



## Supporting Information

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Ultra-Specific Zeptomole MicroRNA Detection by  
Plasmonic Nanowire Interstice Sensor with Bi-Temperature  
Hybridization

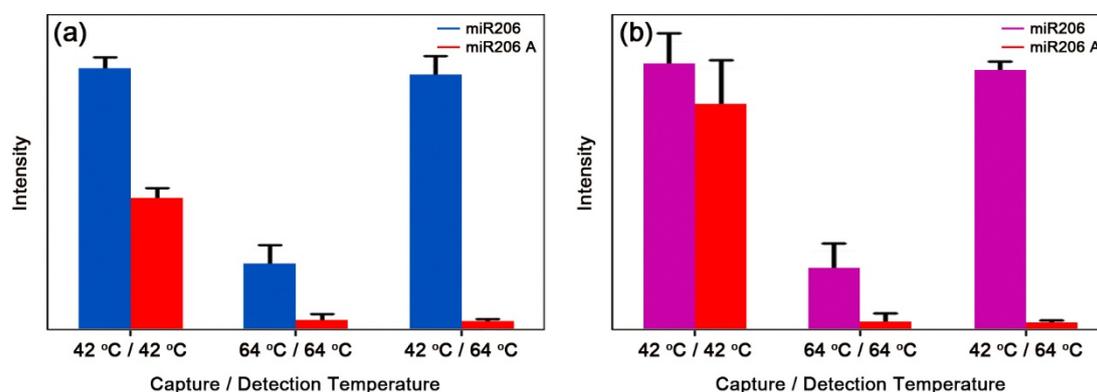
*Taejoon Kang, Hongki Kim, Jeong Min Lee, Hyoban Lee,  
Yun-Seok Choi, Gyeongwon Kang, Min-Kyo Seo, Bong Hyun  
Chung, Yongwon Jung,\* and Bongsoo Kim\**

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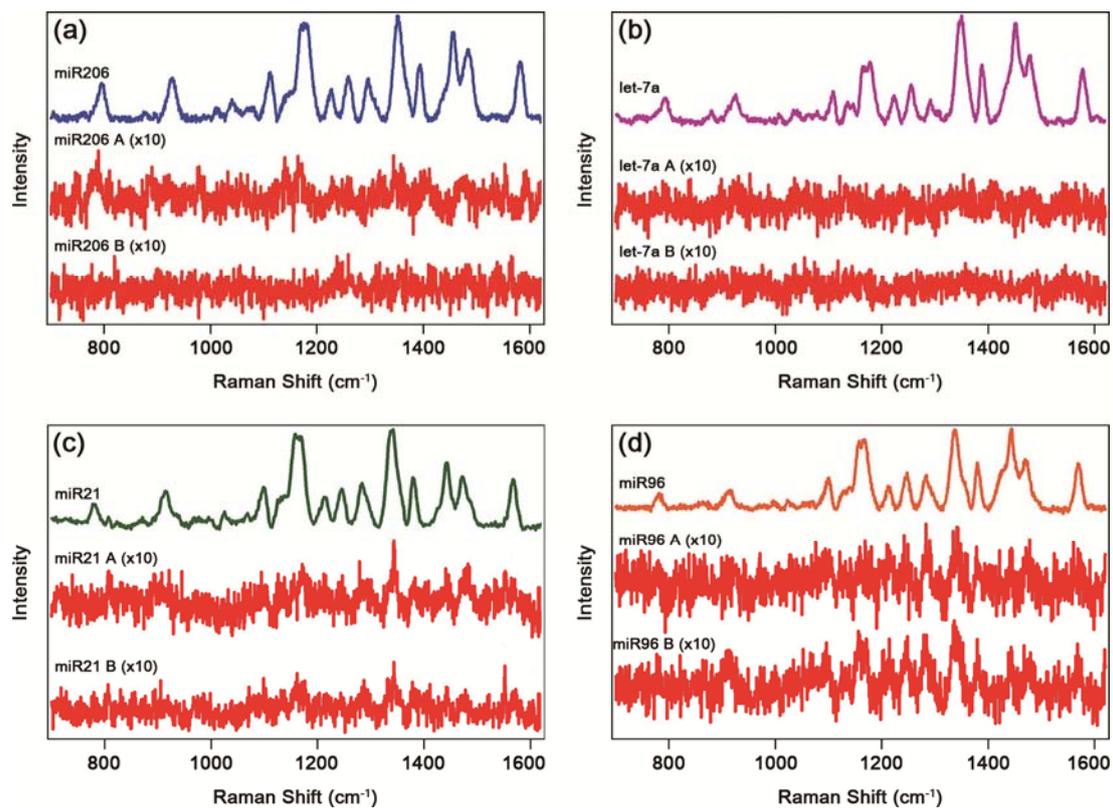
## Supporting Information

### Ultra-Specific Zeptomole MicroRNA Detection by Plasmonic Nanowire Interstice Sensor with Bi-Temperature Hybridization

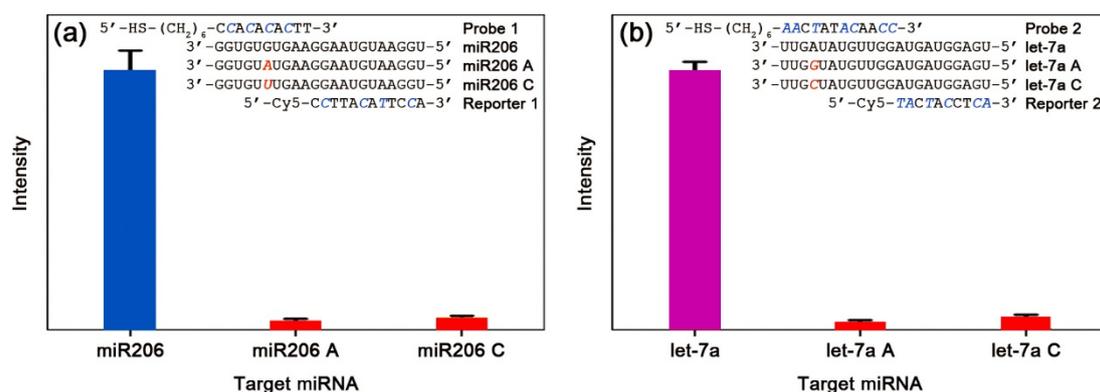
Taejoon Kang, Hongki Kim, Jeong Min Lee, Hyoban Lee, Yun-Seok Choi, Gyeongwon Kang, Min-Kyo Seo, Bong Hyun Chung,\* Yongwon Jung,\* and Bongsoo Kim\*



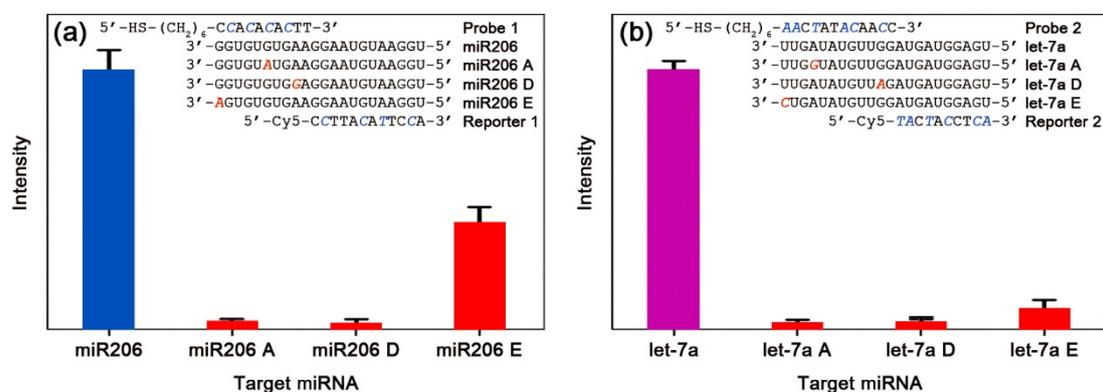
**Figure S1.** Plots of 1580 cm<sup>-1</sup> band intensity with target miRNAs and single base mismatched miRNAs in concentrations of 100 pM after single- and bi-temperature hybridization procedures. Data represent the mean plus standard deviation from ten measurements.



**Figure S2.** (a-d) SERS spectra of Cy5 measured from PNI sensors in the presence of perfectly matched miRNAs (miR206, let-7a, miR21, and miR96) and SERS spectra of Cy5 ( $\times 10$ ) measured from PNI sensors in the presence of single base mismatched miRNAs (miR206 A, B, let-7a A, B, miR21 A, B, and miR96 A, B). The concentrations were all 100 pM



**Figure S3.** (a, b) Plots of 1580 cm<sup>-1</sup> band intensity with target miRNAs (miR206 and let-7a), purine-purine mismatched miRNAs (miR206 A and let-7a A) and purine-pyrimidine mismatched miRNAs (miR206 C and let-7a C) in concentrations of 100 pM. The sequences of probe and reporter LNAs and target miRNAs are written in the inset of each figure. Data represent the mean plus standard deviation from ten measurements.

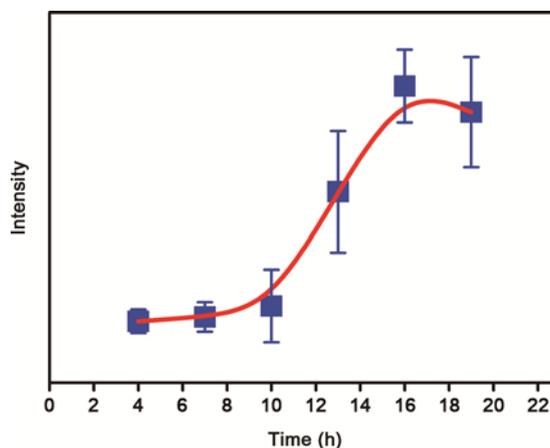


**Figure S4.** (a, b) Plots of 1580 cm<sup>-1</sup> band intensity with target miRNAs (miR206 and let-7a) and single base mismatched miRNAs (miR206 A, D, and E and let-7a A, D, and E) in concentrations of 100 pM. The sequences of probe and reporter LNAs and target miRNAs are written in the inset of each figure. Data represent the mean plus standard deviation from ten measurements.

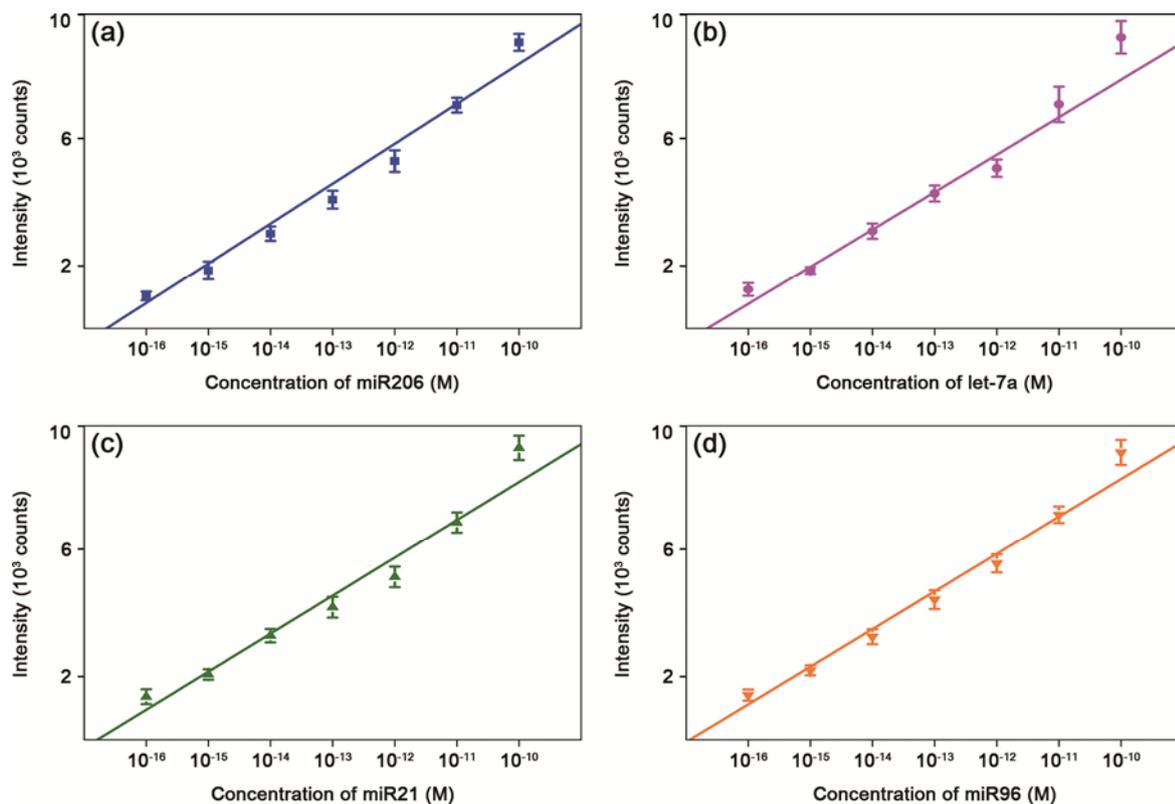
**Table S1.** Statistical analyses of Figure S4.

Target miRNA	Sequence (3' → 5')	Intensity	Intensity ratio (%) <sup>a</sup>
miR206	GGUGUGUGAAGGAAUGUAAGGU	9177.2	100
miR206 A	GGUGU <b>A</b> UGAAGGAAUGUAAGGU	295.0	3.21
miR206 D	GGUGUGUG <b>G</b> AGGAAUGUAAGGU	224.8	2.45
miR206 E	<b>A</b> GUGUGUGAAGGAAUGUAAGGU	3773.6	41.12
let-7a	UUGAUAUGUUGGAUGAUGGAGU	9134.4	100
let-7a A	UUG <b>G</b> UAUGUUGGAUGAUGGAGU	252.3	2.76
let-7a D	UUGAUAUGUU <b>A</b> GAUGAUGGAGU	278.2	3.05
let-7a E	<b>C</b> UGAUAUGUUGGAUGAUGGAGU	750.1	8.21

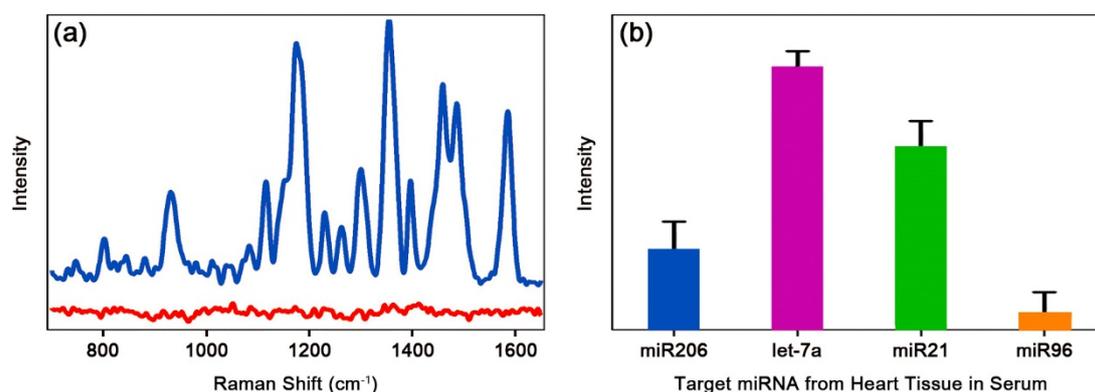
<sup>a</sup>These values are percentage intensity ratio of the SERS signal from single base mismatched miRNA to those from perfectly matched miRNA.



**Figure S5.** Plots of  $1580\text{ cm}^{-1}$  band intensities *versus* the hybridization time. The concentration of target miRNAs (miR206) is 1 fM. Data represent the mean plus standard deviation from ten measurements.



**Figure S6.** (a-d) Plot of 1580 cm<sup>-1</sup> band intensity as a function of the concentration of target miRNAs (miR206, let-7a, miR21, and miR96) and linearly fitted line. Data represent the mean plus standard deviation from ten measurements.



**Figure S7.** (a) SERS spectra of Cy5 measured from PNI sensors in the presence of target miRNA (miR206; blue spectrum) and absence (Red spectrum). Target miRNAs were dissolved in 10 % human serum and the concentration was 100 pM. (b) 1580 cm<sup>-1</sup> band intensities measured from each PNI sensor when the sample contains total RNA extracts from heart in 10 % human serum. Data represent the mean plus standard deviation from ten measurements.

**Table S2.** False positive concentration of single base mismatched miRNAs measured from PNI sensors employing bi-temperature hybridization.

Single base mismatched miRNA	False positive concentration <sup>a</sup>	Single base mismatched miRNA	False positive concentration
miR206 A	39 aM	miR21 A	27 aM
miR206 B	37 aM	miR21 B	25 aM
let-7a A	36 aM	miR96 A	60 aM
let-7a B	36 aM	miR96 B	74 aM

<sup>a</sup>False positive concentrations were obtained from the intensities of Figure 2 and linearly fitted lines of Figure S6.

**Table S3.** Concentration of miRNAs in total RNA extracts measured from PNI sensors employing bi-temperature hybridization.

Human tissue	Concentration of miRNAs <sup>a</sup>			
	miR206	let-7a	miR21	miR96
SKM	>100 pM	>100 pM	14 pM	62 aM
Heart	3.8 fM	>100 pM	15 pM	42 aM

<sup>a</sup>Concentrations of miRNAs were obtained from the intensities of Figure 4 and linearly fitted lines of Figure S6.