ModeScore: approach to infer metabolic activity changes from expression data

Andreas Hoppe[1], Iryna Ilkavets[2], Hauke Busch[3], Stefanie Müller[4], Melanie Boerries[3], Svantje Braun[4], Lorenza D'Alessandro[4], Ursula Klingmueller[4], Steven Dooley[2], Hermann-Georg Holzhütter[1]

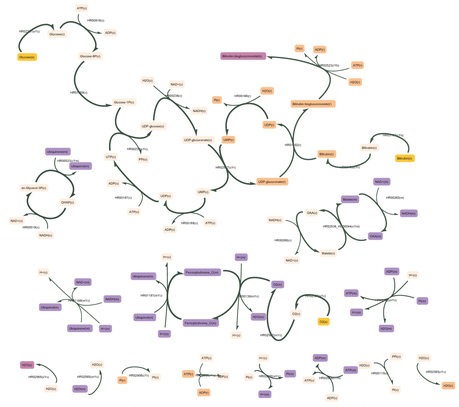
[1]Computational Systems Biochemistry, Institute of Biochemistry, Charité, Universitätsmedizin Berlin, [2]Department of Medicine II, Section Molecular Hepatology, Medical Faculty Mannheim, Heidelberg University, [3]Institute of Molecular Medicine and Cell Research, Albert-Ludwigs-University Freiburg, [4]Deutsches Krebsforschungszentrum Heidelberg, Helmholtz-Gemeinschaft

Background. Full-genome expression profiles for different cell systems under many conditions are available and their interpretation with respect to the metabolism is a major challenge in systems biology. Current approaches are based on the classification of the transcript values in on/off. It turns out that this distinction is particularly difficult for metabolic genes as they may show a relatively high basal RNA expression when not active. Oppositely, several long-lived enzymes show low RNA expression levels although undoubtedly active.

Results. An approach is presented to predict activity changes of metabolic functions by scoring reference flux distributions. Rather than attempting to predict fluxes in a metabolic system which is hard to validate, reference flux distributions obtained with flux-balance computations are the scaffold to interpret transcript changes for metabolic genes. Compared with the annotation-based approach ModeScore relates transcript changes directly to metabolic functions, thus, provides testable hypotheses on the level of cellular function. The process of the algorithm will be demonstrated in time-course study of the effects of the cytokines HGF, TGF, and IL6 on cultured primary mouse hepatocytes.

In conclusion, the novel method provides an enrichment for the most promising functions together with the genes it is based upon for further metabolic analysis solely based on transcript profiles.

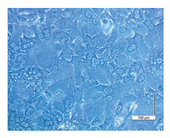
Reference flux distributions (modes)



Plethora of metabolic functions (992), three categories:
- Regeneration of important intemediates (72)
- Function of organismic duty (379)
- Synthesis and degradation of cellular constituents (541)
- Computation with FASIMU [5]

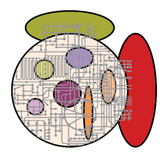
Data.

Full genome RNA transcript profiles (Affymetrix 430.2) of primary mouse hepatocytes cultured on collagen monolayer. Various time points, TGFβ, HGFα, IL6 stimulation.



Network.

HepatoNet1b, manually curated network of the human hepatocyte [4], refined to cover more functions, comprises 1500 localized metabolic species, 2702 reactions, 879 annotated genes [1].

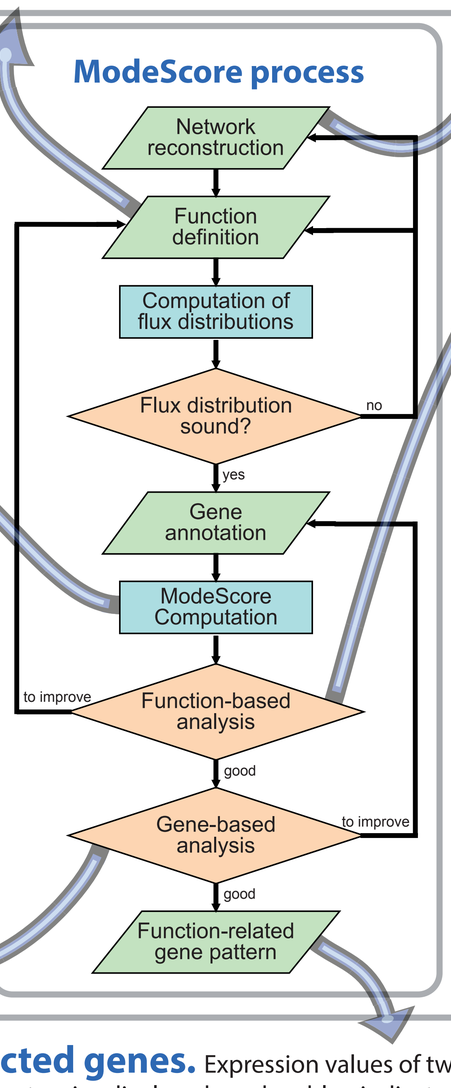


Function ranks by amplitude

Relative expression scores of the third time point (24h) with respect to the first (1h) for the control and TGFb series sorted by the sum of amplitudes (inverse of scaling factor l). Only the most down-regulated shown.

Simulation	control 1h/24h rank	ampl	score	TGFβ/control 24h rank	ampl	score
Aspartate degr	1	-4.7	0.32	23	-0.9	0.34
Asparagine degr	2	-4.6	0.38	21	-0.98	0.34
Proline degr	3	-4.6	0.25	72	-0.51	0.5
Taurine from Cysteine	6	-2.99	0.49	8	-1.46	0.5
Tyrosine	21	-1.51	0.65	2	-2.86	0.65
Gluconeogen from Alanine	4	-3.52	0.4	51	-0.63	0.35
Ethanol degr	65	-0.52	0.64	1	-3.56	0.54
Phenylalanine degr	5	-3.23	0.41	36	-0.76	0.43
Gluconeogen from Glycerol	7	-2.98	0.47	47	-0.69	0.51
Gluconeogen from Lactate	8	-2.54	0.39	62	-0.57	0.51
Arachidonate from Dihomo-gamma-linolenate	11	-1.84	0.39	30	-0.85	0.6

ModeScore process



ModeScore computation.

[2] Flux distributions computed in the stoichiometric network of the hepatocyte's metabolism [2] for a plethora of metabolic functions are matched to transcript changes with a novel method combining the scoring approach [3] to complex functional flux distributions [4], implemented in [5].

$$\mathrm{Score}(M_k, V) = \frac{\sum_{i\in I_k} w_i \mathrm{score}_i(m_i, v_i)}{\sum_{i\in I_k} w_i} \quad \text{total score}$$

$$I_k = \{i \,|\, m_i \neq 0\} \quad \text{indices of nonzero values}$$

$$w_i = \sqrt{|m_i|\omega_i} \quad \text{weights}$$

$$\omega_i \geq 0 \quad \text{fixed weight adjustment}$$

$$\mathrm{score}_i(m_i, v_i) = e^{-\frac{1}{2}\left(2\frac{\lambda v_i - m_i}{|m_i|}\right)^2} \quad \text{score component}$$

$$M_k = (m_i)_i \quad k\text{-th reference mode}$$

$$V = (v_i)_i \quad \text{relative expression profile } (\log_2)$$

$$\lambda \text{ chosen such that } \quad \mathrm{Score}(M_k, V) \text{ is maximal}$$

Detailed reaction list.

Degradation of Phenylalanine

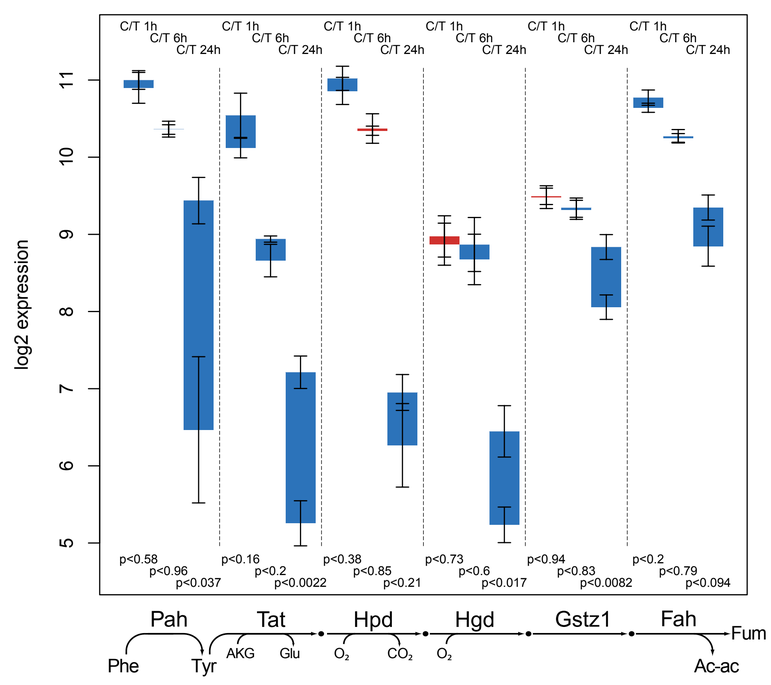
Rea ID	TGFβ/C 1h Score 0.34	Expr Δ	TGFβ/C 6h Score 0.44	Expr Δ	TGFβ/C 24h Score 0.43	Expr Δ	Wght	Reaction
r0399	0.9	-0.1	0.15	-0.005	0	-2.97	14.3	Tetrahydrobiopterin(c) + Phenylalanine(c) + O2(c) ⇌ Dihydrobiopterin(c) + Tyrosine(c) + H2O(c). L-Phenylalanine,tetrahydrobiopterin:oxygen oxidoreductase (4-hydroxylating) Phenylalanine, tyrosine and tryptophan biosynthesis EC:1.14.16.1
Pah								ENSMUSG00000020051 phenylalanine hydroxylase
r0183	0	-0.42	0.74	-0.28	0.01	-1.96	12.6	Tyrosine(c) + AKG(c) ⇌ 4-Hydroxyphenylpyruvate(c) + Glutamate(c). L-Tyrosine:2-oxoglutarate aminotransferase Tyrosine metabolism / Phenylalanine, tyrosine and tryptophan biosynthesis EC:2.6.1.5 EC:2.6.1.57
Tat								ENSMUSG00000001670 tyrosine aminotransferase
r0543	0.002	0.1	1	-0.19	0.49	-1.21	12.8	Homogentisate(c) + O2(c) ⇌ 4-Maleylacetoacetate(c). Homogentisate:oxygen 1,2-oxidoreductase (decyclizing) Tyrosine metabolism / Styrene degradation EC:1.13.11.5
Hgd								ENSMUSG00000022821 homogentisate 1, 2-dioxygenase
r0069	0.02	0.06	0	0.98	0.01	0.37	8.49	CoA(c) + Acetoacetyl-CoA(c) ⇌ 2 Acetyl-CoA(c). Acetyl-CoA:acetyl-CoA C-acetyltransferase Synthesis and degradation of ketone bodies EC:2.3.1.9 (2 genes, 5 sp.)
Acat1 (3)		0.06		1.5		0.54		ENSMUSG00000032047 acetyl-Coenzyme A acetyltransferase 1
Acat2 (2)		0.05		0.2		0.11		ENSMUSG00000023832 acetyl-Coenzyme A acetyltransferase 2
r0544	0.89	-0.16	0.07	0.03	0.98	-0.69	12.7	4-Hydroxyphenylpyruvate(c) + O2(c) ⇌ Homogentisate(c) + CO2(c). 4-Hydroxyphenylpyruvate:oxygen oxidoreductase (hydroxylating,decarboxylating) Tyrosine metabolism EC:1.13.11.27
Hpd								ENSMUSG00000029445 4-hydroxyphenylpyruvic acid dioxygenase
r0034	0.57	-0.06	0.39	-0.06	0.21	-1.43	7.02	2 ATP(m) + CO2(m) + H2O(m) + NH3(m) ⇌ 2 ADP(m) + Pi(m) + Carbamoyl-P(m). Carbon-dioxide:ammonia ligase (ADP-forming,carbamate-phosphorylating) Urea cycle and metabolism of amino groups / Nitrogen metabolism EC:6.3.4.16
Cps1								ENSMUSG00000025991 carbamoyl-phosphate synthetase 1
r0875	0	0.19	0	0.28	0.0003	0.77	8.71	Glutamate(c) + H+(PG)(c) → Glutamate(m) + H+(PG)(m). Mitochondrial Carrier (MC) TCDB:2.A.29.14.3
Slc25a22								ENSMUSG00000019082 solute carrier family 25 (mitochondrial carrier, glutamate), member 22
r0605	0.1	0.01	0.23	-0.03	1	-0.78	12.9	4-Maleylacetoacetate(c) ⇌ Fumarylacetoacetate(c). 4-Maleylacetoacetate cis-trans-isomerase Tyrosine metabolism / Styrene degradation EC:5.2.1.2
Gstz1								ENSMUSG00000021033 glutathione transferase zeta 1 (maleylacetoacetate isomerase)
shortened								

Selected genes.

Expression values of two treatment series displayed as a bar, blue indicates down-regulation, red up/regulation. Standard deviation and Welsh t-test based on the repeats.

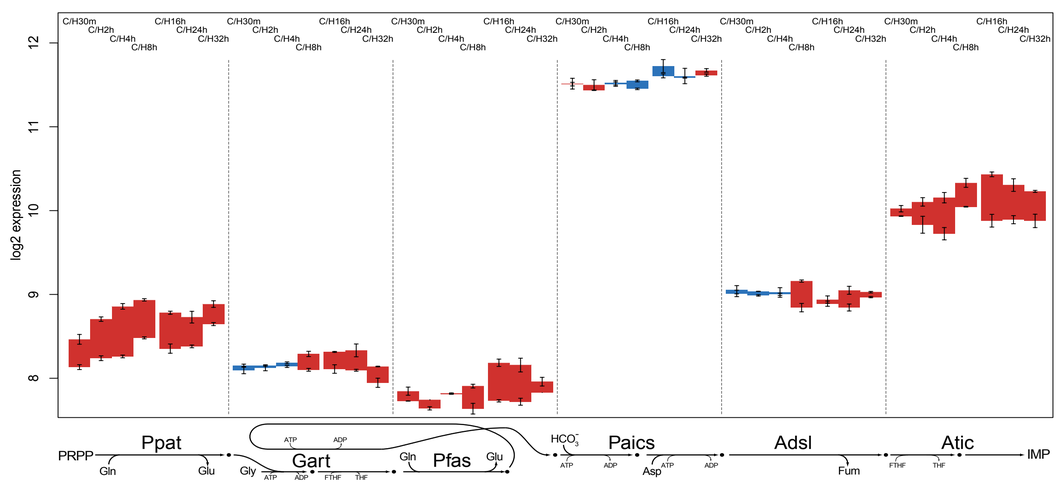
TGFβ (transforming growth factor beta)

leads to a down-regulation of many liver specific metabolic functions including the degradation of amino acids and ethanol as well as urea formation. The down-regulation of **phenylalanine degradation** is particularly strong. Only few genes are up-regulated e.g. collagens.



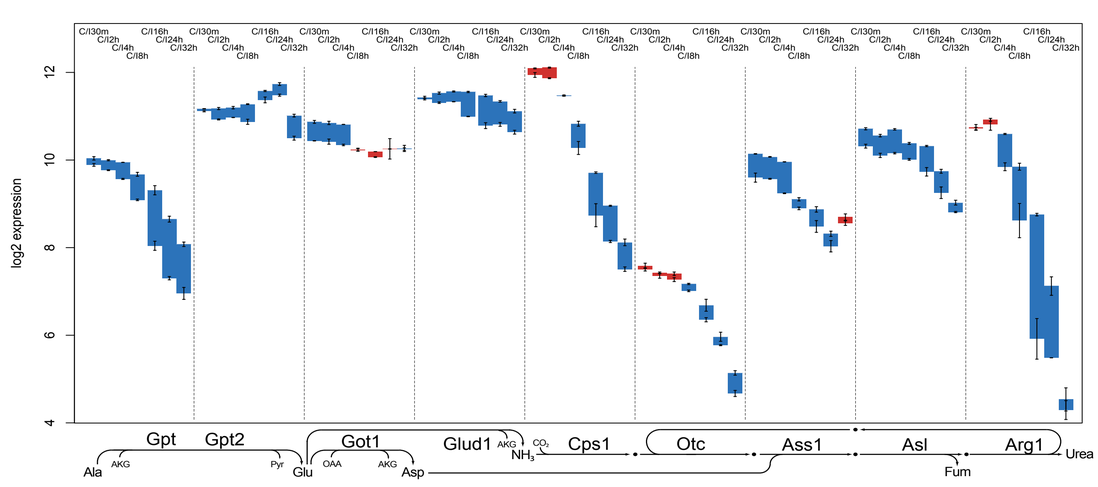
HGFα (hepatocellular growth factor alpha)

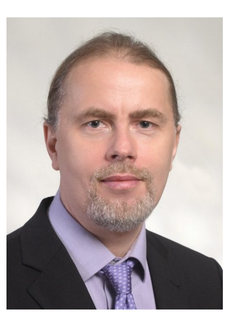
leads to a down-regulation of many liver specific metabolic functions. Genes of the synthesis of cellular constituents, such as lipids, are up-regulated. Its role in hepatocellular proliferation is highlighted by the consistent up-regulation of genes in the **purine de novo synthesis**.



IL6 (interleukin 6)

generally shows a similar response as HGFα. Among the down-regulated liver functions, IL6 shows a particularly strong down-regulation of **urea synthesis.** Among the up-regulated genes, the genes of phospholipid synthesis are specifically up-regulated by IL6.





Contact:
Dr. Andreas Hoppe
Charité, University Medicine Berlin
Institute for Biochemistry
Charitépl. 1/Virchowweg 6
10117 Berlin, Germany
Phone: +49-30-450 528 176
E-mail: **hoppe@bioinformatics.org**

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