDUFB6 is also known as C-IB17 (for complex I-B17) and was incorrectly detected in a publication using the term 'Apolipoprotein C-I' [6]. Finally, EDNRB was correctly detected, but there is no relationship between this gene and APOC1. There is a human LTR that provides alternative promoters for both genes, resulting in this LTR contributing to the expression of APOC1 and EDNRB [7].

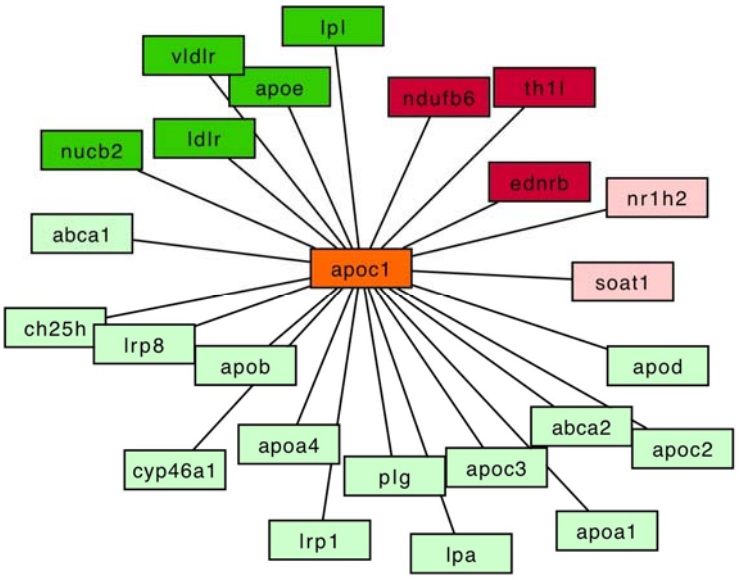


Figure S2. Association network for APOC1. Only direct links to APOC1 are indicated. The majority of these genes also have cross-interactions between each other.

In summary, these results demonstrate the direct implications of APOC1 in cardiovascular disease, as derived from the association network. Direct associations, as well as other indirect associations were detected. A few clear mismatches were identified, due to incorrect gene name and/or association detection. In the complete association network (Figure 5), TH1L was only linked to APOC1 and APOE, and NDUFB6 and EDNRB are only linked to APOC1. Accordingly, the impact of these errors in the network is very limited. Overall, these results highlight the utility of this approach to discover associations between genes, but demonstrate the need to verify the most important associations to avoid the impact of false positives.

Table S3. List of evidence terms for the APOC1 network.

1.	Severe hypertriglyceridemia in human APOC1 transgenic mice is caused by apoC-I-induced inhibition of LPL.[J Lipid Res, 46:(2), Feb, 2005][In Vitro,Journal Article,Research Support, Non-U.S. Gov't] [PubMed]
2.	Although total postheparin plasma LPL activity was not lower in APOC1 mice compared with controls, apoC-I was able to dose-dependently inhibit the LPL-mediated lipolysis of [3H]TG-VLDL-mimicking particles in vitro with a 60% efficiency compared with the main endogenous LPL inhibitor apoC-III[apoc3].[J Lipid Res, 46:(2), Feb, 2005][In Vitro,Journal Article,Research Support, Non-U.S. Gov't] [PubMed]
3.	The analyzed markers correspond to polymorphic sites in several candidate genes for cardiovascular disease including apolipopoteins and their receptors (APOA1, APOB, APOE, APOC1, APOC2, LPA, and LDLR), genes implied in the hemostasis regulation (Factor VII, alpha and beta-fibrinogen, alpha and beta platelet-integrin, tissue plasminogen[plg] activator, and plasminogen[plg] activator inhibitor-1), and the angiotensin converting enzyme gene.[Coll Antropol, 27:(2), Dec, 2003][Journal Article,Research Support, Non-U.S. Gov't] [PubMed]
4.	The common insertional polymorphism in the APOC1 promoter is associated with serum apolipoprotein C-I[ndufb6] levels in Hispanic children.[Atherosclerosis, 179:(2), Apr, 2005][Journal Article,Research Support, N.I.H., Extramural,Research Support, Non-U.S. Gov't,Research Support, U.S. Gov't, P.H.S.] [PubMed]
5.	In this study we investigated the effect of primary hyperlipidaemia on basal and insulin-mediated glucose and on non-esterified fatty acid (NEFA[nucb2]) metabolism and mean arterial pressure in hyperlipidaemic transgenic mice overexpressing apolipoprotein C1 (APOC1).[Diabetologia, 44:(4), Apr, 2001][Journal Article] [PubMed]
6.	Insulin-mediated whole body NEFA[nucb2] uptake, NEFA[nucb2] oxidation (generation of 3H2O) and NEFA[nucb2] storage were lower in APOC1 mice than in wild-type mice (15 +/- 3 vs 33 +/- 6; 3 +/- 2 vs 11 +/- 4 and 12 +/- 2 vs 22 +/- 4 mumol.kg-1.min-1, p 0.05) in the face of higher plasma NEFA[nucb2] concentrations (1.3 +/- 0.3 vs 0.5 +/- 0.1 mmol/l, p 0.05), respectively.[Diabetologia, 44:(4), Apr, 2001][Journal Article] [PubMed]
7.	CONCLUSIONS/INTERPRETATION: 1) Hyperlipidaemic APOC1 mice show reduced NEFA[nucb2] and increased glucose metabolism under both basal and insulin-mediated conditions, suggesting an intrinsic defect in NEFA[nucb2] metabolism.[Diabetologia, 44:(4), Apr, 2001][Journal Article] [PubMed]
8.	Excess of APOC1 protein does inhibit the hepatic clearance of VLDL remnant particles, whereas excess of apoE leads to a hampered extra-hepatic lipolysis of VLDL triglyceride.[Prostaglandins Leukot Essent Fatty Acids, 57:(4-5), Oct, 1997][Journal Article] [PubMed]

9. APOE and APOC1 promoter polymorphisms and the risk of Alzheimer disease in African American and Caribbean Hispanic individuals.[Arch Neurol, 61:(9), Sep, 2004][Comparative Study,Journal Article,Research Support, Non-U.S. Gov't,Research Support, U.S. Gov't, P.H.S.] [PubMed]

10. METHODS: We examined the association between AD and variants in 3 APOE promoters and in the promoter of the adjacent APOC1 gene in African American and Caribbean Hispanic individuals.[Arch Neurol, 61:(9), Sep, 2004][Comparative Study,Journal Article,Research Support, Non-U.S. Gov't,Research Support, U.S. Gov't, P.H.S.] [PubMed]

11. Polymorphisms tested were the -491A/T, -427T/C, and -219G/T (Th1[th1l]/E47cs) in the APOE promoter and the HpaI variant in the APOC1 promoter.[Arch Neurol, 61:(9), Sep, 2004][Comparative Study,Journal Article,Research Support, Non-U.S. Gov't,Research Support, U.S. Gov't, P.H.S.] [PubMed]

12. No promoter variant in APOE or APOC1 was associated with AD before or after adjusting for age, education, sex, and multiple comparisons.[Arch Neurol, 61:(9), Sep, 2004][Comparative Study,Journal Article,Research Support, Non-U.S. Gov't,Research Support, U.S. Gov't, P.H.S.] [PubMed]

13. CONCLUSION: These findings exclude a strong or independent influence of APOE or APOC1 promoter polymorphisms on the variation in APOE-related risk of AD in African American and Caribbean Hispanic individuals.[Arch Neurol, 61:(9), Sep, 2004][Comparative Study,Journal Article,Research Support, Non-U.S. Gov't,Research Support, U.S. Gov't, P.H.S.] [PubMed]

14. We previously reported that the long terminal repeats (LTRs) of retroviral elements belonging to the HERV-E family contribute to the expression of the human apolipoprotein C1 (APOC1) and endothelin B receptor (EDNRB) genes by providing alternative promoters.[J Virol, 77:(13), Jul, 2003][Journal Article,Research Support, Non-U.S. Gov't] [PubMed]

15. In the absence of at least the low-density-lipoprotein receptor (LDLR), it was shown that APOC1 overexpression in transgenic mice inhibited the hepatic uptake of VLDL via the LDLR-related protein.[Biochem J, 338 (Pt 2), Mar, 1999][Journal Article,Research Support, Non-U.S. Gov't] [PubMed]

16. In the present study, we have now examined the effect of apoC1 on the binding of lipoproteins to both the VLDL receptor (VLDLR) and the LDLR.[Biochem J, 338 (Pt 2), Mar, 1999][Journal Article,Research Support, Non-U.S. Gov't] [PubMed]

17. The binding specificity of the VLDLR and LDLR for apoC1-enriched lipoprotein particles was examined in vivo through adenovirus-mediated gene transfer of the VLDLR and the LDLR [giving rise to adenovirus-containing (Ad)-VLDLR and Ad-LDLR respectively] in APOC1 transgenic mice, LDLR-deficient (LDLR-/-) mice and wild-type mice.[Biochem J, 338 (Pt 2), Mar, 1999][Journal Article,Research Support, Non-U.S. Gov't] [PubMed]

18. These results suggest that apoC1 inhibits the clearance of lipoprotein particles via the VLDLR, but not via the LDLR.[Biochem J, 338 (Pt 2), Mar, 1999][Journal Article,Research Support, Non-U.S. Gov't] [PubMed]

19. Chinese hamster ovary (CHO) cells expressing the VLDLR (CHO-VLDLR) or LDLR (CHO-LDLR) bound less APOC1 transgenic VLDL than wild-type VLDL.[Biochem J, 338 (Pt 2), Mar, 1999][Journal Article,Research Support, Non-U.S. Gov't] [PubMed]

20. From these studies, we conclude that apoC1 specifically inhibits the apoE-mediated binding of triacylglycerol-rich lipoprotein particles to the VLDLR, whereas apoC1-enriched lipoproteins can still bind to the LDLR.[Biochem J, 338 (Pt 2), Mar, 1999][Journal Article,Research Support, Non-U.S. Gov't] [PubMed]

21. Intriguingly, however, enrichment with apoE enhanced dose-dependently the binding of wild-type VLDL to CHO-VLDLR cells (up to 5-fold), whereas apoE did not enhance the binding of APOC1 transgenic VLDL to these cells.[Biochem J, 338 (Pt 2), Mar, 1999][Journal Article,Research Support, Non-U.S. Gov't] [PubMed]

22. In contrast, for binding to CHO-LDLR cells, both wild-type and APOC1 transgenic VLDL were stimulated upon enrichment with apoE.[Biochem J, 338 (Pt 2), Mar, 1999][Journal Article,Research Support, Non-U.S. Gov't] [PubMed]

23. We found that APOE promoter polymorphisms and APOC1 insertion alleles were significantly associated with AD.[Neurosci Lett, 350:(1), Oct, 2003][Comparative Study,Journal Article,Research Support, Non-U.S. Gov't] [PubMed]

24. We have recently shown that the predominant hypertriglyceridemia in human apolipoprotein C1 (APOC1) transgenic mice is mainly explained by apoCI-mediated inhibition of the lipoprotein lipase (LPL)-dependent triglyceride (TG)-hydrolysis pathway.[Biochim Biophys Acta, 1761:(2), Feb, 2006][Journal Article,Research Support, Non-U.S. Gov't] [PubMed]

25. Since a deletion/insertion polymorphism in the promoter region of the apolipoprotein C-I[ndufb6] (APOC1) gene has been reported to be associated with late-onset Alzheimer's disease (LOAD), we examined the hypothesis in a Korean population with 120 LOAD cases and 132 age-matched controls.[Neurosci Lett, 319:(2), Feb, 2002][Journal Article,Research Support, Non-U.S. Gov't] [PubMed]

26. They include all of the key components of a glia/neurone cholesterol shuttle including cholesterol binding lipoproteins APOA1, APOA4, APOC1, APOC2, APOC3, APOD, APOE and LPA, cholesterol transporters ABCA1, ABCA2, lipoprotein receptors LDLR, LRP1, LRP8 and VLDLR, and the cholesterol metabolising enzymes CYP46A1 and CH25H, whose oxysterol products activate the liver X receptor NR1H2 and are metabolised to esters by SOAT1.[Neurochem Int, 50:(1), Jan, 2007][Journal Article,Review] [PubMed]

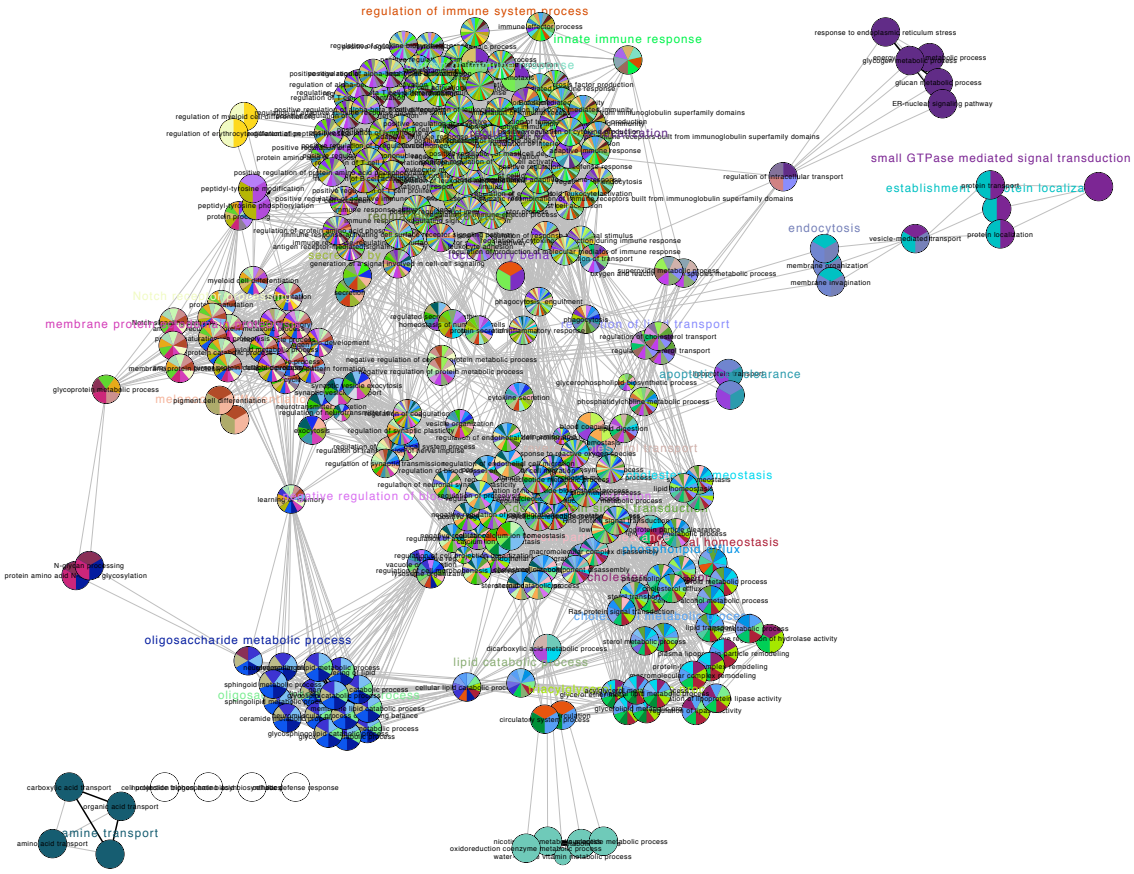


Figure S3. Network of gene ontology (GO) terms obtained from the analysis of the sub-network shown in Figure 7 with *ClueGO*, using all terms in GO Biological Process. The terms are clustered according to the *kappa* statistics, enabling the functional organization of the GO terms.

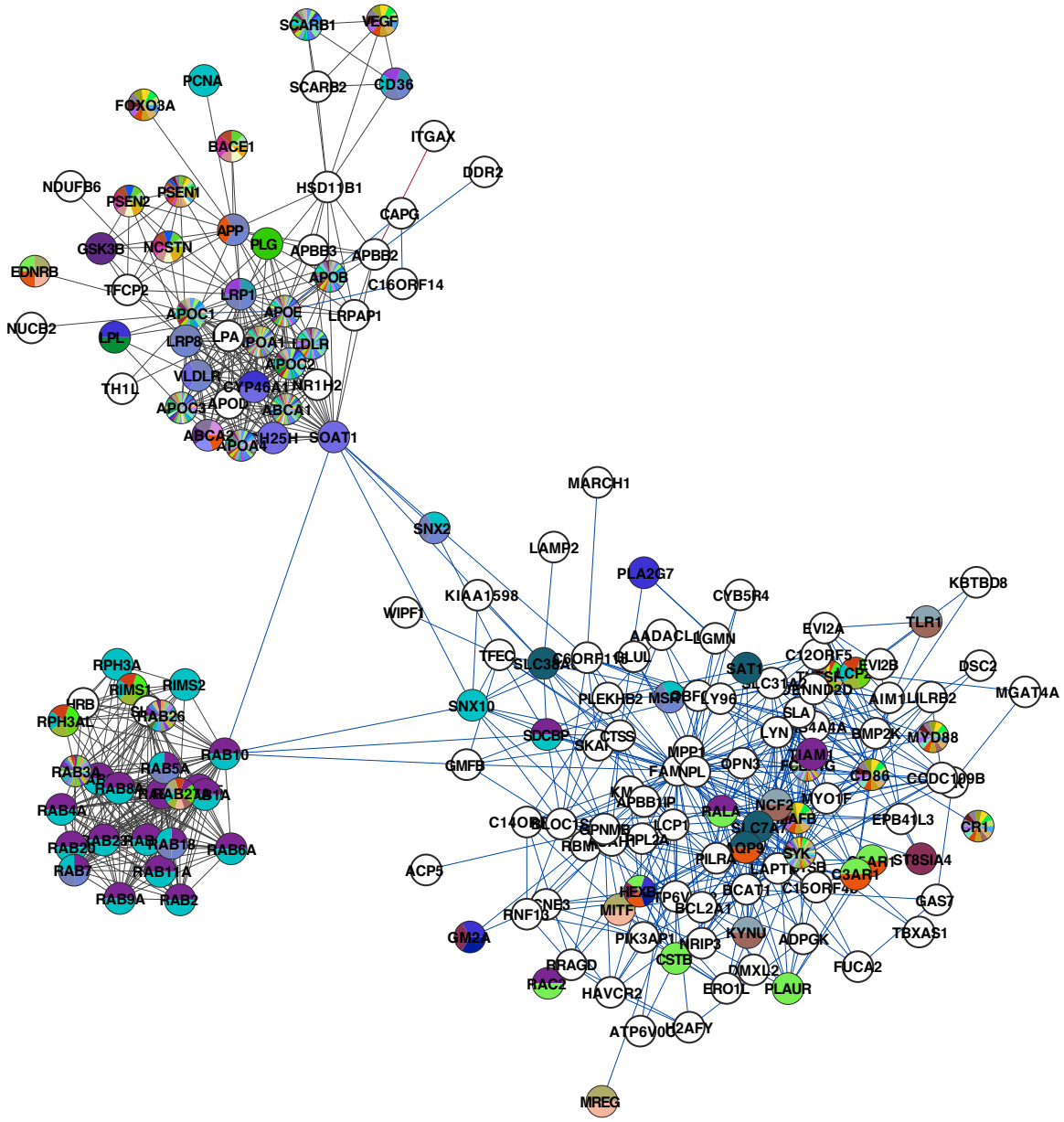


Figure S4. Coloring of the entire sub-network from Figure 7 following analysis with *ClueGO*. *ClueGO* was used to find over-represented GO terms in the category 'Biological Process', using all evidence codes and the Benjamini and Hochberg correction. [8] Several GO terms were found, and clustered into functional groups (Figure S3). The groups were mapped to the sub-network to see the functional distribution.

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