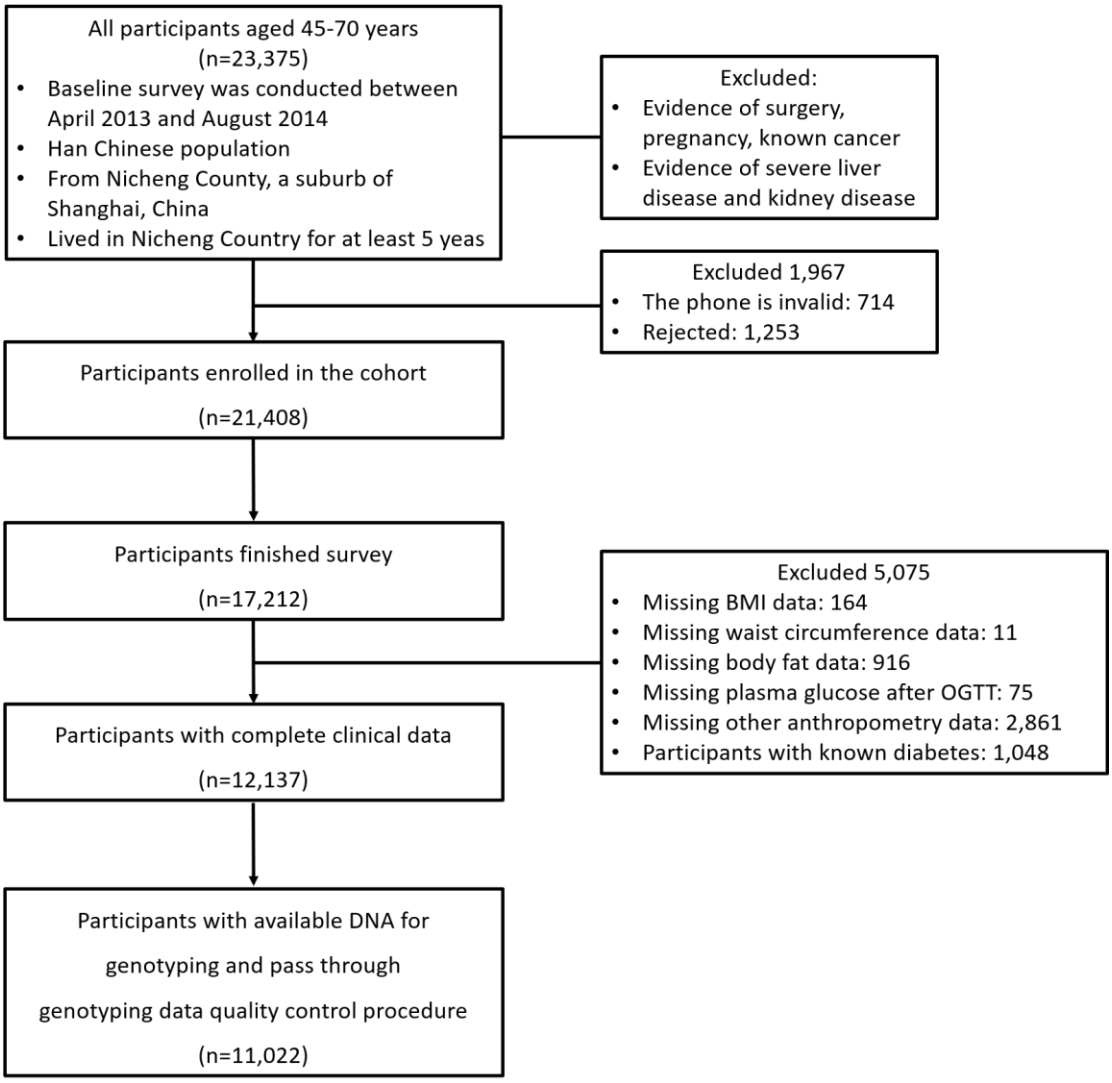
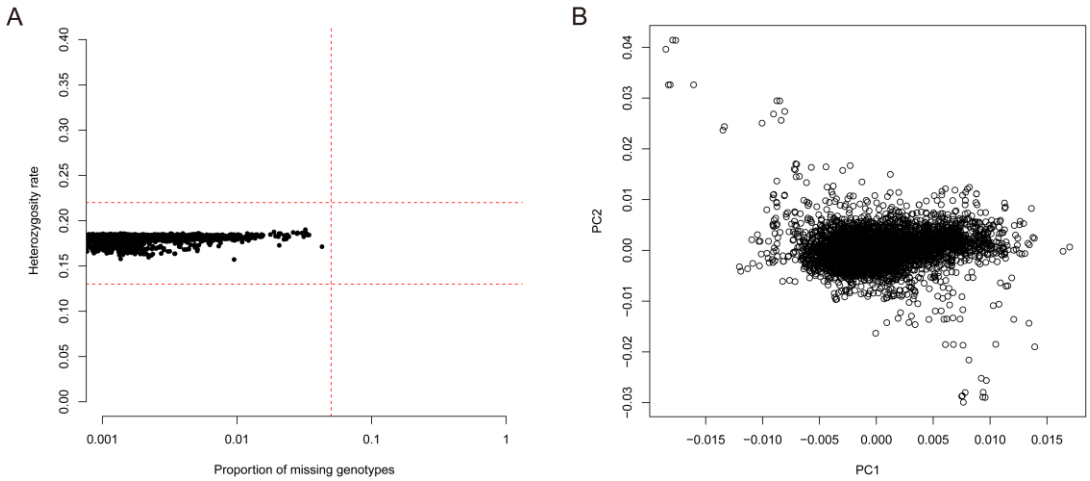
Supplementary Figure 1. The flowchart of participants selection(N=11,022)

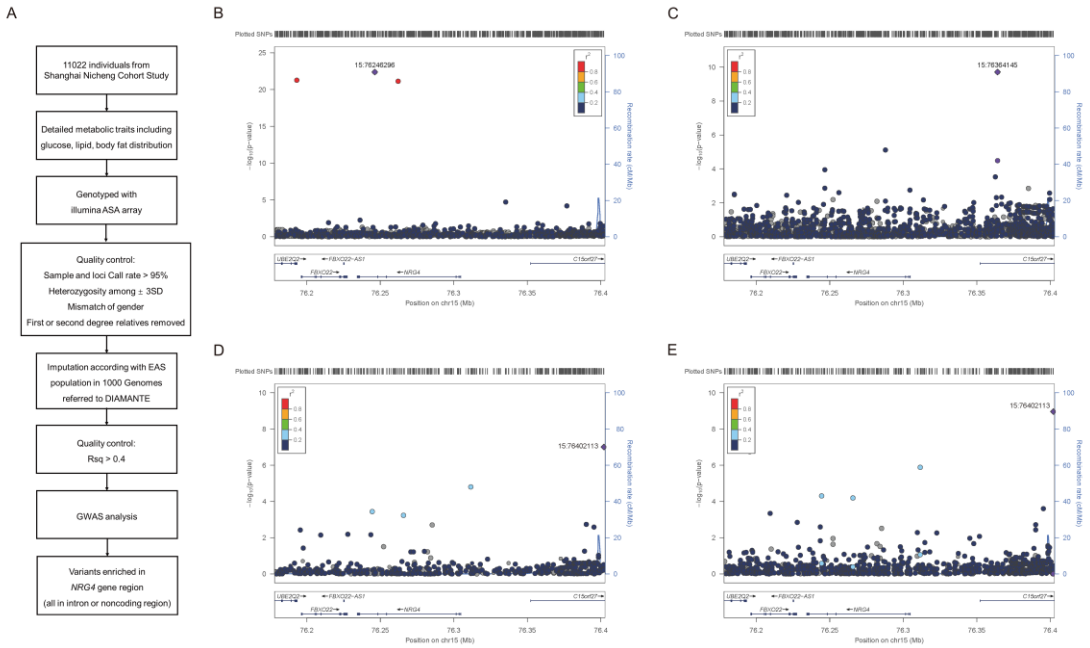


Supplementary Figure 2. Quality control of genome-wide genotype array data



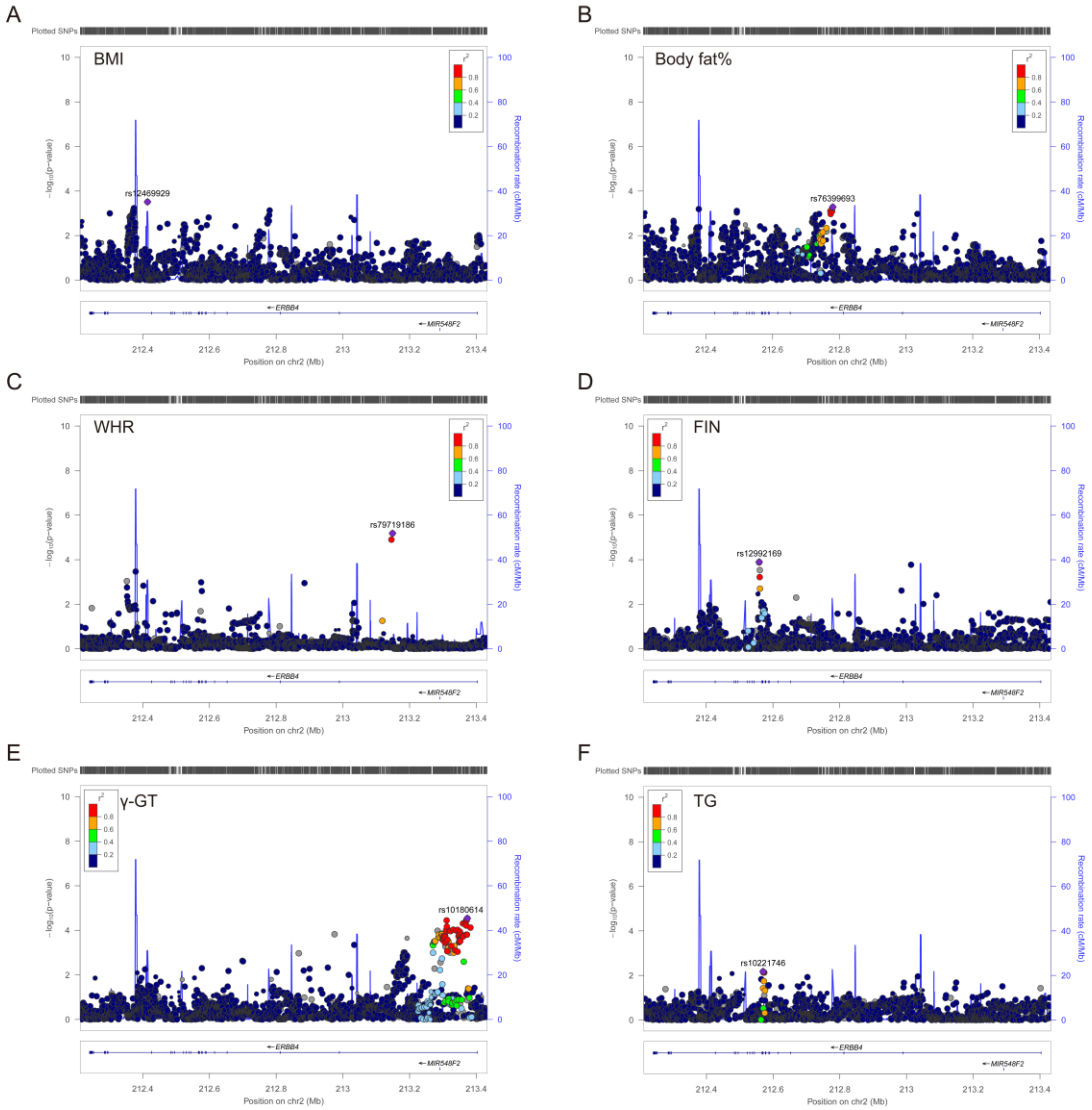
A. Summary of missing genotypes and heterozygosity rate of genome-wide genotyping data. B. Principal component analysis of individual according to the genotyping data.

Supplementary Figure 3. Quality control of genotyping data and association analysis results of *NRG4* gene region.



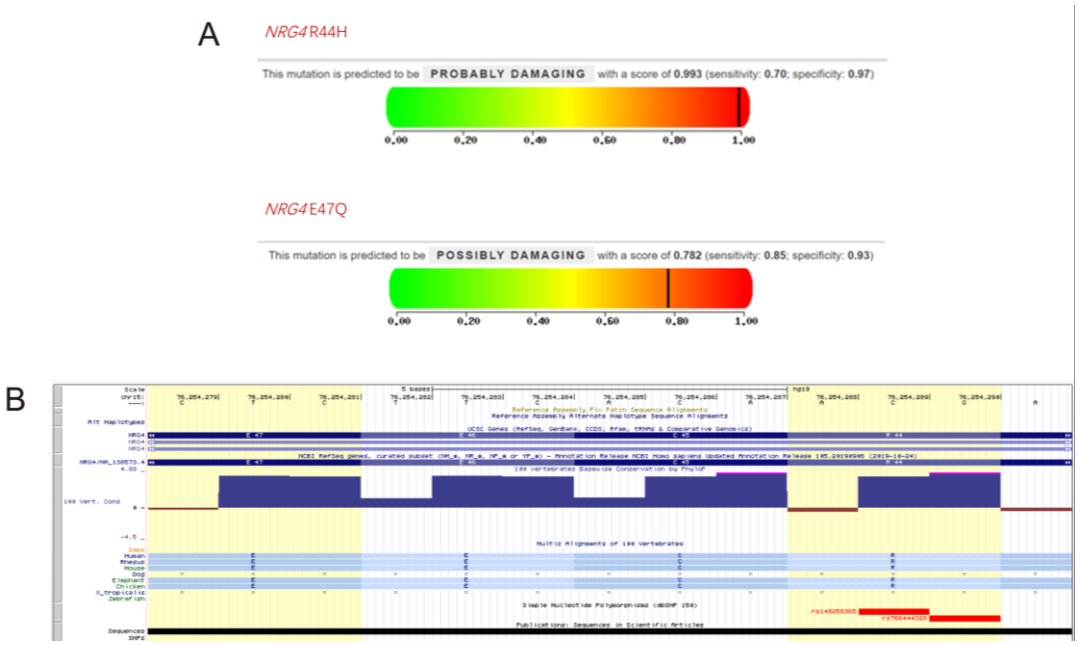
A. Workflow of quality control procedures on genotyping data. B. Region plot of association between loci and Fat mass. C. Region plot of association between loci and plasma triglyceride level. D. Region plot of association between loci and fasting plasma insulin level. E. Region plot of association between loci and HOMA-IR index

Supplementary Figure 4. Association between genetic loci in *ERBB4* gene region with metabolic traits



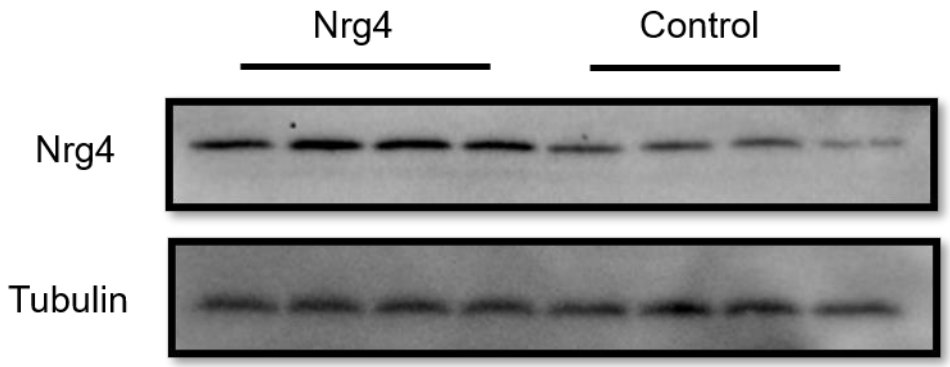
Region plot of association between loci and body mass index (A), body fat (B), waist to hip circumference ratio (C), fasting plasma insulin (D), γ-glutamyltransferase (E), Total triglyceride (F). BMI, body mass index; WHR, waist to hip circumference ratio; FIN, fasting plasma insulin; γ-GT, γ-glutamyltransferase; TG, Total triglyceride.

Supplementary Figure 5. The analysis of Nrg4 R44H and E47Q mutation



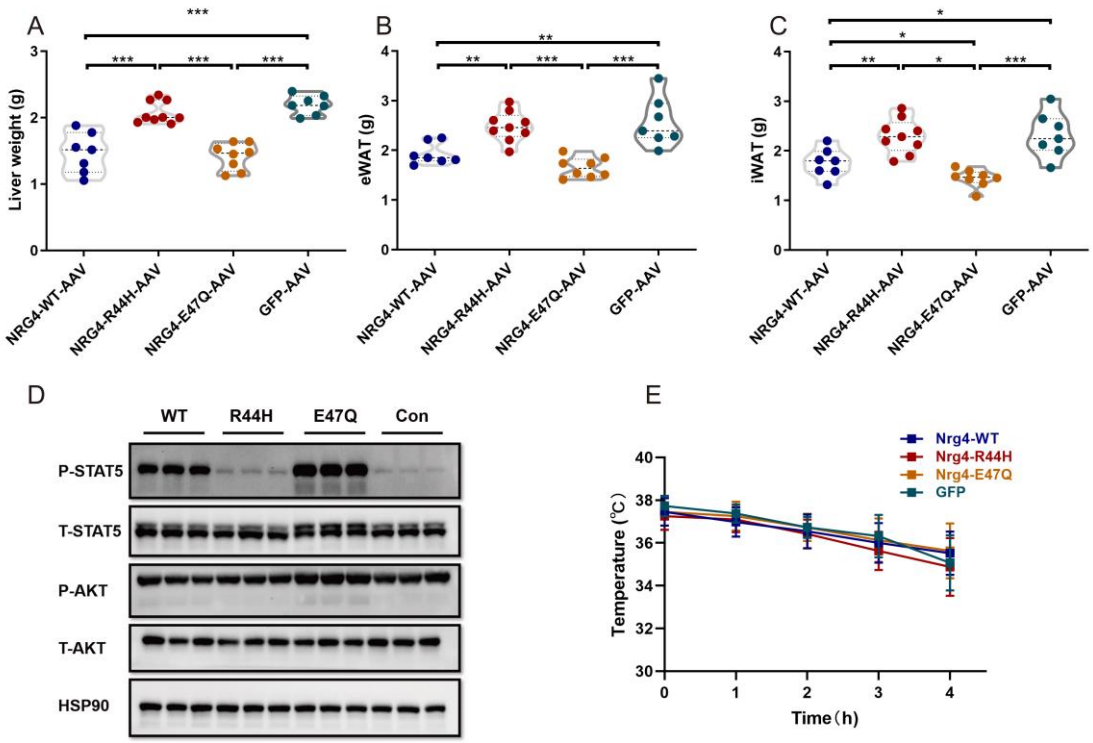
A. Prediction the effect of mutation on Nrg4 protein by polyphen2. B. PhyloP basewise conservation analysis of Nrg4 R44H and E47Q. UCSC genome browser show the mutaion position of p.R44H and p.E47Q. Vertebrate Multiz Alignment & Conservation from 100 Species (100 Vert. Cons) analysis (Basewise Conservation (phyloP)) show the mutations are located in highly conserved position across the species.

Supplementary Figure 6. The expression of Nrg4 in liver



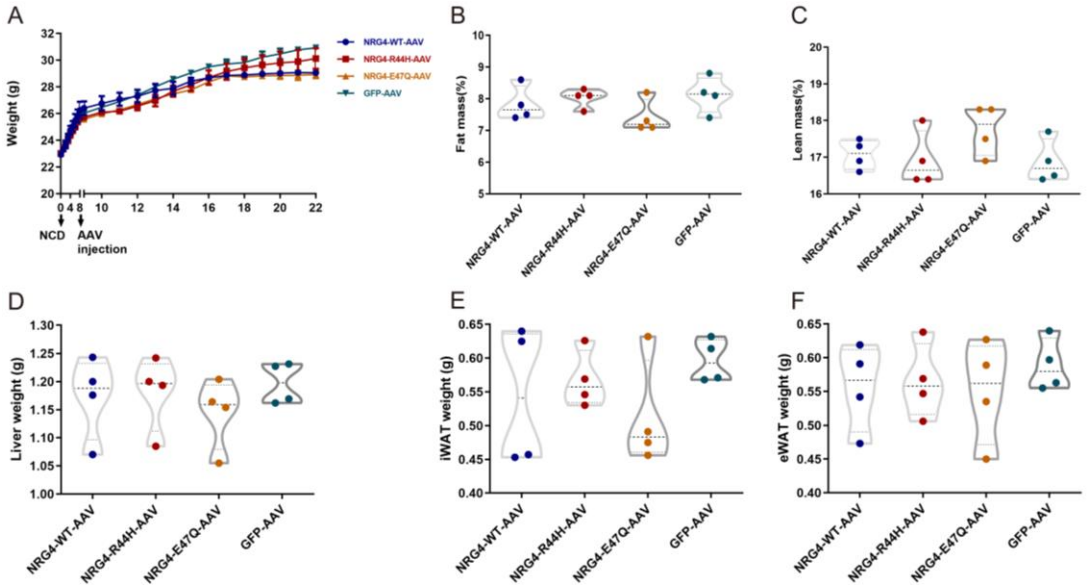
Immunoblots of total liver tissue lysates in Nrg4 overexpression mice and GFP control mice (1 month after AAV injection).

Supplementary Figure 7. Weight of liver and white adipose tissue in different groups under high fat diet feeding



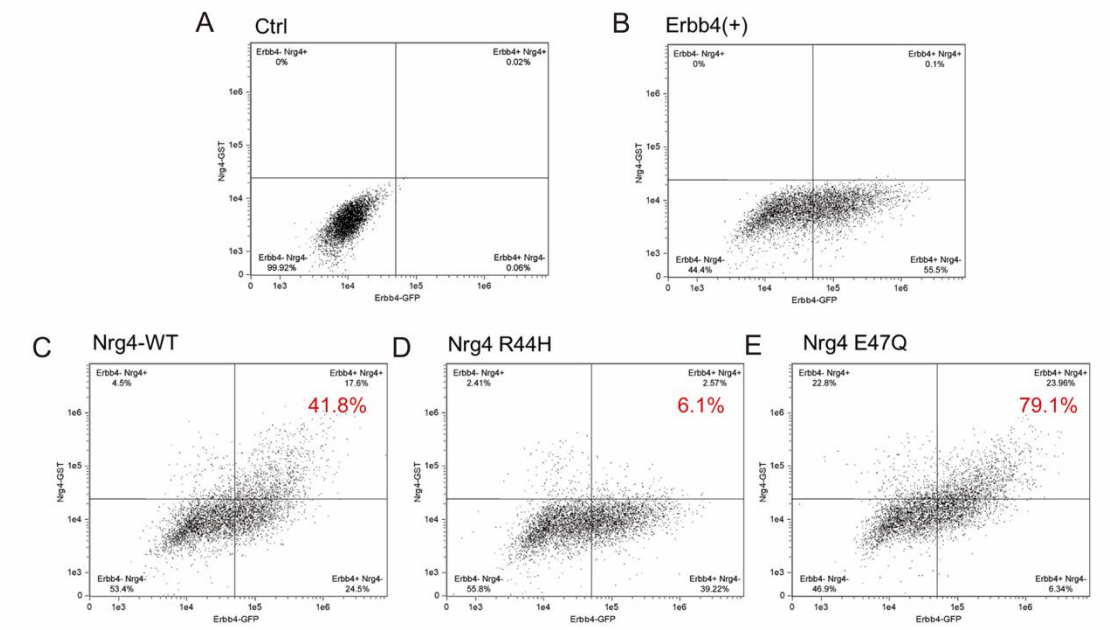
A-C. The comparison of liver, eWAT and iWAT weight among four groups (n=7-9, per group). D. Immunoblots of total liver tissue lysates in Nrg4 wild-type (WT), R44H (M1) and E47Q (M2) mutation overexpression mice and GFP control mice (Con). E. Rectal temperature in mice following cold exposure at 4°C for 4 hours. eWAT, Epididymal white adipose tissue; iWAT, subcutaneous white adipose tissue. The statistical comparison was performed by analysis of variance (ANOVA) and the Student-Newman-Keuls (SNK) multiple pairwise comparison tests with the SAS (version 9.0).

Supplementary Figure 8. Weight and body fat distribution of mice under normal chow diet



A. Body weight of mice fed with NCD starting at 8 weeks age (n=4–5 per group). B-C. Percentage of fat and lean body mass in the four group mice fed NCD for 22 weeks (n=4 per group). D-F: Weight of liver and white adipose tissue. eWAT, Epididymal white adipose tissue; iWAT, subcutaneous white adipose tissue. The statistical comparison was performed by analysis of variance (ANOVA) and the Student-Newman-Keuls (SNK) multiple pairwise comparison tests with the SAS (version 9.0).

Supplementary Figure 9. Binding capacity as measured by flow cytometer in HepG2 cells.



Binding capacity as measured by flow cytometer between GFP-ErbB4 after 36 h transfection and Nrg4 incubation for 1 h at a concentration of 100 μg/mL. (A-E) Dot plot obtained from flow cytometry analysis indicates the four different cell populations. The binding capacity was measured by the proportion of ErbB4+Nrg4+ hepatocytes (top right) in ErbB4+ hepatocytes (top right and bottom right).

Supplementary Table 1. Clinical characteristics of 11,022 community subjects in Chinese Han population

	Male (n=4,571)	Female (n=6,451)
Age (year)	56.10±7.22	55.81±7.43
Body mass index (kg/m^2)	24.85±3.17	24.72±3.37
Newly diagnosed diabetes (%)	14.57	14.88
Waist circumference (cm)	86.67±9.34	82.56±9.28
Waist-to-Hip circumference Ratio	0.93±0.38	0.90±1.06
Body fat (%)	22.83±5.47	34.54±5.99
Fat mass (kg)	16.52±6.80	21.73±7.70
Nonfat mass (kg)	53.78±8.18	39.74±8.39
γ-glutamyltransferase (U)	44.92±55.62	26.89±28.95
Total cholesterol (mmol/L)	5.01±0.96	5.24±1.08
Triglyceride (mmol/L)	1.83±1.81	1.60±1.31
High-density lipoprotein-cholesterol (mmol/L)	1.24±0.34	1.39±0.34
Low-density lipoprotein-cholesterol (mmol/L)	3.04±0.82	3.15±0.83
Impaired fasting glucose (%)	39.73%	35.00%
Impaired glucose regulation (%)	52.71%	52.70%
Fasting plasma glucose (mmol/L)	6.22±1.63	6.09±1.46
30 minute plasma glucose after OGTT (mmol/L)	10.60±2.56	10.17±2.40
2 hour plasma glucose after OGTT (mmol/L)	8.42±4.27	8.75±4.05
Glucose area under curve	18.46±5.50	18.29±5.17
HbA1c (%) [mmol/mol]	5.81±0.94	5.83±0.98
Fasting plasma insulin (mU/L)	8.47±10.61	10.09±19.34
30 minute plasma insulin after OGTT (mU/L)	52.33±41.03	58.73±44.77
2 hour plasma insulin after OGTT (mU/L)	44.03±40.72	59.32±51.77
Insulin area under curve	87.42±61.32	105.79±73.97
HOMA-β	70.45±150.71	84.30±113.92
HOMA-IR	2.51±5.09	2.96±8.56
Stumvoll 1st	489.60±336.32	466.66±338.21
Stumvoll 2nd	179.61±146.20	168.28±143.59
GUTT-ISI index	70.86±21.66	74.91±19.11
Free fatty acid (μmol/L)	529.97±228.62	584.67±229.9
Serum high-sensitivity C-reactive protein (mg/L)	1.84±4.29	1.83±3.80

Data are shown as mean ± standard deviation. OGTT, oral glucose tolerance test; HOMA-β, Homeostatic Model Assessment of β-cell function; HOMA-IR, Homeostatic Model Assessment for Insulin Resistance.

Supplementary Table 2. Clinical characteristics of 2,388 subjects in Shanghai obesity study (SHOS)

	Male (n=1,076)	Female (n=1,312)
Age (year)	52.3±7.47	51.2±7.22
Body mass index (kg/m^2)	24.8±3.29	24.06±3.48
Diabetes status (%)	29.27%	32.06%
Waist circumference (cm)	88.01±9.22	83.19±9.1
Waist to hip circumference ratio	0.93±0.05	0.89±0.06
Body fat (%)	24.83±5.93	32.53±7.01
Fat mass (kg)	18.14±7.08	20.28±9.31
Nonfat mass (kg)	52.69±6.34	39.74±4.05
Visceral fat area (cm^2)	98.42±48.52	71.98±34.68
Subcutaneous fat area (cm^2)	136.8±53.18	185.65±67.19
VSR	0.73±0.31	0.4±0.18
Hypertension (%)	30.76%	16.69%
Systolic blood pressure (mmHg)	127.72±15.36	121.93±15.72
Diastolic blood pressure (mmHg)	82.36±9.99	78.48±13.61
Alanine aminotransferase (U/L)	27.5±20.36	21±15.45
Aspartate aminotransferase (U/L)	24.02±11.33	22.08±10.46
γ-glutamyltransferase (U)	43.7±51.3	24.77±26.04
Total cholesterol (mmol/L)	5.08±1.02	5.28±1.63
Triglyceride (mmol/L)	1.97±1.71	1.49±1.29
High-density lipoprotein-cholesterol (mmol/L)	1.3±0.32	1.55±0.35
Low-density lipoprotein-cholesterol (mmol/L)	3.21±1.01	3.13±0.97
Fasting plasma glucose (mmol/L)	5.96±1.91	5.57±1.42
30 minute plasma glucose after OGTT (mmol/L)	10.24±3.15	9.1±2.71
2 hour plasma glucose after OGTT (mmol/L)	7.67±4.53	7.17±3.58
Glucose area under curve	17.37±6.4	15.79±5.07
HbA1c (%) [mmol/mol]	5.94±1.04	5.8±0.83
Fasting plasma insulin (mU/L)	10.34±6.56	10.62±5.57
30 minute plasma insulin after OGTT (mU/L)	49.55±31.86	51.31±27.34
2 hour plasma insulin after OGTT (mU/L)	36.36±28.3	41.12±25.96
Insulin area under curve	79.4±45	85.05±41.88
Serum high-sensitivity C-reactive protein (mg/L)	1.92±4.72	1.91±5.16

Data are shown as mean ± standard deviation.

VSR, ratio of visceral fat area to subcutaneous fat area; OGTT, oral glucose tolerance test; HOMA-β, Homeostatic Model Assessment of β-cell function; HOMA-IR, Homeostatic Model Assessment for Insulin Resistance.

Supplementary Table 3. Clinical characteristics of *NRG4* R44H family

	MS4	MS15	MS78	MS564	MS997	MS2321
Sex	Male	Male	Male	Female	Female	Female
Relationship	Proband	Sibling	Cousin	Mother	Cousin	Cousin
NRG4 Mutation	R44H	R44H	-	-	-	-
Age (year)	45.00	42.00	43.00	63.20	47.00	48.00
Body mass index (kg/m^2)	23.09	22.22	21.63	22.12	19.23	22.53
Waist circumference (cm)	85.00	85.00	83.00	.	77.00	77.50
Waist to hip circumference ratio	0.95	0.95	0.81	.	0.83	0.83
Body fat (%)	27.60	26.80	22.00	.	24.60	26.00
Fat mass (kg)	19.30	18.50	14.10	.	11.40	16.10
Nonfat mass (kg)	50.60	50.00	49.90	.	34.80	49.20
Visceral fat area (cm^2)	110.75	98.98	80.93	.	49.32	73.22
Subcutaneous fat area (cm^2)	135.00	121.30	118.44	.	128.95	93.35
VSR	0.82	0.82	0.68	.	0.69	0.78
Hypertension	Yes	Yes	No	Yes	No	No
Systolic blood pressure (mmHg)	165.00	140.00	130.00	150.00	130.00	115.00
Diastolic blood pressure (mmHg)	100.00	85.00	75.00	95.00	80.00	70.00
Alanine aminotransferase (U/L)	48.00	44.00	27.00	31.00	24.00	14.00
Aspartate aminotransferase (U/L)	40.00	34.00	28.00	32.00	27.00	20.00
γ-glutamyltransferase (U)	352.00	165.00	17.00	71.00	13.00	22.00
Total cholesterol (mmol/L)	7.72	5.80	2.81	4.21	2.18	5.72
Triglyceride (mmol/L)	3.64	2.54	0.96	1.87	0.90	0.78
High-density lipoprotein-cholesterol (mmol/L)	2.31	1.59	1.58	0.90	1.85	1.97
Low-density lipoprotein-cholesterol (mmol/L)	4.79	4.87	3.75	3.16	3.35	4.87
Fasting plasma glucose (mmol/L)	6.20	6.10	5.30	6.05	4.51	4.86
30 minute plasma glucose after OGTT (mmol/L)	14.55	11.30	9.71	9.12	7.44	9.46
2 hour plasma glucose after OGTT (mmol/L)	12.49	9.20	4.90	19.95	5.62	3.62
HbA1c (%) [mmol/mol]	6.00	6.00	5.20	5.40	5.20	5.10
Fasting plasma insulin (mU/L)	39.88	34.68	3.54	7.27	2.55	14.68
30 minute plasma insulin after OGTT (mU/L)	120.11	104.23	48.71	30.64	25.93	36.32
2 hour plasma insulin after OGTT (mU/L)	130.23	72.12	28.18	.	23.89	41.97

VSR, ratio of visceral fat area to subcutaneous fat area; OGTT, oral glucose tolerance test; HOMA-β, Homeostatic Model Assessment of β-cell function; HOMA-IR, Homeostatic Model Assessment for Insulin Resistance.

Supplementary Table 4. Top associated single nucleotide polymorphisms (SNPs) in the *ERBB4* gene region with different metabolic traits (n=11,022)

Traits	SNP	Chromosome	NCBI37	Minor allele	Minor allele frequency	β±SE	*P* value
Body mass index (kg/m^2)	rs12469929	2	212,414,289	A	0.03	-0.46±0.13	0.0003
Waist circumference (cm)	rs34687398	2	213,023,995	G	0.06	-0.82±0.25	0.0010
Body fat (%)	rs76399693	2	212,780,518	C	0.01	-1.30±0.37	0.0005
Fat mass (kg)	rs147463522	2	212,410,870	A	0.02	-1.02±0.32	0.0016
Waist to hip circumference ratio	rs79719186	2	213,148,893	T	0.05	0.16±0.03	6.52×10^{-6}
Fasting plasma glucose (mmol/L)	rs2135159	2	213,214,465	T	0.09	0.12±0.03	0.0011
30 minute plasma glucose after OGTT (mmol/L)	rs10445809	2	213,160,978	A	0.19	0.13±0.04	0.0010
2 hour plasma glucose after OGTT (mmol/L)	rs10204337	2	212,460,589	T	0.10	-0.30±0.09	0.0014
Glucose area under curve (mmol/L)	rs76004008	2	213,429,691	A	0.02	0.77±0.24	0.0011
Fasting plasma insulin (mU/L)	rs12992169	2	212,558,805	G	0.03	3.08±0.80	0.0001
30 minute plasma insulin after OGTT (mU/L)	rs35510521	2	212,851,142	C	0.39	2.08±0.63	0.0010
2 hour plasma insulin after OGTT (mU/L)	rs59077058	2	213,152,596	C	0.02	7.39±2.30	0.0013
Insulin area under curve	rs199775547	2	212,855,248	T	0.43	3.24±1.01	0.0014
HOMA-β	rs117878120	2	213,076,602	A	0.02	17.36±5.29	0.0010
HOMA-IR	rs117665034	2	213,014,129	C	0.02	1.39±0.43	0.0014
GUTT-ISI index	rs201145892	2	212,754,754	T	0.38	-0.77±0.24	0.0014
Stumvoll 1st	rs2371310	2	212,421,552	A	0.26	13.20±4.02	0.0010
Stumvoll 2nd	rs192392113	2	212,760,400	C	0.01	14.73±4.48	0.0010
HbA1c (%) [mmol/mol]	rs75927397	2	212,578,546	C	0.03	0.14±0.04	0.0011
Low-density lipoprotein-cholesterol (mmol/L)	rs7426163	2	212,594,673	T	0.14	-0.05±0.02	0.0021
High-density lipoprotein-cholesterol (mmol/L)	rs144148947	2	212,401,824	G	0.03	0.04±0.01	0.0024
Total cholesterol (mmol/L)	rs16848730	2	213,375,866	G	0.09	0.07±0.02	0.0017
Triglyceride (mmol/L)	rs10221746	2	212,569,879	C	0.02	0.20±0.07	0.0067
γ-glutamyltransferase (U)	rs10180614	2	213,372,988	T	0.05	5.34±1.28	2.89×10^{-5}

P values were calculated by multiple linear regression under an additive model after adjusting for age and sex; β values were calculated by minor allele. Base positions were referred by NCBI building 37. OGTT, oral glucose tolerance test; HOMA-β, Homeostatic Model Assessment of β-cell function; HOMA-IR, Homeostatic Model Assessment for Insulin Resistance.

Supplementary Table 5. Associations between *ERBB4* exon region variants and metabolic traits

Traits	Nicheng Cohort (n=11,022)			SHOS (n=2,388)			Meta-analysis (n=13,410)		
	β	SE	*P* value	β	SE	*P* value	β	SE	*P* value
Body mass index (kg/m^2)	-1.02	0.40	0.0104	0.32	0.59	0.5855	-0.60	0.33	0.0679
Waist circumference (cm)	-1.57	1.13	0.1651	0.56	1.59	0.7257	-0.86	0.92	0.3530
Body fat (%)	-2.33	0.89	0.0089	0.21	1.25	0.8631	-1.47	0.73	0.0428
Fat mass (kg)	-2.33	0.99	0.0183	-0.35	1.57	0.8232	-1.77	0.84	0.0344
Waist to hip circumference ratio	-0.02	0.13	0.8545	0.00	0.01	0.7284	0.00	0.01	0.7398
Fasting plasma glucose (mmol/L)	-0.20	0.19	0.2734	0.15	0.29	0.6009	-0.10	0.16	0.5216
30 minute plasma glucose after OGTT (mmol/L)	-0.30	0.29	0.2937	0.52	0.51	0.3102	-0.10	0.25	0.6823
2 hour plasma glucose after OGTT (mmol/L)	-0.75	0.50	0.1350	0.22	0.70	0.7557	-0.42	0.41	0.3035
Glucose area under curve	-0.92	0.63	0.1449	1.01	1.06	0.3427	-0.42	0.54	0.4423
Fasting plasma insulin (mU/L)	-2.28	2.40	0.3422	-0.52	1.06	0.6237	-0.81	0.97	0.4053
30 minute plasma insulin after OGTT (mU/L)	-3.77	5.66	0.5053	4.93	5.13	0.3369	1.00	3.80	0.7920
2 hour plasma insulin after OGTT (mU/L)	-5.19	6.21	0.4039	1.24	4.72	0.7934	-1.11	3.76	0.7670
Insulin area under curve	-7.95	9.11	0.3824	0.71	8.07	0.9303	-3.10	6.04	0.6072
HOMA-β	-11.51	13.12	0.3803	-18.36	49.43	0.7104	-11.96	12.68	0.3456
HOMA-IR	-0.85	1.07	0.4260	-0.06	0.46	0.8978	-0.19	0.43	0.6641
GUTT-ISI index	-0.13	0.12	0.2979	0.02	0.16	0.9080	-0.07	0.10	0.4524
Stumvoll 1st	0.22	0.10	0.0219	-0.39	0.17	0.0257	0.08	0.08	0.3682
Stumvoll 2nd	0.01	0.04	0.8436	0.02	0.06	0.7247	0.01	0.03	0.7166
HbA1c (%) [mmol/mol]	0.25	0.12	0.0390	-0.29	0.24	0.2166	0.14	0.11	0.1959
Low-density lipoprotein-cholesterol (mmol/L)	-0.06	0.19	0.7754	-0.41	0.26	0.1211	-0.18	0.16	0.2501
High-density lipoprotein-cholesterol (mmol/L)	-0.14	5.28	0.9792	-0.66	6.93	0.9241	-0.33	4.20	0.9375

P values were calculated by multiple linear regression under an additive model after adjusting for age and sex; β values were calculated by minor allele. Base positions were referred by NCBI building 37. OGTT, oral glucose tolerance test; HOMA-β, Homeostatic Model Assessment of β-cell function; HOMA-IR, Homeostatic Model Assessment for Insulin Resistance.

Supplementary Table 6. Clinical characteristics of the replication subjects (n=7425)

	Male (n=3703)	Female (n=3722)
Age (year)	56.21±7.51	56.54±6.61
Body mass index (kg/m^2)	25.29±3.15	25.34±3.33
Diabetes status (%)	16.24	18.23
Waist circumference (cm)	87.83±9.13	83.68±9.33
Waist to hip circumference ratio	0.92±0.07	0.91±1.29
Body fat (%)	22.96±5.12	35.58±5.64
Fat mass (kg)	16.73±5.92	22.66±6.55
Nonfat mass (kg)	54.32±5.96	39.81±3.86
γ-glutamyltransferase (U)	50.66±72.59	31.24±36.63
Total cholesterol (mmol/L)	5.09±0.97	5.29±1.03
Triglyceride (mmol/L)	1.95±1.95	1.75±1.51
High-density lipoprotein-cholesterol (mmol/L)	1.2±0.33	1.32±0.32
Low-density lipoprotein-cholesterol (mmol/L)	3.07±0.78	3.22±0.84
Impaired fasting glucose (%)	42.71%	39.41%
Impaired glucose regulation (%)	55.12%	57.92%
Fasting plasma glucose (mmol/L)	6.54±1.75	6.36±1.54
30 minute plasma glucose after OGTT (mmol/L)	11.16±2.6	10.71±2.32
2 hour plasma glucose after OGTT (mmol/L)	9.57±4.44	10.02±4.17
Glucose area under curve	18.46±5.50	18.29±5.17
HbA1c (%) [mmol/mol]	5.94±1.00	5.97±0.91
Fasting plasma insulin (mU/L)	9.22±11.25	11.21±20.05
30 minute plasma insulin after OGTT (mU/L)	51.7±44.7	58.66±48.38
2 hour plasma insulin after OGTT (mU/L)	52.81±47.92	70.53±56.67
Insulin area under curve	93.22±67.91	114.04±81.01
HOMA-β	67.25±63.41	84.43±141.16
HOMA-IR	2.85±5.08	3.39±8.14
Stumvoll 1st	330±212.86	299.27±192.94
Stumvoll 2nd	118.88±65.68	104.09±55.86
GUTT-ISI index	67.41±18.63	71.42±17.78
Free fatty acid (μmol/L)	564.88±238.16	629.64±238.7
Serum high-sensitivity C-reactive protein (mg/L)	1.89±4.29	1.87±3.11

Data are shown as mean ± standard deviation. HOMA-β, Homeostatic Model Assessment of β-cell function; HOMA-IR, Homeostatic Model Assessment for Insulin Resistance; OGTT, oral glucose tolerance test.