

Supplement Table 1 Gene list and probe region

gene symbol	Ref_seq transcript ID	DNA probes	RNA probes
<i>ALK</i>	NM_004304	CDS+UTR+intron	CDS+UTR
<i>BRAF</i>	NM_004333	CDS+UTR+intron	CDS+UTR
<i>BRD3</i>	NM_007371	CDS+UTR	CDS+UTR
<i>BRD4</i>	NM_058243	CDS+UTR	CDS+UTR
<i>EGFR</i>	NM_005228	CDS+UTR+intron	CDS+UTR
<i>FGFR1</i>	NM_023110	CDS+UTR+intron	CDS+UTR
<i>FGFR2</i>	NM_000141	CDS+UTR+intron	CDS+UTR
<i>FGFR3</i>	NM_000142	CDS+UTR+intron	CDS+UTR
<i>FGFR4</i>	NM_213647	CDS+UTR	CDS+UTR
<i>MET</i>	NM_001127500	CDS+UTR+intron	CDS+UTR
<i>NRG1</i>	NM_013957	CDS+UTR	CDS+UTR
<i>NTRK1</i>	NM_002529	CDS+UTR+intron	CDS+UTR
<i>NTRK2</i>	NM_006180	CDS+UTR+intron	CDS+UTR
<i>NTRK3</i>	NM_002530	CDS+UTR+intron	CDS+UTR
<i>RET</i>	NM_020975	CDS+UTR+intron	CDS+UTR
<i>ROS1</i>	NM_002944	CDS+UTR+intron	CDS+UTR

Supplement Table 2 Reference standards information

Sample name	product catalog number	company	type	fusion	expected fractional abundance
SD1	CA2192	GeneWell	DNA	<i>EML4::ALK</i> <i>CD74::ROS1</i> <i>SLC34A2::ROS1</i> <i>CCDC6::RET</i> <i>EML4::ALK</i>	8%
SD2	CA2193	GeneWell	DNA	<i>CD74::ROS1</i> <i>SLC34A2::ROS1</i> <i>CCDC6::RET</i> <i>EML4::ALK</i>	5%
SD3	CA2194	GeneWell	DNA	<i>CD74::ROS1</i> <i>SLC34A2::ROS1</i> <i>CCDC6::RET</i> <i>EML4::ALK</i>	2.5%
SD1	CA2197	GeneWell	RNA	<i>CD74::ROS1</i> <i>SLC34A2::ROS1</i> <i>CCDC6::RET</i> <i>EML4::ALK</i>	1000-2000 copies/100ng
SD2	CA2198	GeneWell	RNA	<i>CD74::ROS1</i> <i>SLC34A2::ROS1</i> <i>CCDC6::RET</i> <i>EML4::ALK</i>	500-800 copies/100ng
SD3	CA2199	GeneWell	RNA	<i>CD74::ROS1</i> <i>SLC34A2::ROS1</i> <i>CCDC6::RET</i> <i>BTBD1::NTRK3</i> <i>ETV6::NTRK3</i>	250-400 copies/100ng
SD4		Origimed	DNA	<i>LMNA::NTRK1</i> <i>NACC2::NTRK2</i> <i>QKI::NTRK2</i> <i>TPM3::NTRK1</i>	6%
				<i>BTBD1::NTRK3</i> <i>ETV6::NTRK3</i>	
				<i>LMNA::NTRK1</i> <i>NACC2::NTRK2</i> <i>QKI::NTRK2</i> <i>TPM3::NTRK1</i>	
				<i>BTBD1::NTRK3</i> <i>ETV6::NTRK3</i>	
		Origimed	RNA	<i>LMNA::NTRK1</i> <i>NACC2::NTRK2</i> <i>QKI::NTRK2</i> <i>TPM3::NTRK1</i>	1000 copies/100ng
				<i>BTBD1::NTRK3</i> <i>ETV6::NTRK3</i>	
				<i>LMNA::NTRK1</i> <i>NACC2::NTRK2</i> <i>QKI::NTRK2</i> <i>TPM3::NTRK1</i>	
				<i>BTBD1::NTRK3</i> <i>ETV6::NTRK3</i>	

Supplement Table 3 Tumor type and nucleic acid yields in clinical samples

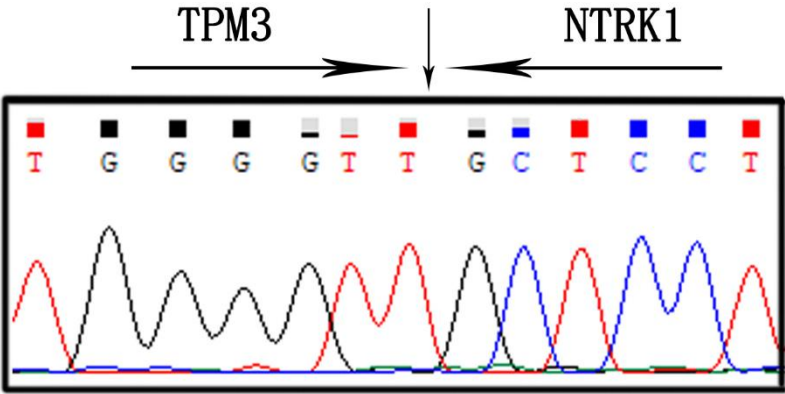
Sample no.	Tumor type	tumor cellularity	DNA yields (ng)	RNA yields (ng)
PS01	chondrosarcoma	90%	9840	17280
PS02	undifferentiated pleomorphic sarcoma	90%	3150	2004
PS03	endometrial stromal sarcoma	85%	5200	16200
PS04	malignant melanoma	70%	2650	3060
PS05	lung adenocarcinoma	20%	1100	2250
PS06	papillary thyroid carcinoma	60%	3340	14100
PS07	lung adenocarcinoma	50%	3440	13800
PS08	lung adenocarcinoma	80%	3300	10620
PS09	lung adenocarcinoma	70%	3890	14580
PS10	lung adenocarcinoma	70%	3920	8760
PS11	lung adenocarcinoma	70%	690	4200
PS12	lung adenocarcinoma	40%	3640	5640
PS13	hamartoma	40%	5400	5640
PS14	lung adenocarcinoma	60%	5100	2622
PS15	lung adenocarcinoma	20%	3000	10680
PS16	lung adenocarcinoma	40%	3040	5400
PS17	lung adenocarcinoma	40%	5660	5388
PS18	lung adenocarcinoma	70%	2740	2916
PS19	lung adenocarcinoma	70%	2890	12600
PS20	lung adenocarcinoma	70%	2830	5220
PS21	lung adenocarcinoma	70%	3020	5820
PS22	lung adenocarcinoma	70%	3960	19500
PS23	lung adenocarcinoma	35%	3240	6900
PS24	epithelioid hemanioendometrioma	20%	84.5	780
PS25	lung adenocarcinoma	50%	4050	11490
PS26	myxofibrosarcoma	80%	7300	6480
PS27	dermatofibrosarcoma protuberans	80%	8060	13320
PS28	renal cell carcinoma	20%	2390	4440
PS29	lung adenocarcinoma	30%	1540	2622
PS30	lung adenocarcinoma	30%	3100	7980
PS31	gastrointestinal schwannoma	80%	4300	8700
PS32	intestinal adenocarcinoma	80%	3560	13800
PS33	endometrial stromal sarcoma	70%	4390	9960
PS34	malignant melanoma	70%	3880	12900
PS35	papillary thyroid carcinoma	80%	4030	5160
PS36	papillary thyroid carcinoma	80%	1510	3720
PS37	papillary thyroid carcinoma	80%	7240	7860
PS38	papillary thyroid carcinoma	70%	2680	4560
PS39	pelvic spindle cell tumor	80%	4070	11280
PS40	thyroid adenoma	60%	427	2256
PS41	papillary thyroid carcinoma	60%	1270	2520
PS42	thyroid adenoma	70%	2000	4740
PS43	pelvic spindle cell tumor	70%	3950	6540
PS44	small cell malignant tumor	90%	9380	30360
PS45	endometrial stromal sarcoma	80%	3580	9660
PS46	renal clear cell carcinoma	50%	1370	1824
PS47	small cell malignant tumor	80%	3320	7620
PS48	fibroblastic/myofibroblastic tumours	40%	4180	4896
PS49	phosphaturic mesenchymal tumor	65%	4620	6480
PS50	skeletal muscle fibrous tumor	80%	2430	2802
PS51	nephroblastoma	80%	3800	22920
PS52	spindle cell sarcoma	90%	3970	8820
PS53	undifferentiated pleomorphic sarcoma	80%	3130	12780
PS54	leiomyosarcoma	90%	3580	11520
PS55	undifferentiated spindle cellsarcoma	70%	3580	18900
PS56	round cell malignant tumor	80%	3420	12300
PS57	gastric adenocarcinoma	60%	2760	3960
PS58	keratinizing squamous cell carcinoma	80%	5800	10440
PS59	malignant mesenchymal tumor	70%	4370	4020
PS60	breast carcinoma	80%	1730	10680

Supplement Table 4 DNA Assay Performance Characteristics for Fusions

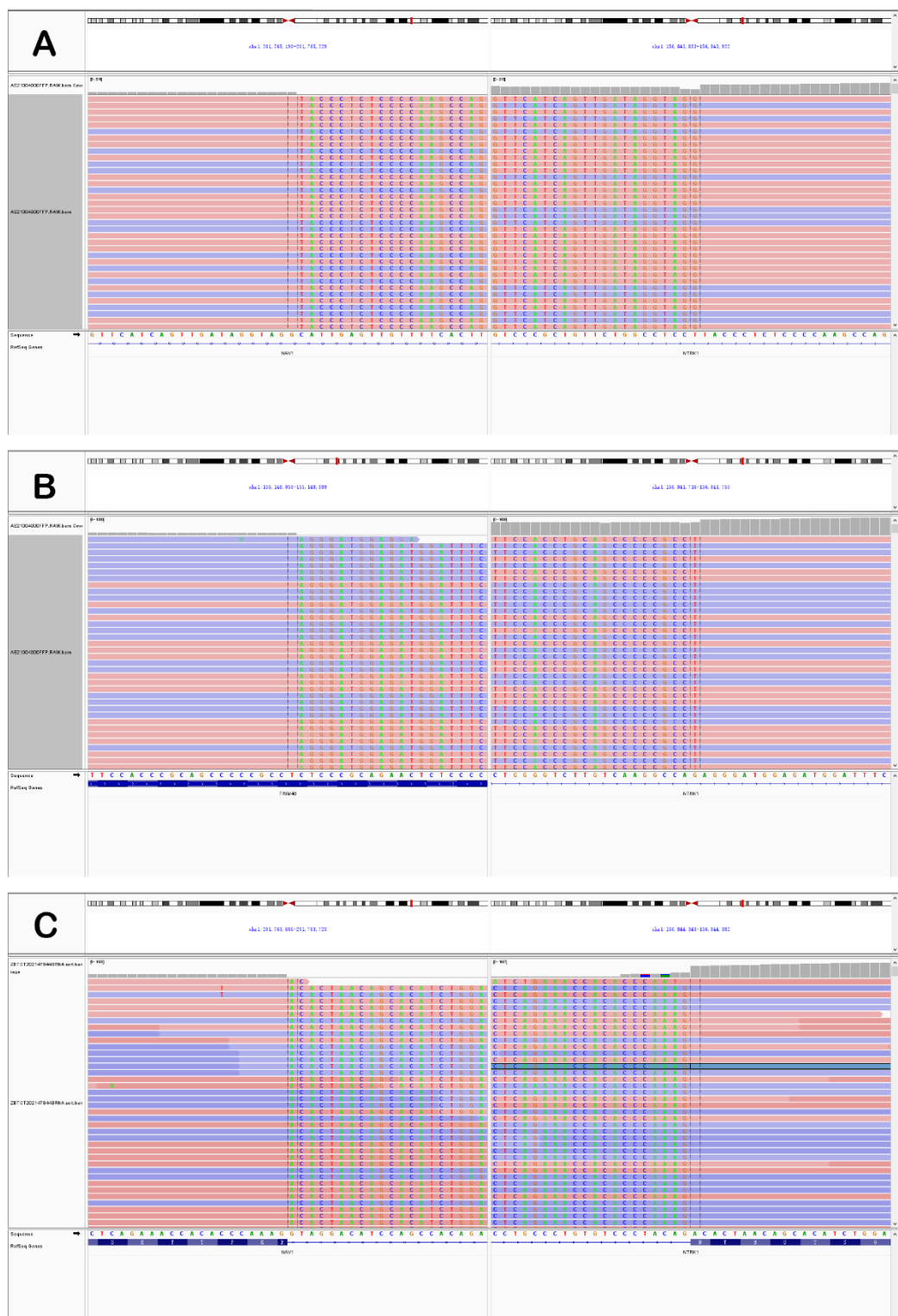
this assay (DNA)	previous results		Performance
	Positive	Negative	
Positive	26	1	96.3% (PPV)
Negative	3	31	91.2% (NPV)
	86.7%	96.9%	93.4% (accuracy)
Performance	(sensitivity)	(specificity)	

Supplement Table 5 RNA Assay Performance Characteristics for Fusions

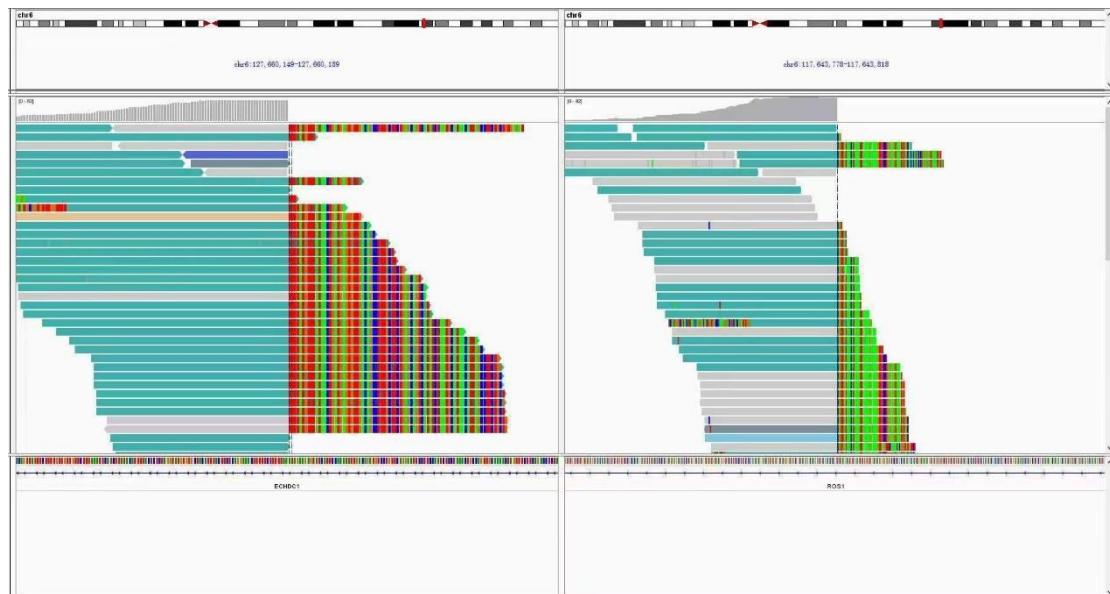
this assay (RNA)	previous results		Performance
	Positive	Negative	
Positive	22	1	95.7% (PPV)
Negative	7	31	81.6% (NPV)
	75.9%	96.9%	86.9% (accuracy)
Performance	(sensitivity)	(specificity)	



Supplemental Figure 1 A *TPM3::NTRK1* fusion was additionally identified in one fusion-negative thyroid papillary carcinoma sample (PS36) as determined by our laboratory's previous test. This *NTRK1* fusion was confirmed by Sanger sequencing.



Supplemental Figure 2 NAV1::NTRK1 (A) and TRIM46::NTRK1 (B) were simultaneously detected at the DNA level in PS04 sample, but only NAV1::NTRK1 undergoes transcription at the RNA level (C).



Supplemental Figure 3 A *ECHDC1::ROS1* rearrangement was detected at the DNA level in PS24 sample, and both *ROS1* and *ECHDC1* genes have only 3' ends left.