

SUPPLEMENTARY MATERIAL

FOR

PHARMACOGENOMICS OF METABOLIC EFFECTS OF

ROSIGLITAZONE

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Pharmacogenomics 2008; 9(2), 141-155

Supplementary table 1. Primer sequences used for real-time PCR validation of microarray results.

Gene	Primer sequence
ADAM metallopeptidase domain 7	FWD 5'-CCATCTGCTTCCCTGTGT-3' REV 5'-AAGGCTTCTCCCCACTCTGT-3'
Aquaporin 9	FWD 5'-GGTTGAGGTCTCACAGTTGG-3' REV 5'-CTTCGAGTGATGCATTGGA-3'
Carnitine palmitoyltransferase 1B	FWD 5'-TTATCGAGTTCAGAACGAACG-3' REV 5'-TGGTGTGTCTCCTGGTCTCA-3'
Catechol-O-methyltransferase	FWD 5'-GAGATCTCACGGGGTTCA-3' REV 5'-TGTTATTGGCGTCTGGACA-3'
Caveolin 1	FWD 5'-GACATCTCTACAGTGTCCC-3' REV 5'-ACACGGCTGATGCACTGAAT-3'
Leptin	FWD 5'-TCCTATGTTCAAGCTGTGCC-3' REV 5'-GTGTAAACACATGGCTCT-3'
Prostaglandin D2 synthase	FWD 5'-GCTTCCACTCCCTCTCAGTG-3' REV 5'-AGAAGCTGGGCTCTGCTGTA-3'

FWD: forward primer, REV: reverse primer.

Supplementary table 2. Two-way ANOVA results.

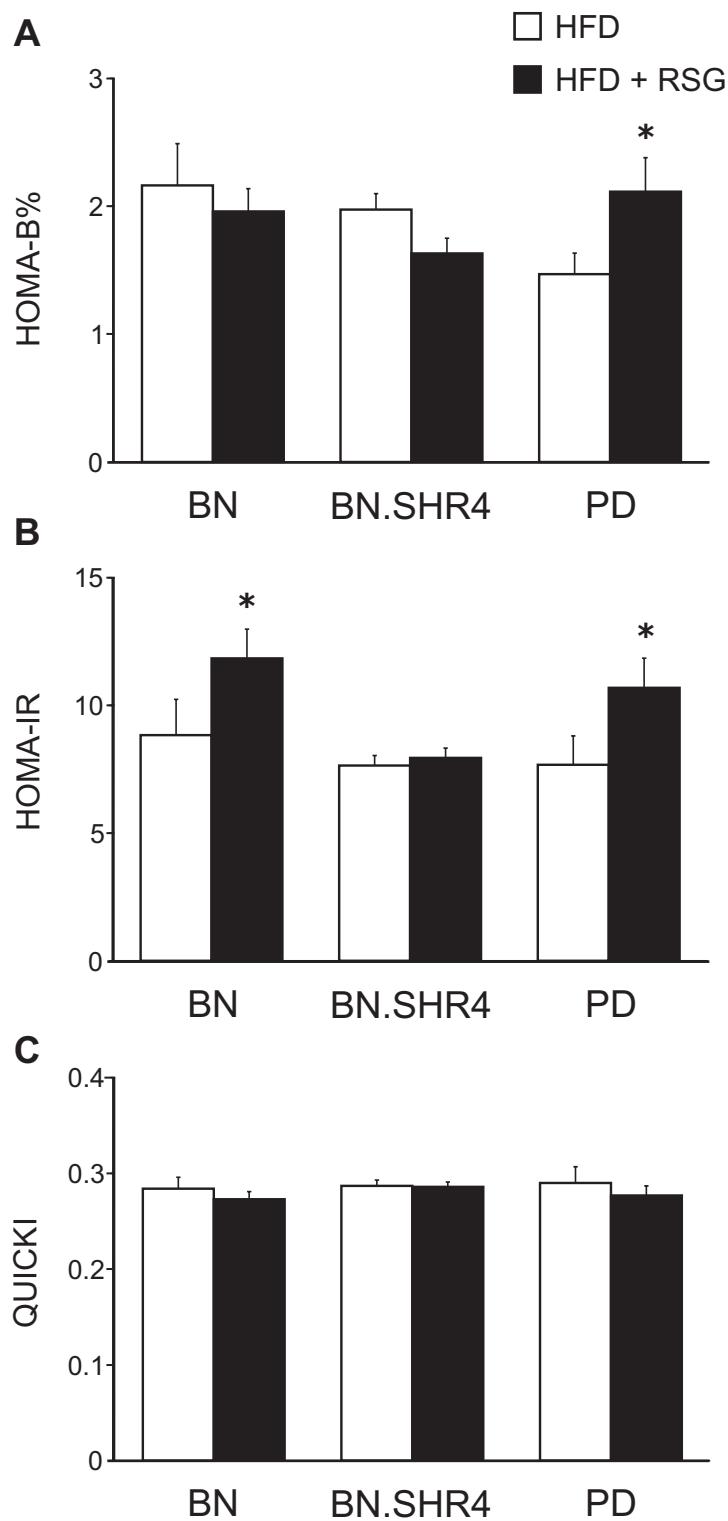
Trait	p STRAIN	p TREATMENT	p S*T
Triglycerides	0.02	0.31	0.12
Cholesterol	<0.0001	0.78	0.31
FPG	0.02	<0.0001	0.009
AUC (OGTT)	0.06	<0.0001	0.68
Insulin	0.11	0.11	0.10
HOMA B%	0.32	0.83	0.026
HOMA IR	0.030	0.009	0.23
QUICKI	0.11	0.009	0.23
Free fatty acids (0 min)	0.002	0.25	0.30
Free fatty acids (60 min)	0.002	0.11	0.76
Initial body weight	<0.0001	0.54	0.18
Final body weight (b.w.)	<0.0001	0.71	0.15
Adiposity index	<0.0001	0.04	0.01
Heart weight/100g b.w.	0.009	0.75	0.94
Kidney weight/100g b.w.	0.16	0.52	0.50
Liver weight/100g b.w.	<0.0001	0.43	0.04
Muscle weight/100g b.w.	<0.0001	0.40	0.20
Liver cholesterol	0.0004	0.007	0.009
Liver triglyceride	0.004	0.88	0.30
Muscle cholesterol	<0.0001	0.59	0.82
Muscle triglyceride	0.007	0.97	0.39
Lipogenesis (insulin -)	<0.0001	0.09	0.01
Lipogenesis (insulin +)	<0.0001	0.08	<0.0001
Glycogenesis (insulin -)	0.13	0.92	0.25
Glycogenesis (insulin +)	0.012	0.92	0.09
Glucose oxidation (insulin -)	0.15	0.03	0.14
Glucose oxidation (insulin +)	0.001	0.09	0.01
Plasma SOD	0.44	0.08	0.54
Liver SOD	<0.001	<0.001	0.009
Plasma CAT	0.62	0.24	0.76
Liver CAT	<0.001	0.59	0.20
Plasma GSH-Px	0.69	0.001	0.47
Liver GSH-Px	0.0002	0.20	0.37
Plasma GSH	0.62	0.006	0.28
Liver GSH	0.78	0.44	0.29
Plasma TBARS	0.52	0.01	0.79
Liver TBARS	0.42	0.0007	0.72
Plasma CD	0.08	<0.001	0.01
Liver CD	0.54	0.76	0.63

S*T: STRAIN*TREATMENT factor interaction, AUC: area under the glycaemic curve, CAT: Catalase, CD: conjugated dienes, FPG: fasting plasma glucose, GSH-Px: Glutathione peroxidase, GSH: Glutathione, HOMA B%: homeostasis model assessment of percent β -cell function index, HOMA IR homeostasis model assessment of insulin resistance index, OGTT: oral glucose tolerance test, QUICKI: quantitative insulin sensitivity check index, SOD: Superoxide dismutase, TBARS: Thiobarbituric acid reactive substances.

Supplementary Table 3.

Gene Identifier	Affymetrix ID	Gene Title	log2 fold change
			PD BN BN.SHR4
Rosiglitazone-upregulated genes in all strains			
NM_019282	1369113_at	Rattus norvegicus cysteine knot superfamily 1, BMP antagonist 1 (Cktsf1b1), mRNA.	1.91 1.09 2.25
NM_080394	1369093_at	Rattus norvegicus Reelin (Rein), mRNA.	1.44 0.84 1.50
NM_013200	1367742_at	Rattus norvegicus Carnitine palmitoyltransferase 1 beta, muscle isoform (Cpt1b), mRNA.*	1.05 0.36 0.81
AA859612	1385438_at	EST, Weakly similar to 810024J URF 4 (H.sapiens)	0.60 0.72 0.70
NM_053289	1368238_at	Rattus norvegicus Pancreatitis-associated protein 1 (Pap1), mRNA.	0.68 1.02 1.12
M57668	1370384_a_at	Rat prolactin receptor (ovarian form) mRNA, complete cds.	0.90 0.76 0.81
M35965	1370764_a_at	Rat thyroglobulin (Tg-2) mRNA, complete cds.	1.09 1.30 1.95
AI029631	1388277_at	Rat Ig germline C-lambda chain gene C1-region, 3 end	0.70 0.82 0.63
NM_024162	1367660_at	Rattus norvegicus Fatty acid binding protein 3, muscle and heart (Fabp3), mRNA.	1.78 0.87 1.43
Rosiglitazone-downregulated genes in all strains			
NM_031556	1370131_at	Rattus norvegicus Caveolin, caveola protein, 22 kDa (Cav1), mRNA.*	-1.65 -0.37 -0.34
NM_024373	1369269_at	Rattus norvegicus polypeptide GalNAc transferase T1 (Galnt1), mRNA.	-1.22 -0.61 -0.54
NM_031787	1369325_at	Rattus norvegicus homeodomain-interacting protein kinase 3 (Hipk3), mRNA.	-1.18 -0.86 -0.37
NM_031315	1369250_at	Rattus norvegicus acyl-CoA thioesterase 1, cytosolic (Cte1), mRNA.	-1.32 -1.01 -0.09
NM_012843	1369736_at	Rattus norvegicus Epithelial membrane protein 1 (Emp1), mRNA.	-1.30 -0.44 -0.69
NM_133315	1387130_at	Rattus norvegicus solute carrier family 39 (iron-regulated transporter), member 1 (Slc39a1), mRNA.	-0.78 -0.81 -0.89
H32486	1385814_at	ESTs, Weakly similar to M3K1 RAT MITOGEN-ACTIVATED PROTEIN KINASE KINASE 1 (R.norvegicus)	-2.02 -1.00 0.47
NM_131914	1370135_at	Rattus norvegicus caveolin 2 (Cav2), mRNA.	-1.42 -0.48 -0.65
NM_022711	1387580_at	Rattus norvegicus sterol 5-alpha-reductase 2 (Srd5a2), mRNA.	-0.41 -1.68 -0.54
NM_017309	1369152_at	Rattus norvegicus protein phosphatase 3, regulatory subunit B, alpha isoform (calcineurin B, type I) (Ppp3r1), mRNA.	-1.16 -0.98 -0.62
AY066016	1368622_at	Rattus norvegicus strain BNCrI glucocorticoid receptor mRNA, complete cds.	-1.33 -0.59 -0.90
NM_013084	1369526_at	Rattus norvegicus Acyl-Coenzyme A dehydrogenase, short-branched chain (Acadsb), mRNA.	-1.42 -0.71 -0.79
L19181	1370488_a_at	Rat receptor-linked protein tyrosine phosphatase (PTP-PS) mRNA, complete cds.	-1.81 -0.88 -0.43
BI300565	1370955_at	A disintegrin and metalloprotease domain (ADAM) 10	-1.29 -1.49 -0.41
NM_053588	1387201_at	Rattus norvegicus Trif gene (Trif-pending), mRNA.	-1.21 -0.30 -1.68
NM_013076	1387748_at	Rattus norvegicus Obesity (murine homolog, leptin) (Lep), mRNA.*	-0.98 -1.08 -1.14
AI235240	1383096_at	Amyloid protein precursor-like protein 2	-1.57 -0.87 -0.78
AI177164	1375528_at	ESTs, Moderately similar to JC7284 phosphoprotein A2 (H.sapiens)	-1.41 -0.86 -0.97
Y13591	1371173_a_at	Rattus norvegicus protein calpastatin, clone RNCAST23.	-1.75 -0.84 -0.97
NM_022589	1368104_at	Rattus norvegicus Tspan-2 protein (Tspan-2), mRNA.	-1.32 -1.49 -0.82
NM_013160	1368963_at	Rattus norvegicus Max interacting protein 1 (Mxi1), mRNA.	-2.61 -0.80 -0.28
NM_012689	1387704_at	Rattus norvegicus Estrogen receptor 1 (Esrl), mRNA.	-2.11 -0.93 -0.71
NM_022210	1387646_a_at	Rattus norvegicus Max (Max), mRNA.	-1.14 -1.42 -1.31
BG378227	1369640_at	Gap junction protein, alpha 1, 43 kD (connexin 43)	-1.02 -1.61 -1.29
NM_019258	1368767_at	Rattus norvegicus cystatin 8 (cystatin-related epididymal spermaticogen) (Cst8), mRNA.	-0.94 -4.06 1.02
AW533292	1376532_at	ESTs, Moderately similar to T46917 hypothetical protein DKFZp762K137.1 (H.sapiens)	-2.57 -0.28 -1.23
NM_017139	1367949_at	Rattus norvegicus preproenkphalin, related sequence (Penk-rs), mRNA.	-0.73 -4.10 0.69
L09653	1369653_at	Rattus norvegicus transforming growth factor-beta type II receptor mRNA, complete cds.	-3.90 -0.69 -0.63
BE349699	1373780_at	ESTs, Moderately similar to A59262 tetraspan TSPAN-1 (H.sapiens)	-1.82 -4.79 0.87
NM_139085	1369925_at	Rattus norvegicus cystatin 11 (Cst11), mRNA.	-0.25 -6.55 0.68
AJ312745	1371179_a_at	Rattus norvegicus partial mRNA for soluble fibroblast growth factor receptor IIb (sKGF-R gene).	-4.32 -0.95 -0.87
NM_020301	1369823_at	Rattus norvegicus a disintegrin and metalloprotease domain (ADAM) 7 (Adam7), mRNA.*	-1.54 -5.90 0.48
Genes with distinct expression change in response to RSG between BN and BN.SHR4 - pharmacogenetic effect of the differential segment			
M31109	1387955_at	Rat UDP-glucuronosyltransferase mRNA, complete cds.	3.88 3.69 -1.80
M18336	1370500_a_at	Rat cytochrome P450 processed pseudogene mRNA, complete cds.	1.95 2.45 -0.90
NM_013105	1387118_at	Rattus norvegicus Cytochrome P450, subfamily IIIA, polypeptide 3 (Cyp3a3), mRNA.	1.79 1.37 -0.78
NM_012696	1387050_s_at	Rattus norvegicus T-kininogen, see also D11Elh1 and D11Mit8 (Kng), mRNA.	1.61 1.39 -0.71
NM_023103	1370027_a_at	Rattus norvegicus alpha(1)-inhibitor 3, variant 1 (Mug1), mRNA.	1.44 2.41 -1.25
NM_012824	1368587_at	Rattus norvegicus Apolipoprotein C1 (Apoc1), mRNA.	1.39 1.71 -1.16
AF368860	1370349_a_at	Rattus norvegicus 3 non-translated beta-F1-ATPase mRNA-binding protein mRNA, complete cds.	1.37 2.11 -0.63
NM_031533	1387825_at	Rattus norvegicus Androsterone UDP-glucuronosyltransferase (Ugl2b2), mRNA.	1.23 2.29 -1.16
NM_012556	1369111_at	Rattus norvegicus Fatty acid binding protein 1, liver (Fabp1), mRNA.	1.15 3.95 -1.59
NM_017222	1368745_at	Rattus norvegicus solute carrier family 10, member 2 (Slc10a2), mRNA.	0.67 1.14 -0.89
NM_020301	1369823_at	Rattus norvegicus a disintegrin and metalloprotease domain (ADAM) 7 (Adam7), mRNA.*	-1.54 -5.90 0.48
AI172274	1374527_at	ESTs, Weakly similar to ENOYL-COA HYDRATASE, MITOCHONDRIAL PRECURSOR (R.norvegicus)	-0.08 1.86 -1.72
NM_012806	1369102_at	Rattus norvegicus Stress activated protein kinase beta (Serk2), mRNA.	-0.16 1.41 -1.88
NM_053718	1368279_at	Rattus norvegicus myelodysplastic or mixed-lineage leukemia (trithorax (Drosophila) homolog); translocated to, 3 (Mllt3), mRNA.	-2.05 2.57 -1.08
NM_024381	1387491_at	Rattus norvegicus ATP-stimulated glucocorticoid receptor translocator protein (Gyr), mRNA.	1.04 -1.07 2.01
NM_022960	1368621_at	Rattus norvegicus neutral solute channel aquaporin 9 (Aqp9), mRNA.*	0.74 -3.24 0.57
BI287326	1384202_at	ESTs, Weakly similar to calcium binding protein P22 (Rattus norvegicus) (R.norvegicus)	0.27 -1.45 1.04
BI291386	1377191_at	ESTs, Highly similar to ATP synthase, H+ transporting, mitochondrial F1FO complex, subunit e (Rattus norvegicus) (R.norvegicus)	0.10 -0.90 1.59
M25143	1370377_at	Rat cytochrome P450CMF1b mRNA, complete cds.	-0.03 -1.27 2.55
NM_053822	1368494_at	Rattus norvegicus S100 calcium-binding protein A8 (calgranulin A) (S100a8), mRNA.	-0.08 -0.94 3.11
M85193	1370629_at	Rattus norvegicus RT6.2 mRNA, complete cds.	-0.82 -1.80 1.76
NM_019258	1368767_at	Rattus norvegicus cystatin 8 (cystatin-related epididymal spermaticogen) (Cst8), mRNA.	-0.94 -4.06 1.02
Genes with distinct expression change in response to RSG between PD and BN.SHR4 - comparison of RSG effect in two d distinct inbred models of metabolic syndrome			
NM_012503	1370149_at	Rattus norvegicus Alkaloylacylprotein receptor 1 (hepatocyte lectin) (Asgr1), mRNA.	1.72 -1.81 -1.42
NM_133543	1387240_at	Rattus norvegicus retinol dehydrogenase type III (RoDH(III)), mRNA.	1.98 -0.72 -0.81
NM_012556	1369111_at	Rattus norvegicus Fatty acid binding protein 1, liver (Fabp1), mRNA.	1.15 3.95 -1.59
NM_012567	1369639_at	Rattus norvegicus Gap junction protein, alpha 1, 43 kD (connexin 43) (Gja1), mRNA.	1.45 -0.36 -0.60
NM_134350	1387283_at	Rattus norvegicus myxovirus (influenza virus) resistance 3 (Mx3), mRNA.	2.54 -0.09 -0.17
NM_017300	1387508_at	Rattus norvegicus bile acid-Coenzyme A hydrolase, amino acid n-acyltransferase (Baat), mRNA.	2.02 -0.73 -1.08
J04526	AFFX_Rat_Hexoki	Rattus norvegicus brain hexokinase mRNA, complete cds.	1.79 -0.55 -1.04
NM_013105	1387118_at	Rattus norvegicus Cytochrome P450, subfamily IIIA, polypeptide 3 (Cyp3a3), mRNA.	1.79 1.37 -0.78
AF134409	1370372_at	Rattus norvegicus Rhes protein mRNA, complete cds.	1.89 -1.14 -0.48
NM_012730	1367917_at	Rattus norvegicus Cytochrome P450, subfamily IID2 (Cyp2d2), mRNA.	0.99 -0.11 -1.06
NM_053482	1370088_at	Rattus norvegicus sperm autoantigenic protein 17 (Sp17), mRNA.	1.02 -0.70 -1.03
NM_053318	1370065_at	Rattus norvegicus hemopexin (Hpx), mRNA.	1.52 1.01 -0.51
BE104506	1398458_at	ESTs, Moderately similar to T50615 hypothetical protein DKFZp761G1515.1 (H.sapiens)	1.61 -0.88 -0.88
NM_031533	1387825_at	Rattus norvegicus Androsterone UDP-glucuronosyltransferase (Ugl2b2), mRNA.	1.23 2.29 -1.16
NM_133599	1368278_at	Rattus norvegicus lectin, galactoside-binding, soluble 2 (galectin 2) (Lgals2), mRNA.	2.03 -0.66 -2.16
NM_023103	1370027_a_at	Rattus norvegicus alpha(1)-inhibitor 3, variant 1 (Mug1), mRNA.	1.44 2.41 -1.25
AB019693	1370592_at	Rattus norvegicus HP33 mRNA, complete cds.	2.48 -0.91 -0.15
NM_017112	1368431_at	Rattus norvegicus hepsin (Hon), mRNA.	0.90 -0.71 -1.30
AA800502	1370241_at	cytochrome P450, 2c39	1.54 0.18 -0.50
NM_053626	1369491_at	Rattus norvegicus D-amino acid oxidase (Dao1), mRNA.	1.72 -1.02 -0.66
NM_019334	1368785_a_at	Rattus norvegicus paired-like homeodomain transcription factor 2 (Pitx2), mRNA.	2.08 -0.88 -1.35
NM_017332	1367707_at	Rattus norvegicus fatty acid synthase (Fasn), mRNA.	3.16 0.30 -0.27
NM_031154	1387023_at	Rattus norvegicus glutathione S-transferase, mu type 3 (Yb3) (Gstm3), mRNA.	1.78 -0.86 -0.84
NM_012824	1368587_at	Rattus norvegicus Apolipoprotein C1 (Apoc1), mRNA.	1.39 1.71 -1.16
AA817993	1375707_at	ESTs, Moderately similar to A53798 58kDa membrane-associated protein - rat (R.norvegicus)	1.51 -0.86 -0.99
NM_019168	1368672_at	Rattus norvegicus arginase type II (Arg2), mRNA.	1.45 -1.20 -0.86
NM_031841	1367668_a_at	Rattus norvegicus stearoyl-Coenzyme A desaturase 2 (Scd2), mRNA.	1.88 -0.65 -0.20
A117934	1367708_a_at	fatty acid synthase	1.88 0.33 -0.13
AF368860	1370349_a_at	Rattus norvegicus 3 non-translated beta-F1-ATPase mRNA-binding protein mRNA, complete cds.	1.37 2.11 -0.63
AF368860	1370349_a_at	Rattus norvegicus 3 non-translated beta-F1-ATPase mRNA-binding protein mRNA, complete cds.	1.37 2.11 -0.63
BF289341	1389946_at	ESTs, Moderately similar to T13614 N-acetyltransferase homolog EG:8D8.6 - fruit fly (D.melanogaster)	1.53 0.05 -0.39
A1008409	1389986_at	unknown Glu-Pro dipeptide repeat protein	0.32 -1.84 -1.60
NM_031736	1368150_at	Rattus norvegicus solute carrier family 27 (fatty acid transporter), member 2 (Slc27a2), mRNA.	1.22 -0.76 -0.68
NM_031712	1368178_at	Rattus norvegicus PDZ domain containing 1 (Pdzk1), mRNA.	1.12 -1.26 -0.77
NM_031536	1368364_at	Rattus norvegicus Mitochondrial capsule selenoprotein (Mscp), mRNA.	0.46 -0.80 -1.42
J04488	1367851_at	Rat prostaglandin D synthetase mRNA, complete cds.*	1.30 -0.77 -0.57

*...validated by RT-PCR.



Supplementary Figure 1. Insulin resistance indices. The homeostasis model assessment (HOMA) of percent β -cell function (HOMA-B%) (top), HOMA of insulin resistance index (HOMA-IR) (middle) and quantitative insulin sensitivity check index (QUICKI) (bottom) in control (HFD, white bars) and rosiglitazone-treated (HFD + RSG, black bars) adult male rats of BN, BN.SHR4 and PD rat strains. The significance levels only for intra-strain rosiglitazone effect are shown (post-hoc Tukey's HSD) as follows: *...p<0.05.