

Coordinated modular functionality and prognostic potential of a heart failure biomarker-driven interaction network

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Supplementary information

Table S1. List of HF biomarkers used as inputs to the PPI network inference and analysis framework

CRP	MMP2	AGT	ST2
TNF	MMP7	AVP	CHGA
IL1B	TIMP1	EDN1	CHGB
IL6	TIMP2	TNNI1	LGALS3
IL18	TIMP3	MYLK	TNFSF11
OLR1	TIMP4	FABP3	ADIPOQ
MPO	COL2A1	CKM	GDF15
MMP20	COL1A2	NPPB	
MMP9	PIINP	NT-proBNP	
MMP3	REN	MR-proADM	

Table S2. List of candidate genes related to the set of HF biomarkers using the Endeavour software system. GSR: Global similarity ranking. The union of this list with the list shown in Table S1 represents the network seeds.

GSR	Gene	GSR	Gene	GSR	Gene
1	TIMP1	34	MPO	67	FBLN1
2	MMP2	35	COL3A1	68	COL5A2
3	IL6	36	COL6A1	69	SERPINA1
4	TIMP2	37	MMP20	70	ACAN
5	IL1B	38	MYLK	71	MMP16
6	MMP3	39	IL18	72	LPL
7	TNF	40	TNFSF11	73	IL6ST
8	MMP9	41	CKM	74	FBN1
9	COL1A2	42	TGFB1	75	APCS
10	TIMP3	43	MMP10	76	COL6A2
11	MMP7	44	COL5A1	77	LTA
12	ENSG00000206439	45	CRP	78	IL6R
13	ENSG00000206328	46	MMP17	79	PDGFRB
14	EDN1	47	VWF	80	COMP
15	AGT	48	IL1R1	81	COL8A1
16	NPPB	49	SPP1	82	ADAM15
17	COL1A1	50	ADAMTS1	83	THBS2
18	MMP1	51	GDF15	84	CCL2
19	REN	52	NPPA	85	FBLN2
20	COL6A3	53	IL1A	86	CYR61
21	CHGA	54	TGFB1	87	COL5A3
22	TIMP4	55	COL11A1	88	C3
23	ADIPOQ	56	LIF	89	DPT
24	MMP8	57	COL7A1	90	AVP
25	COL2A1	58	COL4A1	91	BMP2
26	SERPINE1	59	OLR1	92	C5
27	LGALS3	60	LAMC1	93	F12
28	COL4A2	61	COL10A1	94	IL1RAP
29	FN1	62	IL1RN	95	PDGFRA
30	MMP13	63	FABP3	96	LGALS3BP
31	MMP19	64	COL4A3	97	TNC
32	CHGB	65	SPARC	98	LAMB1
33	MMP14	66	ITGB1	99	ADAMTS5
				100	COL4A5

Table S3. Characteristics of the MI patients used for gene expression analyses.

	High EF (n=16)		Low EF (n=16)		P
1-month EF, % (median-range)	63	45-73	35	20-40	3.E-04
Age, y (median-range)	59	46-86	72	40-85	0.59
Sex (male, n, %)	13	81%	13	81%	1
Body Mass Index (median-range)	28	23-35	26	20-38	0.25
Cardiovascular history, n (%)					
Prior MI	2	13%	3	19%	0.64
CABG	0	0%	1	6%	0.33
PTCA	1	6%	2	13%	0.56
Diabetes	4	25%	6	38%	0.46
Hypertension	6	38%	7	44%	0.73
Hypercholesterolemia	7	44%	9	56%	0.50
Tobacco	4	25%	5	31%	0.71
Medications, n (%)					
Beta-blockers	16	100%	14	88%	0.16
Calcium antagonists	3	19%	0	0%	0.08
Nitrates	6	38%	6	38%	1
ACE inhibitors	10	63%	10	63%	1
Statins	14	88%	14	88%	1
Angiotensin inhibitors	2	13%	0	0%	0.16

All patients had successful mechanical reperfusion and stenting of the infarct artery within 12 hours of chest pain onset. All patients received Aspirin, Clopidogrel, Heparin and Abciximab. ACE: Angiotensin-Converting Enzyme; CABG: Coronary Artery Bypass Grafting; EF: Ejection Fraction; PTCA: Percutaneous Transluminal Coronary Angioplasty; MI: Myocardial Infarction.

Table S4. Description of network modules. BP and CC are examples of biological processes and cellular localizations terms respectively, which are highly over-represented in each module, as defined in the GO (January 2009 release). *P* values estimating the statistical significance, after correcting for multiple-testing, of the enrichment of the terms are also included. “*”: enrichment of GO term in the module was nominally significant at *P* = 0.05, i.e. no statistical significance was observed after correcting *P* value for multiple-testing. “?”: statistically detectable GO terms were not found.

Module	Proteins				BP	CC
1	CD46 C3 PFC IF CFHL5 CFHL3 CFHL4 ITGAX BF	C3AR1 CFB CFHR4 CFHR5 CFI GC GPR77 ITGAM MASP1	OLFM4 PAPPA CFHR3 CFP CR1 CR2 CPN1 C5 C6	C7 C8B C5R1 CPB2 C5AR1 C2	Innate immune response (<i>P</i> = 2.0E-22) Acute inflammatory response (<i>P</i> = 4.4E-22)	Extracellular region part (<i>P</i> = 5.1E-17) Intracellular space (<i>P</i> = 2.8E-7)
2	CRP CFH SNRP70 GP6 HIST1H1A	HIST1H2AL LEP spa FCN2 APCS C4BPA	FCGR3A FCGR3B CALU TG FCGR1A	FCGR2B	Immune response (<i>P</i> = 5.0E-04)	Cell part (<i>P</i> = 0.01) Extracellular space (<i>P</i> = 0.05)
3	COL7A1 LAMB3 LAMC2 LAMA1 FBXL2	HSPA8 SHKBP1 ZC3H7A LAMA5 LAMC1	SNAPAP CGI-116 ATF7IP CCDC53 COL6A3	PDIA3 BALF4 LAMB1 ACHE ATXN7L2 GFI1B	Cell adhesion (<i>P</i> = 1.8E-06) Epithelial cell proliferation (<i>P</i> = 6.0E-06)	Extracellular matrix part (<i>P</i> = 8.4E-14) Laminin complex (<i>P</i> = 8.4E-14)
4	DPT LOX BAT3 CMA1 EDN1 COPS6 ADM ECE1	EDNRB KEL EDNRA MME NPPA EPB41 NPR3 NPR2	NPPB EWSR1 NPR1 AGT ACE2 AGTR2 CTSG DNPEP	KNG1 MAS1 MEP1A PRCP PREP PRG2 AGTR1 ENPEP Agtr1	Circulation (<i>P</i> = 2.3E-19) Regulation of blood vessel size (<i>P</i> = 2.7E-14)	Plasma membrane (<i>P</i> = 0.003) Intracellular (<i>P</i> = 0.01)
5	LRP2 LPL EMD ASCC2 FLJ13855 RPL18A KIAA1377 LUC7L2	PTPN4 VLDLR CETP APOC2 UBE2Z TNFRSF11B VWF F8	TRA1 HSPA5 PDIA4 ADAMTS13 EBNA6 BILF1 q1hvi2_ebvg BHRF1	ZNF512B CALR F12 APOH KLKB1 MMP12 KRT1 GP1BA	Coagulation (<i>P</i> = 1.6E-08) Regulation of body fluids (<i>P</i> = 3.2E-08)	Extracellular region part (<i>P</i> = 2.0E-04) Chylomicron (<i>P</i> = 2.0E-04)

Table S4 (cont.)

6	TGFB1	VTN	ITGB8	MGP	Tissue remodeling ($P = 8.4E-05$)	Integral to plasma membrane ($P = 0.005$)
	FNTA	FCN1	LTBP3	BMPR1B		
	SLITL2	ACVRL1	LTBP4	ACVR1	Cell communication ($P = 3.3E-09$)	Intracellular ($P = 3.0E-05$)
	SNIP1	YWHAE	VASN	ASCL1		
	EIF3S2	TGFBRAP1	BMP2	BMPER		
	FMOD	FKBP1A	BMPR1A	CHRD2		
	TGFB1	CCL3	BMPR2	GREM2		
	TGFB3	ITGB6	SOSTDC1	NOG		
	TGFB2	DAXX	ACTR2	ENG		
				TGFB2		
7	MYOC	COCH	TGFB1	MEP1B	Cell adhesion ($P = 2.7E-18$)	Extracellular region part ($P = 9.9E-47$)
	COL1A2	PRELP	COL4A4	ITGB7		
	SHBG	BGN	FBLN2	REG3A	Anatomical structure development ($P = 9.8E-09$)	Collagen ($P = 6.9E-33$)
	q69hr1_cioin	CHAD	BFLF2	TSHR		
	CD36	COL9A2	FBN1	TRIB3		
	ITGA2B	COL9A3	FBN2	CTSD		
	ITGB3	LOXL4	MFAP2	GALNT6		
	ANXA5	TNFRSF10A	CSPG2	LRG1		
	COL10A1	MATN1	ELN	NT5E		
	P4HB	DCN	VCAN	SCGB1A1		
	COL2A1	COL4A1	HSPG2	TAC1		
	COL9A1	DISC1	FBLN1	FST		
	COMP	SMAD1	MPG	FSTL3		
	FGF7	SAA2	FGF	HGF		
	COL6A1	SAA4	ECM1	SMAD9		
	MAG	COL16A1	PREI3	CD79A		
	COL3A1	COL4A2	E6	NOV		
	PCOLCE	FAM46A	ATN1	C1QA		
	DDR1	ANTXR2	MFAP5	COL4A6		
	COL11A1	BMP3	NID	TMPRSS6		
	THBS1	COL4A3	NID1	COL6A2		
	COL5A1	CAMK2B	CTGF	ABCD3		
	SDC3	COL4A3BP	SMAD4	ADAMTS1		
	COL5A3	USH2A	LTBP1	FURIN		
	BMP1	SAA1	SKIL	VEGF		
	COL5A2	OSM	FN1	A2M		
	COL1A1	COL4A5	LACRT	VEGFA		
	TXN	RNF10	MIA	SPARC		
	NID2	HABP2	AMBP	CTSK		
	CAPN1	SERPINE2	VHL	PDGFB		
	DDR2	APP	LPA	COL13A1		
	HTRA1	CD93	GSN	PLAT		
	PKD1	MATN2	F13A1	SDC2		
				TGM2		

Table S4 (cont.)

9	SPP1	IL8	IGFBP3	CLDN1	Protein digestion (<i>P</i> = 2.6E-13)	Extracellular region part (<i>P</i> = 5.1E-33)
	SGTA	BTC	BCAN	SPOCK1		
	PDLIM7	RECK	CCL7	TFPI	Collagen metabolic process (<i>P</i> = 6.1E-13)	Extracellular space (<i>P</i> = 10.0E-19)
	CTNBNL1	MMP7	MMP13	MMP8		
	ABCF3	FASLG	ADAMTS5	UMOD		
	ACP5	HBEGF	MMP14	TIMP2		
	PRKG1	MBP	LUM	SNCG		
	SERPINE1	NGFB	C1QBP	PSMA7		
	PLAU	CD44	ADI1	AGC1		
	ORM1	PLG	SDC1	MMP19		
	KLK2	CHGA	BCAR1	MMP20		
	LRP1B	SCG3	ACTB	TNFSF11		
	F2	THBS2	LRP1	KHDRBS1		
	IGFBP5	THBS3	MMP17	ADAM19		
	UBQLN4	TIMP1	KISS1	AKT1		
	ITGAV	RECQL5	MMP16	MAPK1		
	CYR61	ECH1	MMP2	MAPK8		
	ITGB5	EEF1B2	COL18A1	NFKB1		
	HAPLN1	MMP3	FGFR1	NFKBIA		
	MMP10	CCL8	BACE1	TNFRSF11A		
	MMP9	SPOCK3	CXCL12	TRAF6		
	CXCL5	MMP1	MMP25	TIMP3		
	CXCL6	BSG	PZP	EFEMP1		
	AREG	SERPINA3	LCN2	KDR		
	CXCL1	CCL13	TIMP4	ADAM17		
10	GEMIN4	CUBN	PHB2	MAP3K1	Positive regulation of signal transduction (<i>P</i> = 1.3E-05)	?
	FCGR2A	CYHR1	CD14	IKBKE		
	C1GALT1C1	LIM2	PPIC	TRADD		
	CSNK1A1	SUFU	LMP2	MAP3K7IP2		
	CSNK2A1	LGALS1	NFKB2	TBK1		
	CSNK2A2	LGALS3BP	RELA			
11	ELA2	PRTN3	ADAMTS4	MMP11	Proteolysis (<i>P</i> = 2.4E-13)	Endoplasmatic reticulum (<i>P</i> = 0.004)
	SERPINA1	CTRB1	DERL2	MMP26		
	KLK13	ELA1	DERL3	PRSS2	Digestion (<i>P</i> = 1.4E-07)	
	KLK3	PRSS1	KLK5	PRSS3		
				try1_bovin SSR1 CANX		
12	TNFRSF1A	POU2F1	IL18	MYD88	Immune response (<i>P</i> = 7.1E-13)	Extracellular space (<i>P</i> = 0.002)
	LTA	ADAM9	IL18BP	TICAM2		
	LTB	IFNG	CASP3	IL1RAP		
	LGALS2	PDIK1L	IL18RAP	IRAK1		
	LTBR	VACCL3_190	IL1RL2	TIRAP		
	TNFRSF14	crmE	CASP4	SIRPA		
	TUBA4A	IL1B	IL18R1	PRPF40A		
	TUBA1A	MAPK8IP2	IL1R1	RAC1		
	TNFRSF1B	ZNF675	SIGIRR	IL1R2		
	TNF	ADRB2	IRAK2	IL1RN		
	RALBP1	UBE2N	IL1F10	IL1A		
	TRAF2	CASP1	TOLLIP	NDN		
				S100A13		
				HAX1		
			NFKBIE			

Table S4 (cont.)

13	ITGA2	FLNB	ITGA10	ITGB1BP2	Cell adhesion ($P = 4.4E-15$)	Receptor complex ($P = 7.5E-12$)
	COL8A1	GNB2L1	SLC3A2	Apba1		
	KLHL12	MAP4K4	TGOLN2	Tln1		
	KRTAP4-12	ACTN1	ICAM4	Dab1	Cell matrix adhesion ($P = 2.4E-14$)	Integrin complex ($P = 1.2E-11$)
	EFEMP2	TSPAN4	IGF1R	tenc1_human		
	CD81	CD81	ACTN4	ANKS1B		
	COL8A2	FLT4	ARHGAP5	RABGAP1		
	ITGA1	CD151	CD82	RGS12		
	CD63	ITGA9	DOK1	Shc1		
	ITGB1	FBXO2	EPS8	GULP1		
	FLNA	CD47	ITGA11	EMILIN1		
	ITGA3	TLN1	ITGA4	CKM		
	RAB8B	ITGA6	ITGB1BP3	TRIM63		
	ITGB1BP1	YWHAB	NF2	UBC		
	CD9	NME1	PRKCE	CKB		
	ILK	PXN	PTK2	MYOM2		
	ITGA8	LGALS8	VCAM1	FHL2		
	MCP					
14	ITGA5	CSPG3	PTPRB	NCAN	Cell adhesion ($P = 2.0E-04$)	?
	TNC	EGFR	CNTN1			
15	MAPK3	CTTN	CAMK2G	TNNI1	Muscle contraction ($P = 5.1E-05$)	Myofibril ($P = 9.0E-04$)
	MYLK	PAK1	PAK2	DIPA		
	PRKG2	MLC1	PRKCA	PKD2L1		
	CALM1	ACTC1	ACTA1	TNNC1	Protein amino phosphorylation ($P = 10.0E-05$)	Contractil fiber part ($P = 9.0E-04$)
				CCDC85B		
16				TNNT1		
	SRC	PLAUR	FYN	PRDX2	Cell communication ($P = 7.7E-27$)	Cytoplasm ($P = 0.007$)
	IL6R	HCK	YES1	SOCS1		
	MAGI1	VAV1	NCK1	SYNGAP1		
	WWP1	JAK2	PDGFRB	E5	Signal transduction ($P = 6.5E-23$)	
	WWP2	TYK2	VAV2	ATP6V0C		
	CNTF	SHC1	VAV3	Ptpn1		
	STAT3	PTPN11	COPA	Ptpn11		
	JAK1	GRB2	NCK2	SLC9A3R2		
	IL6	ADAM15	RAF1	PDGFRA		
	PTHLH	SNX9	SH3KBP1	PDGFA		
	ZBTB16	TRIP13	EDG1	PDGFC		
	SH3GL2	MAD2L2	EIF2AK2	CRKL		
	IL6ST	LCK	SNX1	SNX6		
	LIFR	MAD2L1	RASA1	SHB		
	LIF	ABL1	ARAF	SHF		
	TLE1	PACSIN3	GAB1	CBL		
	AR	NPHP1	GRB7	CRK		
	CNTFR	ArgBP2a	BAG1	PLCG1		
	SOCS3	LYN	COPB1	SLC9A3R1		
	CDK9	PTPN3	GRB10	CAV1		
	ERBB2	ARHGEF7	PDAP1	CAV3		
	ERBB3	ARHGEF6	PDGFD	SNX2		
	OSMR	SH3PXD2A	PTPN1	SNX4		
	PRKCD	SH3MD4	PTPN2	GRB14		
	PTPN6	SH3PX3	PTPRJ	STAT1		
	CTF1	SORBS1	PIK3CA	STAT5A		
	IL31RA	q9ham2_human	PIK3R2	STAT5B		
	PIK3CG	SH3GLB1	PIK3R3	PIK3R1		

Table S4 (cont.)

17	PTEN	MGLL	S100A8	UBTF	Response to stress* ($P = 0.01$)	DNA-directed RNA polymerase complex* ($P = 0.04$)
	CHGB	OGG1	SLC25A6	TUBB2A		
	ACACA	PTK9L	YLPM1	SGK223		
	CBFB	SAFB2	TUBB2	q8nbb9_human		
	DKFZp761P0423	ATXN2	CCDC92	hCG_1723909		
	FLJ22471	TAZ	POLR2E	OGG1 type 1e		
	GCS1	FGFR3	RXRG			
	MARK3	POLD1	TWF2			

Software code.

Java-written routines for searching and retrieving interactions of biomarkers ("biomarkers.txt") from a (binary) protein-protein interaction text file (tab-separated, two-column).

```
/*
 * @author Francisco Azuaje
 */

package datamanager;
import java.io.*;
import java.util.*;
import java.lang.Math.*;

public class NewMain {

    public static int maxLines = 10000;
    public static int maxFeatures = 20;
    public static int maxInputBiomarkers = 800;
    static String [] biomarkers = new String[maxInputBiomarkers];
    static String [] numberProteinInteractions= new String[maxInputBiomarkers];
    static String [] retrievedGenes = new String[maxInputBiomarkers];
    static String [][] interactions = new String [maxLines][2];
    static String [][] retrievedInteractions= new String [maxLines][2];
    static String [][] retrievedPathways= new String [maxLines][maxFeatures];
    static String [][] GOresults = new String[38][6];
    static String [][] sample = new String[maxLines][maxFeatures];
    public static int numberSamples = 0;

    public static int numberBiomarkers = 0;
    public static int numInteractions = 0;

    static boolean [] interactionRetrieved = new boolean [maxLines];
    public static int numberFeatures = 0;

    /**
     * @param args the command line arguments
     */

    public static void main(String[] args) {
        // TODO code application logic here
    }

    /** procedure for opening file with interaction data, and search/retrieval of interactions
     * containing biomarkers stored in "biomarkers.txt"

    public static void fileReader(File inputFile) {
```

```

int linecounter = 0;
String str = new String("");
String entry [] = new String[maxLines];
BufferedReader in = null;

if (inputFile.getName() != null) {

    try {
        in = new BufferedReader(new FileReader(inputFile));
    }

    catch (Exception e1) {
        e1.printStackTrace();
    }
}

try {
    while ( (str = in.readLine()) != null) {
        entry[linecounter] = str;
        linecounter++;
    }
    numberSamples = linecounter;
    in.close();

} //try

catch (Exception e1) {
    e1.printStackTrace();
}

String tempTok = new String();

for(int i=0; i<numberSamples; i++)
{
    StringTokenizer st = new StringTokenizer(entry[i], "\t");
    int numberTok = 0;
    while (st.hasMoreTokens()) {
        tempTok = st.nextToken();
        if (!tempTok.equals(null))
            if((!tempTok.equals("\t") || (!tempTok.equals("\n")) || (!tempTok.equals(" "))))
                sample[i][numberTok] = tempTok;
        numberTok++;
    }
}

}

openBiomarkerFile();
searchInteractions();

```

```
}
```

```
/* search/retrieval of interactions
```

```
public static void searchInteractions()
```

```
{
```

```
    numInteractions = 0;
```

```
    for (int i = 0; i < numberBiomarkers; i++)
```

```
    {
```

```
        for (int j = 0; j < numberSamples; j++)
```

```
        {
```

```
            if((biomarkers[i].equals(sample[j][0])) ||
```

```
            (biomarkers[i].equals(sample[j][1])))
```

```
            {
```

```
                if(!interactionRetrieved[j])
```

```
                {
```

```
                    retrievedInteractions[numInteractions][0]=
```

```
                    sample[j][0];
```

```
                    retrievedInteractions[numInteractions][1]=
```

```
                    sample[j][1];
```

```
                    interactionRetrieved[j]=true;
```

```
                    numInteractions++;
```

```
                }
```

```
            }
```

```
        } // for j
```

```
    } // for i
```

```
}
```

```
public static void saveRetrievedInteractions (File inputFile){
```

```
    BufferedWriter out = null;
```

```
    if (inputFile != null) {
```

```
        try {
```

```
            out = new BufferedWriter(new FileWriter(inputFile));
```

```
            PrintWriter outputFile = new PrintWriter(out);
```

```
            outputFile.println("num inter:" + numInteractions);
```

```
            outputFile.println("num biomar:" + numberBiomarkers);
```

```
            outputFile.println("num inter ori:" + numberSamples);
```

```
            for (int i=0; i<numInteractions; i++)
```

```
            {
```

```
                for (int j=0; j<2; j++)
```

```
                {
```

```
                    if(retrievedInteractions[i][j]!= null)
```

```
                    outputFile.print(retrievedInteractions[i][j]);
```

```

        Double value = new Double (sample[i][j]);
        if (value.doubleValue() > 0.0) outputFile.print(value*1000);
        outputFile.print("\t");
    }
    outputFile.println();

}

    outputFile.close();
} //try

    catch (IOException e1) {
        e1.printStackTrace();
    }

}

}

```

```

public static void openBiomarkerFile (){

    int linecounter = 0;
    String str = new String("");
    String entry [] = new String[maxLines];
    BufferedReader in = null;
    File inputFile = new File ("biomarkers.txt");
    if (inputFile.getName() != null) {
        try {
            in = new BufferedReader(new FileReader(inputFile));
        }
        catch (Exception e1) {
            e1.printStackTrace();
        }
    }

    try {
        while ( (str = in.readLine()) != null) {
            entry[linecounter] = str;
            linecounter++;
        }
        numberBiomarkers=linecounter;
        in.close();

    } //try

    catch (Exception e1) {
        e1.printStackTrace();
    }

    String tempTok = new String();

```

```

for(int i=0; i<numberBiomarkers; i++)
{
    StringTokenizer st = new StringTokenizer(entry[i], "\t");
    int numberTok = 0;
    while (st.hasMoreTokens()) {
        tempTok = st.nextToken();
        if (!tempTok.equals(null))
            if ((!tempTok.equals("\t") || (!tempTok.equals("\n")) || (!tempTok.equals(" "))))
                biomarkers[i] = tempTok;
        numberTok++;
    }
}

}

/* procedure for saving results

public static void fileSaver(File inputFile) {

    BufferedWriter out = null;
    if (inputFile != null) {
        try {
            out = new BufferedWriter(new FileWriter(inputFile));
            PrintWriter outputFile = new PrintWriter(out);

            for (int i=0; i<numberSamples; i++)
            {
                for (int j=0; j<numberFeatures; j++)
                {
                    if(sample[i][j]!= null)
                        outputFile.print(sample[i][j]);
                    if (value.doubleValue() > 0.0) outputFile.print(value*1000);
                    outputFile.print("\t");
                }
                outputFile.println();

            }
            outputFile.close();

        } //try

        catch (IOException e1) {
            e1.printStackTrace();
        }

    }

}

*/

```

```
saveRetrievedInteractions(inputFile);  
}  
}
```