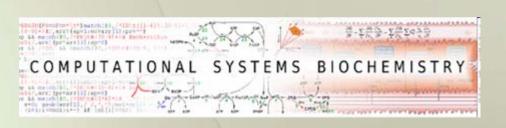




Linking transcription and metabolism

Infer metabolic changes from RNA expression

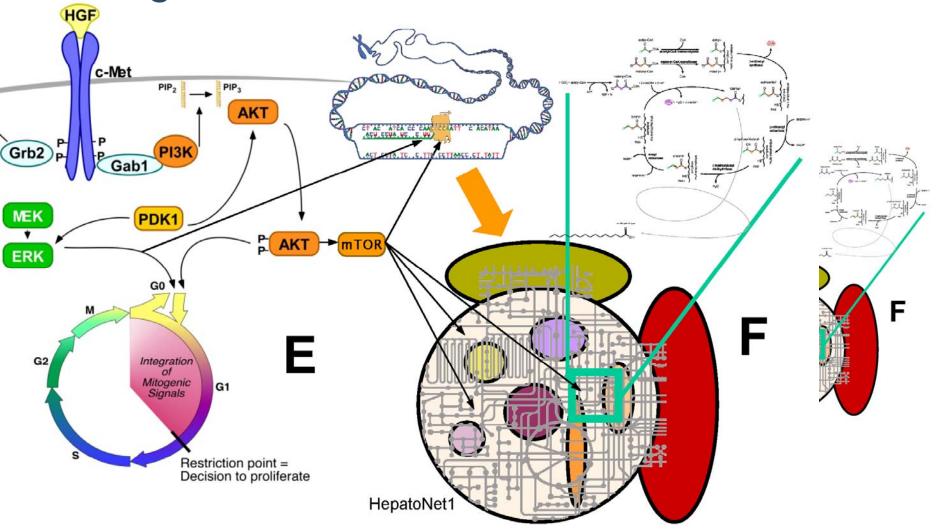
Andreas Hoppe, Charité Universtitätsmedizin Berlin Computational systems biochemistry group







Integrative model





Questions

- a) Which metabolic functions are strongly affected by HGF?
- b) What are the key regulation points?
- c) What is the metabolic basis for increased lipid production?

Data: RNA profiles by time/treatment

Approach: ModeScore

virtual liver

RNA-metabolism chain



RNA-metabolism chain

Specific degradation Allosteric effects

Complex forming
Specific degradation

Metabolic channeling

Metabolite concentration changes



RNA



Protein 🖚



Enzymatic activity



Flux rate

Specific degradation

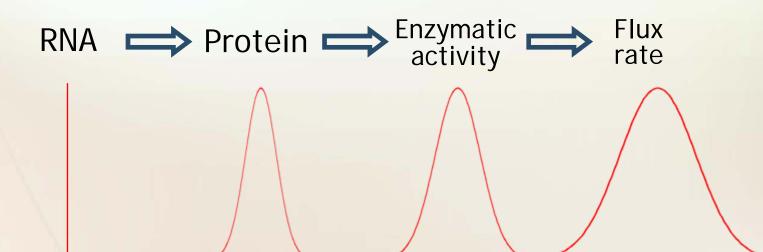
Translation regulation

Inhibition Activation Enzyme kinetics



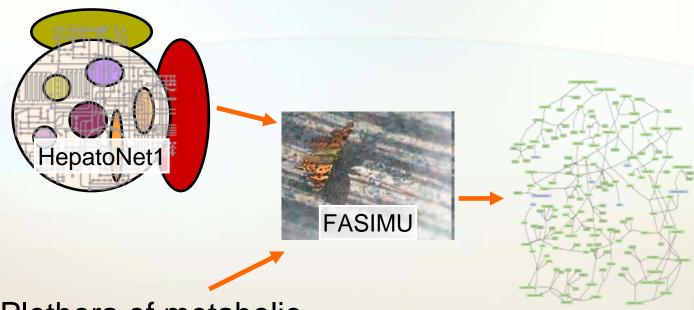
RNA-metabolism chain

- Probability field
- Normal distribution





Representative flux modes



Plethora of metabolic functions

Reference flux modes

HepatoN et 1 ... , Gille et al., 2010, Mol Syst Biol FASIMU ... Hoppe et al., 2011, BMC Bioinf

virtual liver

- a) Function change prediction
- matches relative RNA profile with reference mode
- predicts amplitude of regulation
- gives confidence score
- Ranking list (by amplitude)

Data from Ilkavets, Dooley

Simulation			control			TGFβ	
Gluconeogen. from Glycerol Aerobic ATP rephosph (FA) 3 -6.75 0.28 4 -8.68 0.31	Simulation	rank			rank		score
Gluconeogen. from Glycerol Aerobic ATP rephosph (FA) 3 -6.75 0.28 4 -8.68 0.31	Anserobic rephosph of ATP	2	-8 74	0.31	1	-12.6	0.29
Aerobic ATP rephosph (FA) Galactose degr Complete oxidation of oleate Ethanol(s) degr min Aerobic reduction of NADP+ Triacylglycerol(r) synt phys Gly-CD-cholate(b) phys Lysine degr Urea from alanine Clycocholate(b) synt phys Clycocholate(b) synt min C							
Calactose degr							
Complete oxidation of oleate Financial State		5			5	-8.13	
Ethanol(s) degr min 68		7	-5.31	0.28	6	-7.2	0.33
Aerobic reduction of NADP+ Triacylglycerol(r) synt phys Gly-CD-cholate(b) synt min Gly-CD-cholate(b) synt min Gly-CD-cholate(c) synt min Gly-CD-cholate(b) synt phys Gly-CD-cholate(b) synt synt synt synt synt synt synt synt		68		0.24	2	-11.5	0.25
Cally-CD-cholate(b) phys 6 -5.41 0.27 10 -4.59 0.28	1, 4	8	-5.25	0.24	7	-6.34	0.26
Cly-CD-cholate(b) phys	Triacylglycerol(r) synt phys	1	-10.6	0.23	64	-0.97	0.29
Lysine degr 10 -3.91 0.22 14 -4.22 0.28 Urea from alanine 12 -3.04 0.25 11 -4.52 0.28 Glycocholate(b) synt phys 20 -2.04 0.26 12 -4.42 0.27 LDL(s) degr phys 9 -4.12 0.2 37 -1.89 0.24 Alanine(c) synt min 49 -0.83 0.31 9 -4.61 0.31 Taurocholate(b) synt phys 17 -2.25 0.27 16 -3.66 0.28 ATP salvage from Adenosine 72 -0.08 0.21 8 -5.08 0.23 CTP(c) synt min 15 -2.41 0.21 23 -2.62 0.24 Acetoacetate 19 -2.19 0.35 19 -2.79 0.36 (R)-3-Hydroxybutanoate 21 -2.02 0.4 21 -2.66 0.43 THF(c) synt min 74 -0.03 0.33 13 -4.34 0.25		6	-5.41	0.27	10	-4.59	0.28
Clycocholate(b) synt phys 20 -2.04 0.26 12 -4.42 0.27	1,7-2	10	-3.91	0.22	14	-4.22	0.28
LDL(s) degr phys 9	Urea from alanine	12	-3.04	0.25	11	-4.52	0.28
LDL(s) degr phys	Glycocholate(b) synt phys	20	-2.04	0.26	12	-4.42	0.27
Taurocholate(b) synt phys ATP salvage from Adenosine CTP(c) synt min 15	1,,	9	-4.12	0.2	37	-1.89	0.24
ATP salvage from Adenosine 72	Alanine(c) synt min	49	-0.83	0.31	9	-4.61	0.31
CTP(c) synt min 15 -2.41 0.21 23 -2.62 0.24 Acetoacetate 19 -2.19 0.35 19 -2.79 0.36 (R)-3-Hydroxybutanoate 21 -2.02 0.4 21 -2.66 0.43 THF(c) synt min 74 -0.03 0.33 13 -4.34 0.25 Aspartate degr 27 -1.59 0.24 22 -2.66 0.28 Gluconeogen. from Lactate 42 -1.09 0.26 18 -2.93 0.33 Methionine degr 13 -2.86 0.25 57 -1.13 0.32 Stearate(c) synt min 38 -1.23 0.35 20 -2.69 0.37 ATP salvage from Hypoxanthine 86 0.36 0.21 15 -4.21 0.23 Bilirubin conjugation 11 -3.45 0.23 88 -0.15 0.26 UTP(c) synt min 30 -1.51 0.2 33 -2.08 0.27 <tr< td=""><td>Taurocholate(b) synt phys</td><td>17</td><td>-2.25</td><td>0.27</td><td>16</td><td>-3.16</td><td>0.28</td></tr<>	Taurocholate(b) synt phys	17	-2.25	0.27	16	-3.16	0.28
Acetoacetate	ATP salvage from Adenosine	72	-0.08	0.21	8	-5.08	0.23
(R)-3-Hydroxybutanoate 21 -2.02 0.4 21 -2.66 0.43 THF(c) synt min 74 -0.03 0.33 13 -4.34 0.25 Aspartate degr 27 -1.59 0.24 22 -2.66 0.28 Gluconeogen. from Lactate 42 -1.09 0.26 18 -2.93 0.33 Methionine degr 13 -2.86 0.25 57 -1.13 0.32 Stearate(c) synt min 38 -1.23 0.35 20 -2.69 0.37 ATP salvage from Hypoxanthine 86 0.36 0.21 15 -4.21 0.23 Bilirubin conjugation 11 -3.45 0.23 88 -0.15 0.26 UTP(c) synt min 30 -1.51 0.2 33 -2.08 0.27 udpglcur(c) synt min 24 -1.74 0.22 39 -1.82 0.26 Gluconeogen. from Alanine 45 -0.96 0.31 24 -2.59 0.35	CTP(c) synt min	15	-2.41	0.21	23	-2.62	0.24
THF(c) synt min Aspartate degr Aspartate degr Gluconeogen. from Lactate 42	Acetoacetate	19	-2.19	0.35	19	-2.79	0.36
Aspartate degr Gluconeogen. from Lactate 42 -1.09 0.26 18 -2.93 0.33 Methionine degr 13 -2.86 0.25 57 -1.13 0.32 Stearate(c) synt min 38 -1.23 0.35 20 -2.69 0.37 ATP salvage from Hypoxanthine Bilirubin conjugation 11 -3.45 0.23 88 -0.15 0.26 UTP(c) synt min 30 -1.51 0.2 33 -2.08 0.27 udpglcur(c) synt min 24 -1.74 0.22 39 -1.82 0.26 Gluconeogen. from Alanine 45 -0.96 0.31 24 -2.59 0.35 cmpneuNAc(n) synt min 26 -1.62 0.25 36 -1.9 0.25 udpgal(c) synt min 27 -1.70 0.23 40 -1.75 0.27 NH3 degr 40 -1.17 0.25 35 -2.01 0.32 Pyridoxal-P(c) synt min 46 -2.27 0.25 75 -0.76 0.32 UDP-glucose(c) synt min 47 -1.27 0.22 41 -1.73 0.27 Creatine(c) synt min 48 -2.98 0.25 Creatine(c) synt min 49 -1.56 0.34	(R)-3-Hydroxybutanoate	21	-2.02	0.4	21	-2.66	0.43
Gluconeogen. from Lactate 42 -1.09 0.26 18 -2.93 0.33 Methionine degr 13 -2.86 0.25 57 -1.13 0.32 Stearate(c) synt min 38 -1.23 0.35 20 -2.69 0.37 ATP salvage from Hypoxanthine 86 0.36 0.21 15 -4.21 0.23 Bilirubin conjugation 11 -3.45 0.23 88 -0.15 0.26 UTP(c) synt min 30 -1.51 0.2 33 -2.08 0.27 udpglcur(c) synt min 24 -1.74 0.22 39 -1.82 0.26 Gluconeogen. from Alanine 45 -0.96 0.31 24 -2.59 0.35 cmpneuNAc(n) synt min 26 -1.62 0.25 36 -1.9 0.25 udpgal(c) synt min 25 -1.7 0.23 40 -1.75 0.27 NH3 degr 40 -1.17 0.25 35 -2.01 0.32	THF(c) synt min	74	-0.03	0.33	13	-4.34	0.25
Methionine degr 13 -2.86 0.25 57 -1.13 0.32 Stearate(c) synt min 38 -1.23 0.35 20 -2.69 0.37 ATP salvage from Hypoxanthine 86 0.36 0.21 15 -4.21 0.23 Bilirubin conjugation 11 -3.45 0.23 88 -0.15 0.26 UTP(c) synt min 30 -1.51 0.2 33 -2.08 0.27 udpglcur(c) synt min 24 -1.74 0.22 39 -1.82 0.26 Gluconeogen. from Alanine 45 -0.96 0.31 24 -2.59 0.35 cmpneuNAc(n) synt min 26 -1.62 0.25 36 -1.9 0.25 udpgal(c) synt min 25 -1.7 0.23 40 -1.75 0.27 NH3 degr 40 -1.17 0.25 35 -2.01 0.32 Pyridoxal-P(c) synt min 73 -0.03 0.21 17 -3.13 0.23 </td <td>Aspartate degr</td> <td>27</td> <td>-1.59</td> <td>0.24</td> <td>22</td> <td>-2.66</td> <td>0.28</td>	Aspartate degr	27	-1.59	0.24	22	-2.66	0.28
Stearate(c) synt min 38 -1.23 0.35 20 -2.69 0.37 ATP salvage from Hypoxanthine 86 0.36 0.21 15 -4.21 0.23 Bilirubin conjugation 11 -3.45 0.23 88 -0.15 0.26 UTP(c) synt min 30 -1.51 0.2 33 -2.08 0.27 udpglcur(c) synt min 24 -1.74 0.22 39 -1.82 0.26 Gluconeogen. from Alanine 45 -0.96 0.31 24 -2.59 0.35 cmpneuNAc(n) synt min 26 -1.62 0.25 36 -1.9 0.25 udpgal(c) synt min 25 -1.7 0.23 40 -1.75 0.27 NH3 degr 40 -1.17 0.25 35 -2.01 0.32 Pyridoxal-P(c) synt min 73 -0.03 0.21 17 -3.13 0.23 Asparagine(c) synt min 16 -2.27 0.25 75 -0.76 0.32	Gluconeogen. from Lactate	42	-1.09	0.26	18	-2.93	0.33
ATP salvage from Hypoxanthine 86 0.36 0.21 15 -4.21 0.23 Bilirubin conjugation 11 -3.45 0.23 88 -0.15 0.26 UTP(c) synt min 30 -1.51 0.2 33 -2.08 0.27 udpglcur(c) synt min 24 -1.74 0.22 39 -1.82 0.26 Gluconeogen. from Alanine 45 -0.96 0.31 24 -2.59 0.35 cmpneuNAc(n) synt min 26 -1.62 0.25 36 -1.9 0.25 udpgal(c) synt min 25 -1.7 0.23 40 -1.75 0.27 NH3 degr 40 -1.17 0.25 35 -2.01 0.32 Pyridoxal-P(c) synt min 73 -0.03 0.21 17 -3.13 0.23 Asparagine(c) synt min 16 -2.27 0.25 75 -0.76 0.32 UDP-glucose(c) synt min 37 -1.27 0.22 41 -1.73 0.27 </td <td>Methionine degr</td> <td>13</td> <td>-2.86</td> <td>0.25</td> <td>57</td> <td>-1.13</td> <td>0.32</td>	Methionine degr	13	-2.86	0.25	57	-1.13	0.32
Bilirubin conjugation 11 -3.45 0.23 88 -0.15 0.26 UTP(c) synt min 30 -1.51 0.2 33 -2.08 0.27 udpglcur(c) synt min 24 -1.74 0.22 39 -1.82 0.26 Gluconeogen. from Alanine 45 -0.96 0.31 24 -2.59 0.35 cmpneuNAc(n) synt min 26 -1.62 0.25 36 -1.9 0.25 udpgal(c) synt min 25 -1.7 0.23 40 -1.75 0.27 NH3 degr 40 -1.17 0.25 35 -2.01 0.32 Pyridoxal-P(c) synt min 73 -0.03 0.21 17 -3.13 0.23 Asparagine(c) synt min 16 -2.27 0.25 75 -0.76 0.32 UDP-glucose(c) synt min 32 -1.38 0.25 44 -1.56 0.34	Stearate(c) synt min	38			20	-2.69	
UTP(c) synt min udpglcur(c) synt min 24 -1.74 0.22 39 -1.82 0.26 Gluconeogen. from Alanine 45 -0.96 0.31 24 -2.59 0.35 cmpneuNAc(n) synt min 26 -1.62 0.25 36 -1.9 0.25 udpgal(c) synt min 25 -1.7 0.23 40 -1.75 0.27 NH3 degr Pyridoxal-P(c) synt min 73 -0.03 0.21 17 -3.13 0.23 Asparagine(c) synt min 16 -2.27 0.25 75 -0.76 0.32 UDP-glucose(c) synt min 37 -1.27 0.22 41 -1.73 0.27 Creatine(c) synt min 30 -1.51 0.2 33 -2.08 0.27 24 -2.59 0.35 -1.9 0.25 0.27 0.25 35 -2.01 0.32 0.27 0.28 40 -1.75 0.22 0.29 41 -1.73 0.27 0.29 41 -1.73 0.27	ATP salvage from Hypoxanthine	86	0.36		15	-4.21	
udpglcur(c) synt min 24 -1.74 0.22 39 -1.82 0.26 Gluconeogen. from Alanine 45 -0.96 0.31 24 -2.59 0.35 cmpneuNAc(n) synt min 26 -1.62 0.25 36 -1.9 0.25 udpgal(c) synt min 25 -1.7 0.23 40 -1.75 0.27 NH3 degr 40 -1.17 0.25 35 -2.01 0.32 Pyridoxal-P(c) synt min 73 -0.03 0.21 17 -3.13 0.23 Asparagine(c) synt min 16 -2.27 0.25 75 -0.76 0.32 UDP-glucose(c) synt min 37 -1.27 0.22 41 -1.73 0.27 Creatine(c) synt min 32 -1.38 0.25 44 -1.56 0.34		11	-3.45		88	-0.15	
Gluconeogen. from Alanine cmpneuNAc(n) synt min 26 -1.62 0.25 36 -1.9 0.25 udpgal(c) synt min 25 -1.7 0.23 40 -1.75 0.27 NH3 degr 40 -1.17 0.25 35 -2.01 0.32 Pyridoxal-P(c) synt min 73 -0.03 0.21 17 -3.13 0.23 Asparagine(c) synt min 16 -2.27 0.25 75 -0.76 0.32 UDP-glucose(c) synt min 37 -1.27 0.22 41 -1.73 0.27 Creatine(c) synt min 32 -1.38 0.25 44 -1.56 0.34	1 7 2	30			33		
cmpneuNAc(n) synt min 26 -1.62 0.25 36 -1.9 0.25 udpgal(c) synt min 25 -1.7 0.23 40 -1.75 0.27 NH3 degr 40 -1.17 0.25 35 -2.01 0.32 Pyridoxal-P(c) synt min 73 -0.03 0.21 17 -3.13 0.23 Asparagine(c) synt min 16 -2.27 0.25 75 -0.76 0.32 UDP-glucose(c) synt min 37 -1.27 0.22 41 -1.73 0.27 Creatine(c) synt min 32 -1.38 0.25 44 -1.56 0.34	1, , ,	24	-1.74		39	-1.82	
udpgal(c) synt min 25 -1.7 0.23 40 -1.75 0.27 NH3 degr 40 -1.17 0.25 35 -2.01 0.32 Pyridoxal-P(c) synt min 73 -0.03 0.21 17 -3.13 0.23 Asparagine(c) synt min 16 -2.27 0.25 75 -0.76 0.32 UDP-glucose(c) synt min 37 -1.27 0.22 41 -1.73 0.27 Creatine(c) synt min 32 -1.38 0.25 44 -1.56 0.34		45			24		
NH3 degr 40 -1.17 0.25 35 -2.01 0.32 Pyridoxal-P(c) synt min 73 -0.03 0.21 17 -3.13 0.23 Asparagine(c) synt min 16 -2.27 0.25 75 -0.76 0.32 UDP-glucose(c) synt min 37 -1.27 0.22 41 -1.73 0.27 Creatine(c) synt min 32 -1.38 0.25 44 -1.56 0.34	1,7,2				36		
Pyridoxal-P(c) synt min 73 -0.03 0.21 17 -3.13 0.23 Asparagine(c) synt min 16 -2.27 0.25 75 -0.76 0.32 UDP-glucose(c) synt min 37 -1.27 0.22 41 -1.73 0.27 Creatine(c) synt min 32 -1.38 0.25 44 -1.56 0.34						-1.75	
Asparagine(c) synt min 16 -2.27 0.25 75 -0.76 0.32 UDP-glucose(c) synt min 37 -1.27 0.22 41 -1.73 0.27 Creatine(c) synt min 32 -1.38 0.25 44 -1.56 0.34			-1.17			-2.01	
UDP-glucose(c) synt min 37 -1.27 0.22 41 -1.73 0.27 Creatine(c) synt min 32 -1.38 0.25 44 -1.56 0.34							
Creatine(c) synt min 32 -1.38 0.25 44 -1.56 0.34	- 1, , -						
1,7 =		1					
Histidina degr. 18 _9 91 0.95 77 _0.79 0.90	1 / -	1					
	Histidine degr	18	-2.21	0.25	77	-0.72	0.29
Arginine(c) synt min 34 -1.35 0.35 43 -1.57 0.4	. , , -						
Glutamate degr 14 -2.75 0.21 87 -0.17 0.3		1					
udpgalnac(c) synt min 31 -1.43 0.23 45 -1.47 0.25	udpgalnac(c) synt min	31	-1.43	0.23	45	-1.47	0.25



b) Regulation points

- Contribution of individual reactions
- Expression difference of individual genes
- → Prediction of regulation points

Glyconeogenesis from Glycerol

Rea	Scre	Impct	Expr	Expr	Expr	Expr	Ref	Wght	Reaction
ID	0.42			C1D	C3D	*-0.16	value		
r025	5 1	0.51	-5.07	8.86	3.79	0.8	0.77	0.88	$DHAP(c) + GAP(c) \rightleftharpoons Fructose-1,6PP(c)$
r024	0.94	0.46	-5.77	18.5	12.7	0.91	0.77	0.88	$DHAP(c) \rightleftharpoons GAP(c)$
r247	0.9	0.42	-3.77	4.72	0.95	0.59	0.77	0.88	$Glucose\text{-}6P(c) + Pi(r) \to Glucose\text{-}6P(r) + Pi(c)$
r001	7 0.12	-0.37	0.19	7.35	7.53	-0.03	0.77	1.24	$O2(m) + 4 H+(PG)(m) + 4 Ferrocytochrome C(m) \rightarrow 4 Ferricytochrome C(m) + 2 H2O(m) + 4 H+(PG)(c)$
r050	7 0.13	-0.36	0.13	7.93	8.06	-0.02	1.54	1.24	Ubiquinol(m) + 2 Ferricytochrome $C(m) + 4 H+(PG)(m) \rightarrow Ubiquinone(m) + 4 H+(PG)(c) + 2 Ferrocytochrome C(m)$
r019	2 0.82	0.35	-6.49	17	10.5	1.02	0.77	0.88	$Fructose-6P(c) \rightleftharpoons Glucose-6P(c)$
r020	0.14	-0.35	-0.003	1.75	1.74	0.001	1.54	1.24	$Ubiquinone(m) + sn-Glycerol-3P(c) \rightleftharpoons DHAP(c) + Ubiquinol(m)$
r0019	0.14	-0.34	-0.05	0.69	0.64	0.01	1.54	1.24	$ADP(m) + Pi(m) + 3 H+(PG)(c) \rightarrow ATP(m) + H2O(m) + 3 H+(PG)(m)$
r020	0.14	-0.34	-0.14	0.69	0.55	0.02	1.54	1.24	$ATP(c) + Glycerol(c) \rightleftharpoons ADP(c) + sn-Glycerol-3P(c)$
r048	7 0.75	0.29	-3.06	4.99	1.93	0.48	0.77	0.88	$Fructose\text{-}1,6PP(c) + H2O(c) \to Fructose\text{-}6P(c) + Pi(c)$
r039	5 0.2	-0.19	-0.53	0.89	0.36	0.08	0.77	0.88	$H2O(r) + Glucose-6P(r) \rightarrow Glucose(r) + Pi(r)$
r104	7 0.09	-0.18	1.08	16.7	17.8	-0.17	1.54	0.56	$Pi(c) + H+(PG)(c) \rightarrow Pi(m) + H+(PG)(m)$
	1 0.47			2.86				0.88	$Glucose(r) \rightarrow Glucose(c)$
r103	0.47	0.04	-1.88	2.86	0.97	0.3	0.77	0.88	$Glucose(c) \rightleftharpoons Glucose(s)$

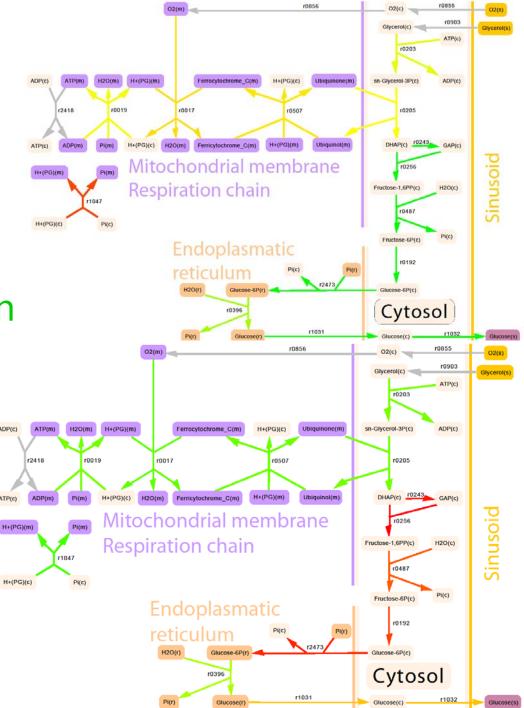
virtual liver Glyconeogenesis from Glycerol

Expression change:

up / unchanged / down

Score:

high / medium / low



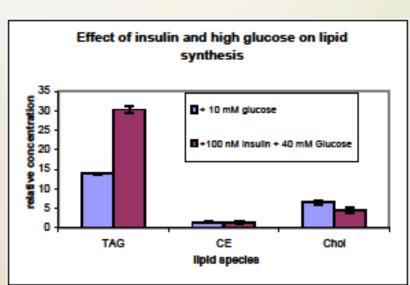


c) Lipid metabolism

- Increased lipid formation (confirmed)
- Focused analysis of lipid-related functions
- Alternative flux scenarios

Wet lab analysis: Gebhardt, Schiller,

Shevchenko





Summary/Outlook

- ModeScore: Quantitative assessment of metabolic regulation by RNA profiles
- Allows comparative analysis of HGF effect
- In silico predictions
- Focus on lipid metabolism: testing flux scenarios with RNA profiles