

Table S1 Ferroptosis-related factors

Gene	Protein	Function
<i>ABCC1</i>	ATP Binding Cassette Subfamily C Member 1	Mediates the export of organic anions and drugs from the cytoplasm
<i>ACACA</i>	Acetyl-CoA carboxylase 1	Catalyzes acetyl-CoA to malonyl-CoA
<i>ACO1</i>	Cytoplasmic aconitate hydratase	Bifunctional iron sensor
<i>ACSF2</i>	Acyl-CoA synthetase family member 2	Catalyzes the initial reaction in fatty acid metabolism
<i>ACSL3</i>	Acyl-CoA synthetase long chain family member 3	Catalyzes the conversion of long-chain fatty acids into fatty acyl-CoAs
<i>ACSL4</i>	Acyl-CoA synthetase long-chain family member 4	Converts long-chain fatty acids into acyl-CoA
<i>AKR1C1</i>	Aldo-keto reductase family 1 member C1	Catalyzes ketosteroids to hydroxysteroids
<i>CARS</i>	Cysteinyl-tRNA synthetase	Catalyzes the ligation of Cys (ATP-dependent) to transfer RNA(tRNA)
<i>CBS</i>	Cystathionine-β-synthase	Catalyzes L-homocysteine to L-cysteine
<i>CISD1</i>	CDGSH iron-sulfur domain 1	Participates in Fe-S cluster shuttling and redox reactions
<i>CS</i>	Citrate synthase	Catalyzes the first step of the citric acid cycle
<i>FADS2</i>	Acyl-CoA 6-desaturase	Participates in the biosynthesis of highly unsaturated fatty acids
<i>FANCD2</i>	Fanconi anemia group D2 protein	Is involved in DNA damage repair and maintaining chromosome stability
<i>FDFT1</i>	Squalene synthase	Catalyzes the production of the squalene
<i>FTH1</i>	Ferritin heavy chain	intracellular iron storage subunit
<i>G6PD</i>	Glucose-6-phosphate 1-dehydrogenase	Facilitates reduced form of nicotinamide adenine dinucleotide phosphate (NADPH) reduction and pentose phosphate for synthesis of fatty acids and nucleic acids
<i>GCLC</i>	Glutamate-cysteine ligase catalytic subunit	Is involved in glutathione biosynthesis
<i>LOX</i>	Lysyl Oxidase	
<i>GLS2</i>	Glutaminase 2	Is involved in the regulation of glutamine catabolism
<i>GOT1</i>	Glutamic-Oxaloacetic Transaminase 1	Is involved in the biosynthesis of glutamate
<i>GPX4</i>	Glutathione Peroxidase 4	Essential antioxidant peroxidase
<i>HMGR</i>	3-hydroxy-3-methylglutaryl-coenzyme A reductase	Catalyzes the synthesis of mevaleric acid
<i>HMOX1</i>	Heme Oxygenase 1	Catalyzes the degradation of heme to biliverdin
<i>HSBP1</i>	Heat Shock Factor Binding Protein 1	Negative regulator of the heat shock response
<i>IREB2</i>	Iron Responsive Element Binding Protein 2	RNA-binding protein that binds to Iron-responsive elements (IREs). Binding to the IRE element in ferritin results in the repression of its mRNA translation
<i>KEAP1</i>	Kelch Like Epichlorohydrin (ECH)-associated Protein 1	Targeting NFE2L2/NRF2 regulates the oxidative stress response
<i>LPCAT3</i>	Lysophosphatidylcholine Acyltransferase 3	Catalyzes fatty acid chain transfer to 1-acyl lysophospholipids to form various classes of phospholipids
<i>MIR137</i>	–	Regulates ferroptosis by targeting the glutamine transporter, SLC1A5
<i>NCOA4</i>	Nuclear Receptor Coactivator 4	Ligand-independent co-activator of the peroxisome proliferator-activated receptor (PPAR) gamma
<i>NFS1</i>	Cysteine desulfurase	Is involved in the synthesis of iron-sulfur clusters by using sulfur from cysteine
<i>NOX1</i>	NADPH Oxidase 1	a pyridine nucleotide-dependent oxidoreductase that generates superoxide
<i>NQO1</i>	NAD(P)H Quinone Dehydrogenase 1	A quinone reductase
<i>NFE2L2</i>	NFE2-like BZIP Transcription Factor 2	Key transcription factor in the response to oxidative stress
<i>PEBP1</i>	Phosphatidylethanolamine-binding protein 1	Phosphatidylethanolamine binding protein
<i>PGD</i>	Phosphogluconate Dehydrogenase	Catalyzes the oxidative decarboxylation of 6-phosphate gluconate
<i>PHKG2</i>	Phosphorylase Kinase Catalytic Subunit Gamma 2	Activation of glycogen phosphorylase and mediates glycogen breakdown
<i>SLC1A5</i>	Solute Carrier Family 1 Member 5	Sodium-dependent amino acids transporter
<i>SLC7A11</i>	Solute Carrier Family 7 Member 11	Specific binding to the anionic form of cystine and glutamate
<i>SQLE</i>	Squalene Epoxidase	Rate-limiting enzyme in steroid biosynthesis
<i>STEAP3</i>	Metalloreductase STEAP3	Reduces Fe ³⁺ to Fe ²⁺
<i>TFRC</i>	Transferrin Receptor	Cellular uptake of iron
<i>ZEB1</i>	Zinc Finger E-Box Binding Homeobox 1	Transcriptional repressor

Table S2 Copy number variation of ferroptosis-related genes in LUAD from TCGA data

Gene	CNV rate	CNV type
<i>SQLE</i>	10.71%	Amplification: 51/51
<i>FDFT1</i>	9.24%	Amplification: 1/44
<i>TFRC</i>	4.62%	Amplification: 13/22; Deletion: 9/22
<i>AKR1C1</i>	3.78%	Amplification: 14/18; Deletion: 4/18
<i>LPCAT3</i>	3.15%	Amplification: 9/15; Deletion: 6/15
<i>NFS1</i>	2.52%	Amplification: 12/12
<i>GPX4</i>	2.52%	Amplification: 3/12; Deletion: 9/12
<i>GLS2</i>	2.52%	Amplification: 12/12
<i>ACSF2</i>	2.31%	Amplification: 11/11
<i>CS</i>	2.10%	Amplification: 10/10
<i>PGD</i>	1.89%	Amplification: 7/9; Deletion: 2/9
<i>HSBP1</i>	1.89%	Amplification: 1/9; Deletion: 8/9
<i>ACO1</i>	1.89%	Amplification: 4/9; Deletion: 5/9
<i>ACACA</i>	1.89%	Amplification: 9/9
<i>NFE2L2</i>	1.68%	Amplification: 8/8
<i>MIR137</i>	1.68%	Amplification: 2/8; Deletion: 6/8
<i>KEAP1</i>	1.68%	Deletion: 8/8
<i>STEAP3</i>	1.47%	Amplification: 7/7
<i>PHKG2</i>	1.47%	Amplification: 7/7
<i>SLC1A5</i>	1.26%	Amplification: 5/6; Deletion: 1/6
<i>HMOX1</i>	1.26%	Amplification: 4/6; Deletion: 2/6
<i>ZEB1</i>	1.05%	Amplification: 5/5
<i>PEBP1</i>	1.05%	Amplification: 3/5; Deletion: 2/5
<i>HMGCR</i>	1.05%	Amplification: 2/5; Deletion: 3/5
<i>GCLC</i>	1.05%	Amplification: 4/5; Deletion: 1/5
<i>LOX</i>	0.84%	Amplification: 1/4; Deletion: 3/4
<i>IREB2</i>	0.84%	Amplification: 4/4
<i>FADS2</i>	0.84%	Amplification: 4/4
<i>CISD1</i>	0.84%	Amplification: 2/4; Deletion: 2/4
<i>CBS</i>	0.84%	Amplification: 3/4; Deletion: 1/4
<i>SLC7A11</i>	0.63%	Amplification: 2/3; Deletion: 1/3
<i>NQO1</i>	0.63%	Amplification: 2/3; Deletion: 1/3
<i>NCOA4</i>	0.63%	Amplification: 2/3; Deletion: 1/3
<i>FTH1</i>	0.63%	Amplification: 3/3
<i>ACSL3</i>	0.63%	Amplification: 3/3
<i>FANCD2</i>	0.42%	Amplification: 1/2; Deletion: 1/2
<i>GOT1</i>	0.21%	Deletion: 1/1

LUAD, lung adenocarcinoma; TCGA, The Cancer Genome Atlas; CNV, copy number variation

Table S3 Expression of six ferroptosis-related factors in the tumor tissues of 12 LUAD patients

Patient	GPX4	NRF2	KEAP1	FTH1	ACSL4	LPCAT3
Protein (tumor vs. adjacent)						
Patient #1	High	Low	Low	High	High	High
Patient #2	High	Low	High	Low	Consistent	High
Patient #3	Consistent	Low	Low	High	High	High
Patient #4	High	Low	Low	High	Consistent	High
Patient #5	Low	Low	Low	High	High	High
Patient #6	High	Low	Low	High	Consistent	Low
Patient #7	High	Low	Low	Consistent	High	High
Patient #8	High	Low	Low	Consistent	High	High
Patient #9	High	Low	Low	High	High	High
Patient #10	Consistent	Low	Low	Low	Consistent	Consistent
Patient #11	Low	Low	Low	High	Low	Low
Patient #12	High	Consistent	Low	Low	Consistent	High
Total	High: 8/12; low: 2/12; consistent: 2/12	High: 0; low: 11/12; consistent: 1/12	High: 1/12; low: 11/12; consistent: 0	High: 7/12; low: 3/12; consistent: 2/12	High: 6/12; low: 1/12; consistent: 5/12	High: 9/12; low: 2/12; consistent: 1/12

LUAD, lung adenocarcinoma.

Table S4 Cox regression analysis of six ferroptosis-related factors in LUAD

Variables	Univariate analysis			Multivariate analysis		
	HR	95% CI	P value	HR	95% CI	P value
GPX4	1.540	1.137-2.087	0.005*	1.606	1.174-2.198	0.003*
NRF2	1.585	1.029-2.442	0.037*	1.944	1.243-3.039	0.004*
KEAP1	0.751	0.539-1.047	0.091	0.751	0.533-1.059	0.102
FTH1	0.744	0.515-1.076	0.116	0.753	0.514-1.102	0.144
ACSL4	0.694	0.507-0.949	0.022*	0.739	0.537-1.016	0.063
LPCAT3	1.518	1.018-2.265	0.041*	1.371	0.909-2.068	0.132
Stage	1.621	1.407-1.867	2.18e-11*	1.591	1.375-1.842	4.48e-10*
Age	1.007	0.991-1.022	0.408			
Sex	1.059	0.783-1.431	0.71			

*, P<0.05. LUAD, lung adenocarcinoma; HR, Hazard ratio; CI, confidence interval.

Table S5 Correlation between GPX4 expression and clinicopathological variables in LUAD patients from tissue microarray

Clinicopathological variables	n	GPX4 expression		χ^2 value	P value
		Low	High		
All patients	82	39	43		
Age (years)				2.204	0.138
<65	39	21	18		
≥65	43	30	13		
Gender				1.685	0.194
Male	39	21	18		
Female	43	17	26		
TNM stage ¹				0.006	0.941
I-II		26	13		
III-IV		29	14		
Lymph node metastasis				0.005	0.939
Negative		23	16		
Positive		25	18		
Tumor size				0.339	0.560
≤3 cm		13	26		
>3 cm		17	26		
Tissue types				7.922	0.005*
Tumor	62	44	18		
Adjacent	82	39	43		

*, P<0.05. LUAD, lung adenocarcinoma; GPX4, Glutathione Peroxidase 4; ADC, adenocarcinoma; TMA, tissue microarray; TNM, tumor-node-metastasis.

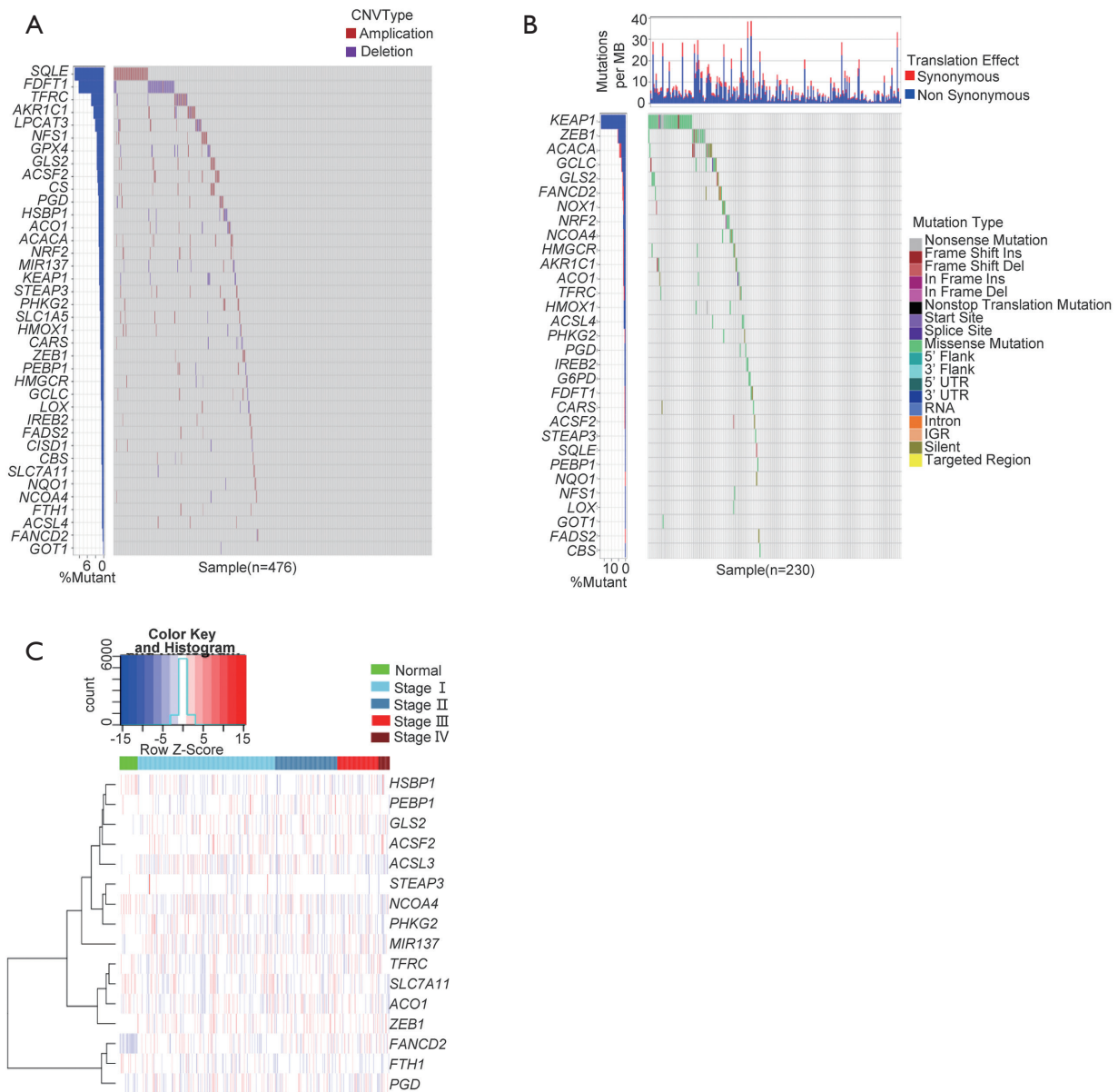


Figure S1 The expression of ferroptosis-related factors in LUAD. (A) CNV profile of ferroptosis-related genes in clinical LUAD samples. (B) Mutation profile of ferroptosis-related genes in clinical LUAD samples. (C) Promoter region DNA methylation profile of ferroptosis-related genes in clinical LUAD samples. LUAD, lung adenocarcinoma; CNV, copy number variation.

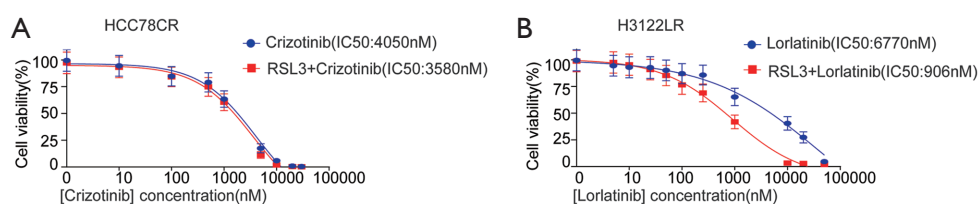


Figure S2 TKI or/and RSL3 synergistically targeted drug-resistant cells. (A) Proliferation inhibition curves of HCC78CR cells after treatment with crizotinib or RSL3 plus Crizotinib. (B) Proliferation inhibition curves of H3122LR cells after treatment with lorlatinib or RSL3 plus lorlatinib.