

	LUAD	tissues	CTCs			
Variables	Training cohort Study cohort (n=40) (n=54)		Detection by CanPatrol [™] (n=80)*	Simultaneous Detection of CD274 (n=20)	Detection by CytoploRare (n=21)	
Gender(male: female)	16:24	24:30	39: 41	13: 7	8:13	
Age(year)	58.60±9.46	60.4±9.1	58.10±10.30	64.58±7.51	55.85±13.30	
Tumor size(cm)	3.07±1.21	2.3±0.7	1.93±0.91	2.34±1.11	1.93±1.43	
Stage(1a:1b:2a)	22:13:5	46:8:0	70:8:2	14: 4: 2	19:2 : 0	
Survive time(days)	OS:2732.1±880.8	DFS:582.5±385.9	DFS:582.66±375.60	N.A.	N.A.	

Table S1. Clinical and demographical characteristics of training cohort and study cohort	
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Note: * 80 cases included 54 cases in Study Cohort.

name	Cat number	company	dilution rate	usage
BCAR1	#13846S	CST	1:200/1: 1000/1: 200	IHC/WB/IF
BCAR1	SC-20029	Santa cruz	1:500	IP
CD274	# 13684S	CST	1: 100/1:1000	IHC/WB
BRD4	ab128874	Abcam	1:200/1:500/1:1000	IHC/IP/WB
RAC1	ab33186	Abcam	1:50/1:1000	IHC/WB
CD8	ab17147	Abcam	1:50	IHC
CD4	ab133616	Abcam	1:200	IHC
CDH1	#14472S	CST	1:1000	WB
CDH2	#13116S	CST	1:1000	WB
Vimentin	#5741S	CST	1:1000	WB
GAPDH	#2118	CST	1:1000	WB
TSG101	ab125011	Abcam	1:1000	WB
Flag	F1804	Sigma	1:2000	IP

Table S2. information of antibodies

Table S3. Primers (5'->3') of probes for BCAR1 and CD274 detection in CTC.

ID	seq
BCAR1	cacgtcgtagaggtcaggag;gcatacaccactgtcgac;cagaaggtgggcaacggtgg;cagatgtgtgggcagcattg;
	tgtaggtggacgtagtcata;tgttccagtcgttcaaactg;cactgctccaggtagaagag;cggtaaagaaggcgtccacg
CD274	ctactgggaatttgcattca; tagtgcagccaggtctaatt; tcctctccatgcacaaattg; tgtagctactatgctgaacc;
	gagagctggtccttcaacag; gatctgaagtgcagcatttc; catcctgcaatttcacatct; ctgatcatgcagcggtacac

	= /		<u><u></u></u>
NO.	5	STEM	3
BCAR1-sgRNA-a	CACCg	CAGCAACGGGTCTCGGCCAT	
BCAR1-sgRNA-b	aaac	ATGGCCGAGACCCGTTGCTG	С
5			
NC		CCCTTCCCCCCCCCTTCAA	

Table S4. Synthetic oligos for BCAR1 sgRNA construction

Table S5. Primers of construction of cDNA library of BCAR1

ID	seq
BCAR1(45429-1)-P1	AGGTCGACTCTAGAGGATCCCGCCACCATGCCTGCCAAGCCCTTCCTCTTCTG
BCAR1(45429-1)-P2	CACAGGCTAGCTCAACCGGTTCAGGCGGCTGCCAGCTGGCCTAGGACGCGG

Gene	Upstream primer sequences	downstream primer sequences	Amplified fragment size (bp)
GAPDH	TGACTTCAACAGCGACA CCCA	CACCCTGTTGCTGTAGCC AAA	121
BCAR1	CAATGCCTCACTGCTCT TCA	CTCCTTCTGGGTCTTCTC AAAC	247

Table S6. Primers of Q-PCR to detect BCAR1 expression in cells

Gene	Upstream primer sequences	downstream primer sequences	Amplified fragment size (bp)
GAPDH	TGACTTCAACAGCGACACCC A	CACCCTGTTGCTGTAGCCA AA	121
CD274	ACTGGCATTTGCTGAACG	TCCTCCATTTCCCAATAGAC	148

Table S7. Primers of Q-PCR to detect CD274 expression in cells

Table S8 Primers of the construction and identification of FLAG-RAC1 vector

Purpose	ID	seq
Construction of vector RAC1(63107-6)-p1 GAGGATCCCCGGGTACCGGTCGCCACCATGGAC		GAGGATCCCCGGGTACCGGTCGCCACCATGGACTACAAGGATGACGATG
	RAC1(63107-6)-p2	CACACATTCCACAGGCTAGCTTACAACAGCAGGCATTTTCTCTTC
PCR identification	RAC1(63107-6)-p3	GGGTCAATATGTAATTTTCAGTG
	RAC1(63107-6)-p4	CGTCGCCGTCCAGCTCGACCAG

Figure2A	H1975		H1299		A549		
	NC	KO	NC KO		NC	OE	
BCAR1-gray value	90197.588	50284.35	86894.856	47738.463	61985.309	84887.836	
GAPDH-gray value	83499.128	84346.16	82677.924	81200.714	87430.501	88206.602	
Relative BCAR1	1.0802219	0.596166	1.051004329	0.587906936	0.708966645	0.962375084	

Table S9Gray values of WB bands

Figure2E	EXSOMES					
		OE	NC			
	RAC1-gray value	65233.019	26003.43			
	TSG101-gray value	71831.693	60072.93			
	Relative RAC1	0.908137	0.432864			

Figure2F		H19	975	H1299		A549	
		NC	KO	NC	KO	NC	OE
	RAC1-gray value	70275.208	49973.7	74572.48	35744.712	48657.501	76248.534
	GAPDH-gray value	66460.693	60104.14	64833.338	62439.986	53402.643	74803.472
	Relative RAC1	1.0573951	0.831452	1.150218118	0.572465087	0.911144061	1.019318114
	CDH2-gray value	9884.983	5621.184	16120.276	12424.619	8602.326	23759.276
	GAPDH-gray value	14352.912	14109.84	21986.083	21072.548	12760.104	21245.518
	Relative CDH2	0.6887092	0.398387	0.733203636	0.589611612	0.674157985	1.11831945

Vimentin-gray value	26851.326	10861.26	24154.154	11020.205	8741.912	15779.276
GAPDH-gray value	18609.548	18468.74	22079.69	21890.962	23194.205	21764.548
Relative Vimentin	1.442879	0.588089	1.093953493	0.503413464	0.376900696	0.724999021

Figure4B			H1	1975		H1299				A549			
		Total		Nucleus		Total		Nucleus		Total		Nucleus	
		NC	KO	NC	KO	NC	KO	NC	KO	NC	OE	NC	OE
	BRD4-L gray	0	0	55844.3	70220.2	0	0	52615.7	62904.4	3462.98	3824.50	4692.87	4562.97
	value			62	38			54	75		7	5	
	BRD4-S gray	26595.9	27705.	80201.7	61082.1	64658.2	50139.6	35124.9	24563.5	17439.5	18493.9	39519.3	44851.1
	value	66	77	47	39	91	61	34	37	4	8	43	15
	GAPDH-gray	79754.0	52731.	85803.3	91723.3	69898.2	57636.5	81906.1	85731.2	28029.1	24961.9	43871.3	51971.2
	value	57	14	21	8	79	57	58	5	25	83	09	96

Figure4C				Nu	cleus			Total						
		T1	N1	T2	N2	Т3	N3		T1	N1	T2	N2	T3	N3
	BRD4-L	0	0	52533.	0	0	0	BRD4 gray	7340.2	4553.7	32823.	1446.2	9468.2	1084.2
	gray value			919				value	05	9	045	13	96	34
	BRD4-S	1263.9 252 6 75	2529.	29933.	0	26549.	41477.	CD274 gray	1115.1	1883.7	1907.8	5141.5	30146.	6780.9
	gray value		75	208		803	442	value	63	7	2	98	681	62
	GAPDH-gr	19068.	1892	41212.	10586.	24698.	20609.	GAPDH-gray	30498.	16466.	28827.	14410.	24625.	34259.
	ay value	104	0.3	811	933	761	347	value	205	477	861	497	033	225