

Figure S1

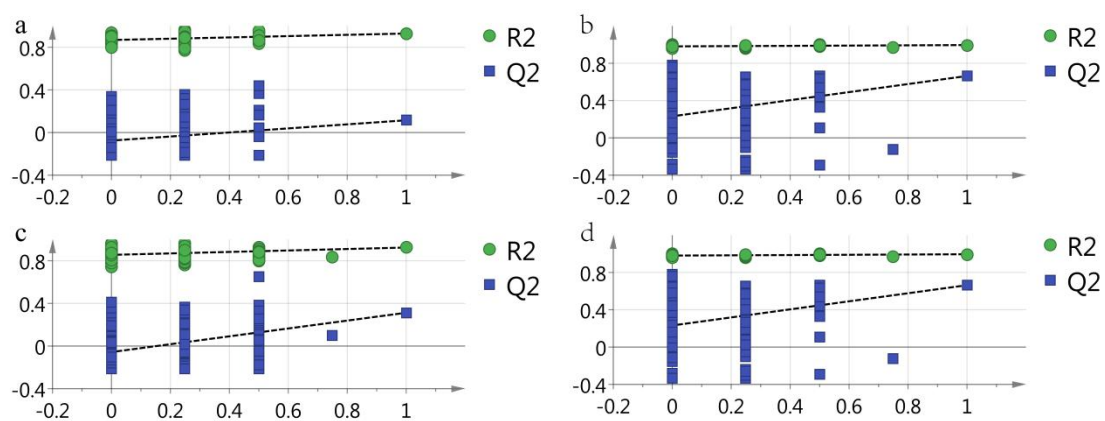


Figure S1: PLS-DA of the control and model groups based on the data obtained in the positive ion mode (a) and the negative ion mode (b); PLS-DA of the model and DST groups based on the data obtained in the positive ion mode (c) and the negative ion mode (d).

Table S1: Candidate biomarkers and their mode of action that might be related to memory impairment.

Name	Model/Control	DST/Model	Mode of action that may be related to AD
Prolylphenylalanine	↓*	↑#	-
Arachidoyl Ethanolamide	↑*	↑	Protect the heart and nervous system, improve blood pressure and immune function
Folic acid	↓*	↑	Reduce β-amyloid protein levels, tau protein hyperphosphorylation, oxidative stress
1-hexadecanoyloxy-3-hydroxypropan-2-yl-octadec-9-enoate	↑*	↓#	-
Carnosine	↓**	↑#	Oxidative stress, reduce mitochondrial damage
CMPF	↓*	↑###	Oxidative stress
6-Aminopenicillanic acid	↓*	↑###	-
N-Acetylleucine	↓*	↑	-
Glycine	↓*	↑#	Oxidative stress, inflammatory/immune response
Trehalose-6-phosphate	↓*	↑	-
3-Methyl-L-histidine	↓**	↑###	-
Mesaconic acid	↑*	↑	Apoptosis and oxidative stress
Deoxyguanylic acid	↓**	↓	-
Bilirubin	↓*	↓	Oxidative stress, neurotoxicity, amyloid, tau and neurodegeneration
PI 320	↑*	↓	-
3-Dehydrocholic acid	↑*	↓	-
Hydrocinnamic acid	↑***	↓###	-
L-Histidine	↑*	↓#	Oxidative stress
Galactosamine 1-phosphate	↑*	↓#	-
Ketoisovaleric acid	↑*	↓##	-
Asparagine	↑*	↓##	Aβ deposition

S-MALATE	↑*	↓##	-
Gly-Tyr	↑*	↓##	-
8-Hydroxycarapinic Acid	↓*	↑###	-
Pantothenic acid	↓*	↑	Aβ deposition
Creatinine	↑*	↓	Oxidative stress
Erucamide	↑***	↓	Cholinergic deficiency
Threonic acid	↓*	↑	-
Anserine	↓*	↑#	Antioxidant, anti-aging
3-hydroxybutanoic acid	↓*	↑	Oxidative stress, apoptosis, reduce free radical damage
Chlorothalonil-4-hydroxy	↓**	↑#	-
Pyroglutamic acid	↓*	↑	Reduce inflammation of the small intestine
DL-Lactic acid	↑*	↓	Oxidative stress

* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$, the model group versus the control group; # $p < 0.05$, ## $p < 0.01$, ### $p < 0.001$, the Dushen Tang (DST) group versus the model group

Table S2: List of metabolic pathways related to candidate biomarkers (p < 0.05).

Pathway name	Overlapping metabolites	P value
Beta-Alanine Metabolism	Pantothenic acid; Carnosine; L-Histidine; Anserine	1.68E-06
Metabolism of amino acids and derivatives	Creatinine; Asparagine; Carnosine; Glycine; Hydrocinnamic acid; Anserine; L-Histidine	4.89E-06
Disorders of Folate Metabolism and Transport	L-Histidine; Glycine; Folic acid	4.26E-05
SLC-mediated transmembrane transport	Pantothenic acid; Asparagine; L-Histidine; Glycine; Creatinine	6.00E-05
leukotriene biosynthesis	Asparagine; Glycine; L-Histidine	6.61E-05
Na ⁺ /Cl ⁻ -dependent neurotransmitter transporters	Asparagine; Glycine; L-Histidine	7.30E-05
Amino acid transport across the plasma membrane	Asparagine; Glycine; L-Histidine	8.04E-05
carnosine biosynthesis	Carnosine; L-Histidine	0.000147
One carbon metabolism and related pathways	Glycine; Pyroglutamic acid; Folic acid	0.000183
Histidine metabolism	Carnosine; Glycine; L-Histidine	0.000225
Protein digestion and absorption	Asparagine; Glycine; L-Histidine	0.000256
Amino acid and oligopeptide SLC transporters	Asparagine; Glycine; L-Histidine	0.000308
Aminoacyl-tRNA biosynthesis	Asparagine; Glycine; L-Histidine	0.000347
Glutathione synthesis and recycling	Glycine; Pyroglutamic acid	0.000632
tRNA Aminoacylation	Asparagine; Glycine; L-Histidine	0.000701
Cytosolic tRNA aminoacylation	Asparagine; Glycine; L-Histidine	0.000701
Mitochondrial tRNA aminoacylation	Asparagine; Glycine; L-Histidine	0.000701
Creatine metabolism	Glycine; Creatinine	0.000728
Glutathione metabolism	Glycine; Pyroglutamic acid	0.00118
Glucose Homeostasis	3-hydroxybutanoic acid; L-Histidine	0.00144
Synthesis and degradation of ketone bodies	3-hydroxybutanoic acid	0.0164
Utilization of Ketone Bodies	3-hydroxybutanoic acid	0.0272
ketogenesis	3-hydroxybutanoic acid	0.0325

Coenzyme A biosynthesis	Pantothenic acid	0.0352
Pyruvate Carboxylase Deficiency	Glycine	0.0458
Butanoate metabolism	3-hydroxybutanoic acid	0.0494
