Electronic Supplementary Material (ESI) for Lab on a Chip.

Supplementary Material

Detection of leukemia markers using long-range surface plasmon waveguides functionalized with Protein G

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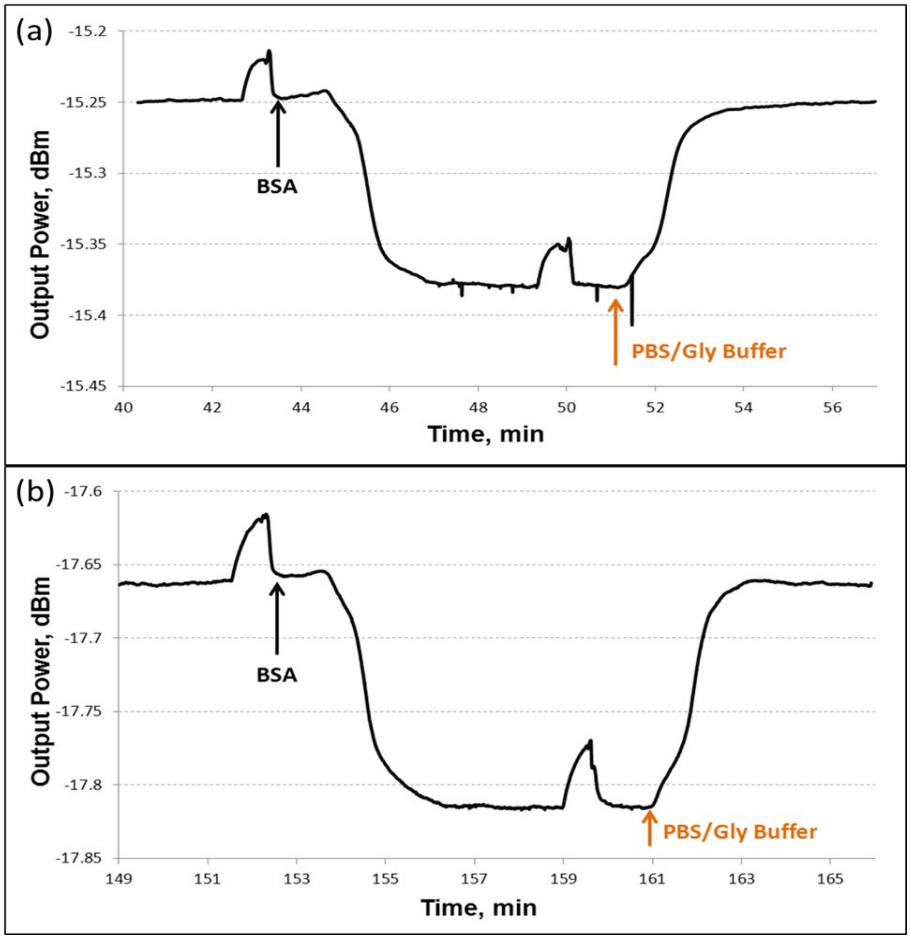

Fig. S1 Blocking non-specific binding sites with bovine serum albumin (BSA, 1 mg/ml): (a) after Protein G functionalization step and (b) after first IgG functionalization step (no prior BSA blocking). Shown results are for validation experiments of human IgGκ:AK using direct approach. Decreased output power after BSA injection is due to the RI bulk effect.

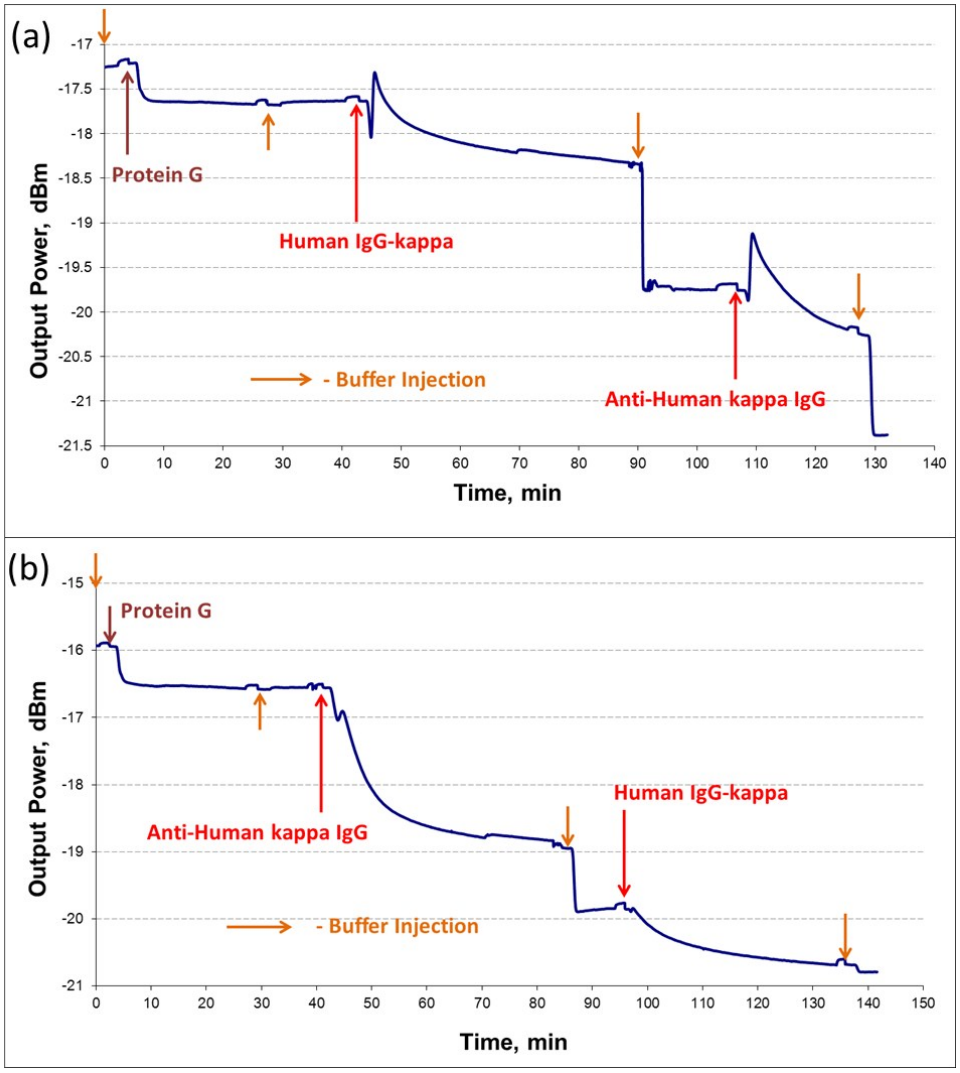

Fig. S2 Full experimental procedures for validation with standard human IgGκ and AK using both approaches: (a) Reverse approach; (b) Direct approach.

Table S3A Experimental results and computational analysis to obtain binding strength for validation of human IgGκ/λ and goat anti-human κ/λ interactions for Fig. 4 in the main text. Reverse approach.

Surface Testing	Test	Replicate	Step	$Response \left[\frac{P_{ao}}{P_{a1}} - 1\right] \times 100$	$Binding\ Strength, g/g \left[\frac{\Delta\Gamma(analyte)}{\Delta\Gamma(surface)}\right] \times 100$
KAPPA	Kappa Binding Strength	1	PG+IgGκ	**62.55**	**72.27**
			IgGκ+AK	**45.21**	
		2	PG+IgGκ	**86.64**	**58.47**
			IgGκ+AK	**50.66**	
		3	PG+IgGκ	**46.89**	**62.10**
			IgGκ+AK	**29.12**	
	Average Binding Strength				**64.28**
	Standard Deviation				**7.15**
	Kappa-Lambda Cross-reactivity	1	PG+IgGκ	**68.27**	**14.81**
			IgGκ+AL	**2.33**	
		2	PG+IgGκ	**73.78**	**48.16**
			IgGκ+AL	**3.04**	
		3	PG+IgGκ	**36.14**	**13.66**
			IgGκ+AL	**1.86**	
	Average Binding Strength				**4.22**
	Standard Deviation				**0.87**
LAMBDA	Lambda Binding Strength	1	PG+IgGλ	**77.83**	**48.06**
			IgGλ+AL	**37.40**	
		2	PG+IgGλ	**52.05**	**52.54**
			IgGλ+AL	**27.35**	
		3	PG+IgGλ	**43.55**	**60.79**
			IgGλ+AL	**26.47**	
	Average Binding Strength				**53.80**
	Standard Deviation				**6.46**
	Lambda-Kappa Cross-reactivity	1	PG+IgGλ	**75.79**	**5.84**
			IgGλ+AK	**4.42**	
		2	PG+IgGλ	**34.28**	**5.12**
			IgGλ+AK	**1.76**	
		3	PG+IgGλ	**53.11**	**6.95**
			IgGλ+AK	**3.69**	
	Average Binding Strength				**5.96**
	Standard Deviation				**0.75**

Table S3B Experimental results and computational analysis to obtain binding strength for validation of human IgGκ/λ and goat anti-human κ/λ interactions for Fig. 4 in the main text. Direct approach.

Test	Step	$Response \left[\frac{P_{ao}}{P_{a1}} - 1\right] \times 100$	$Binding\ Strength,\ g/g \left[\frac{\Delta\Gamma(analyte)}{\Delta\Gamma(surface)}\right] \times 100$
Kappa Binding Strength	PG+AK	**113.80**	**21.24**
	AK+IgGκ	**24.17**	
Kappa-lambda Cross-reactivity	PG+AK	**106.54**	**3.30**
	AK+IgGλ	**3.51**	
Lambda Binding Strength	PG+AL	**162.42**	**16.12**
	AL+IgGλ	**26.18**	
Lambda-Kappa Cross-reactivity	PG+AL	**137.68**	**4.84**
	AL+IgGκ	**6.66**	

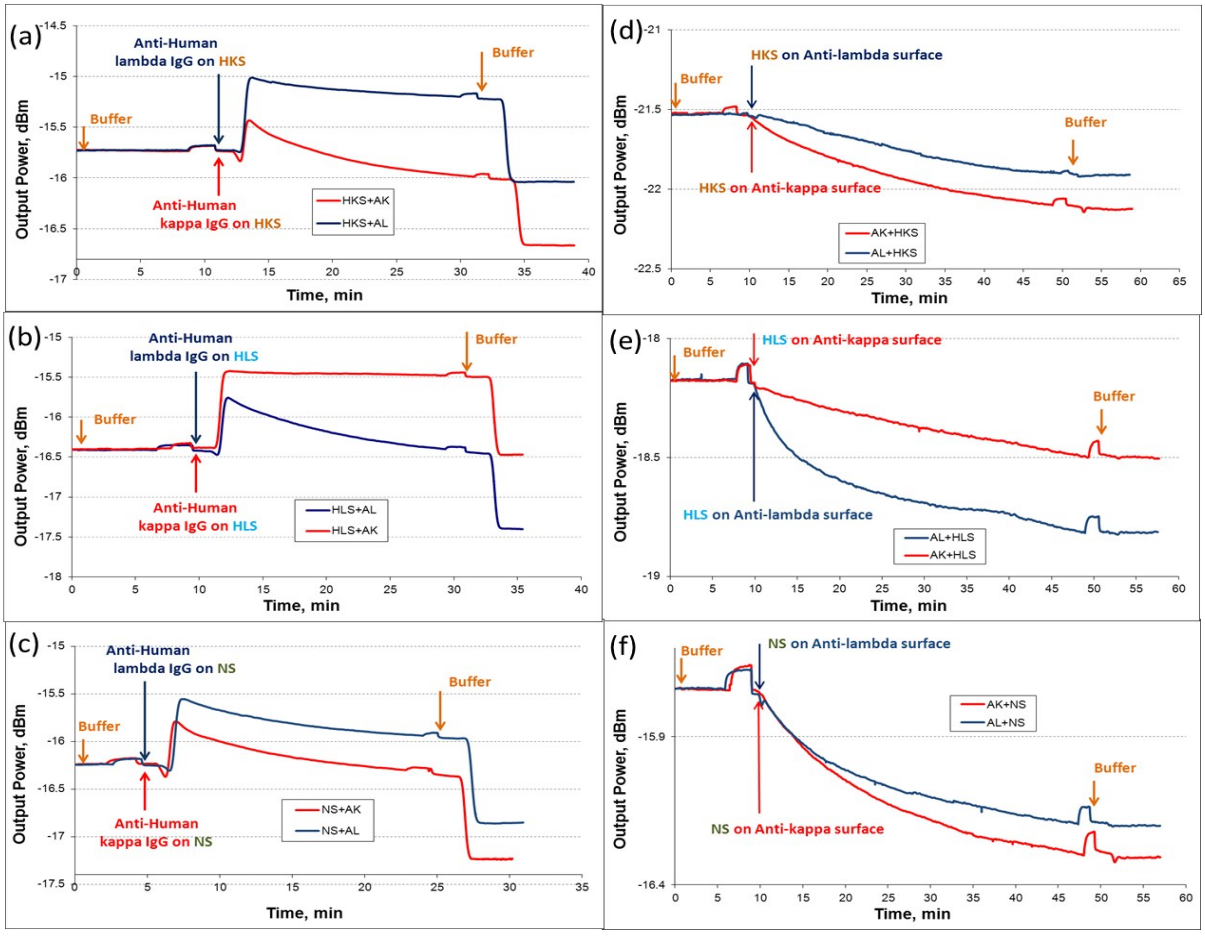

Fig. S4. Real-time responses for HKS, HLS and NS. Reverse approach: (a) Response of HKS-functionalized surface for AK and AL. (b) Response of HLS-functionalized surface for AK and AL. (c) Response of NS-functionalized surface for AK and AL. Direct approach: (d) Response of AK- and AL-functionalized surface for HKS, (e) Response of AK- and AL-functionalized surface for HLS, (f) Response of AK- and AL-functionalized surface for NS.

Table S5. Experimental results and computational analysis to obtain κ-λ ratio for HKS, HLS and NS; direct approach.

Patient Sample	Step	$Response \left[\frac{P_{ao}}{P_{a1}} - 1\right] \times 100$	$Binding\ Strength,\ g/g \left[\frac{\Delta\Gamma(recognition\ IgG)}{\Delta\Gamma(serum\ response)}\right] \times 100$	IgG light chain Ratio
High Kappa Serum	PG+AK	**53.11**	**26.41**	2.61 (κ:λ)
	AK+HKS	**14.02**		
	PG+AL	**53.82**	**10.11**	
	AL+HKS	**5.44**		
High Lambda Serum	PG+AL	**84.93**	**19.01**	2.55 (λ:κ)
	AL+HLS	**16.14**		
	PG+AK	**102.77**	**7.44**	
	AK+HLS	**7.65**		
Normal Serum	PG+AK	**52.76**	**27.08**	1.72 (κ:λ)
	AK+NS	**14.29**		
	PG+AL	**71.00**	**15.74**	
	AL+NS	**11.17**		