

## *Supplementary Information*

# **The Specific gravity-free method for Isolation of Circulating tumor KRAS mutant DNA and exosome in colorectal cancer**

*Tae Hee Lee<sup>†,\*1</sup>, Eunsook Park<sup>2</sup>, Young-gon Goh<sup>1</sup>, Han Byul Lee<sup>1</sup>, Woo Sun Rou<sup>3</sup>, Hyuk Soo*

*Eun<sup>†,\*4</sup>*

<sup>1</sup>Research Institute for Future Medical Science, Chungnam National University Sejong Hospital (CNUSH), Sejong, Republic of Korea

<sup>2</sup>Department of Chemistry, Korea Advanced Institute of Science and Technology (KAIST),  
291 Daehak-ro, Daejeon, Republic of Korea

<sup>3</sup>Division of Gastroenterology and Hepatology, Department of Internal Medicine, Chungnam National University Sejong Hospital, Sejong, Republic of Korea

<sup>4</sup>Division of Gastroenterology and Hepatology, Department of Internal Medicine, Chungnam National University Hospital, 282, Daejeon, Republic of Korea

<sup>†</sup>Both authors contributed equally to this work

<sup>\*</sup>Both authors contributed as corresponding authors

<sup>\*</sup>Address all correspondence to:

Tae Hee Lee, MT. PhD

Research Institute for Future Medical Science,

Chungnam National University Sejong Hospital (CNUSH),

Bodeum 7ro 20, Sejong, 30099, Republic of Korea

email: taehee1155@gmail.com

phone: (82)10-7112-8577

Hyuk Soo Eun, MD. PhD.

Division of Gastroenterology and Hepatology,

Department of Internal Medicine, Chungnam National University Hospital,

282, Daejeon, Republic of Korea

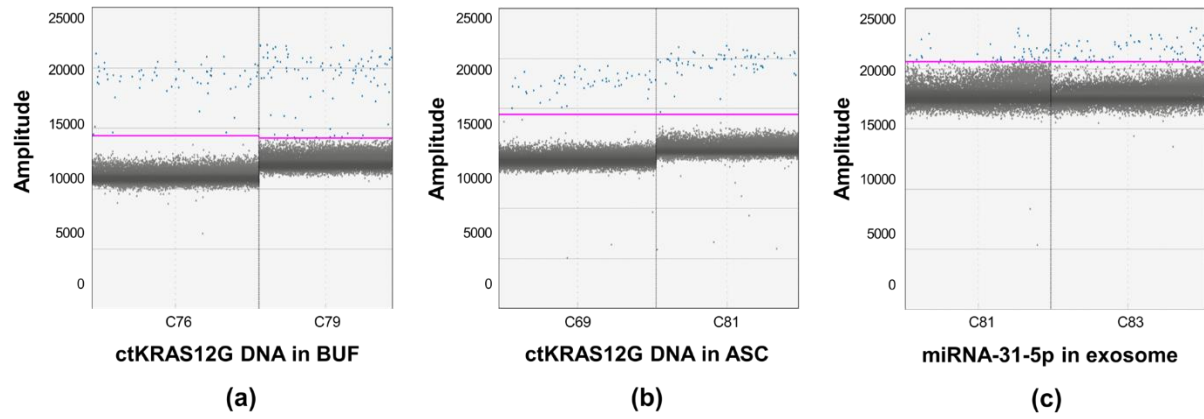
email: [liver@kaist.ac.kr](mailto:liver@kaist.ac.kr), [hyuksoo@cnuh.co.kr](mailto:hyuksoo@cnuh.co.kr)

phone: (82)10-3470-0767

## **Statistics**

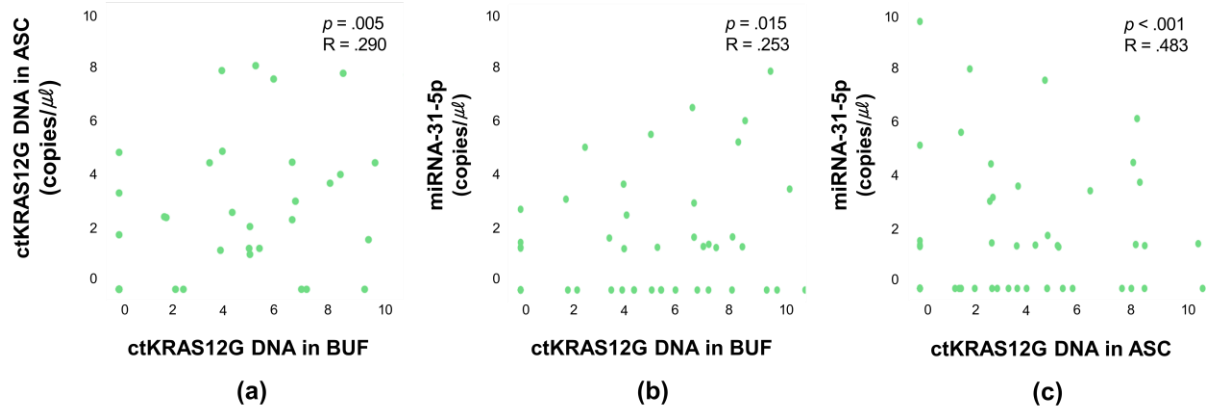
Various associations were determined by calculating Spearman correlation coefficient ( $r$ ) between ctDNA and exosome. A p-value  $<0.05$  was considered significant. All statistical analyses were made through SPSS Statistics 26 (SPSS, IL).

### Supplementary Figure.



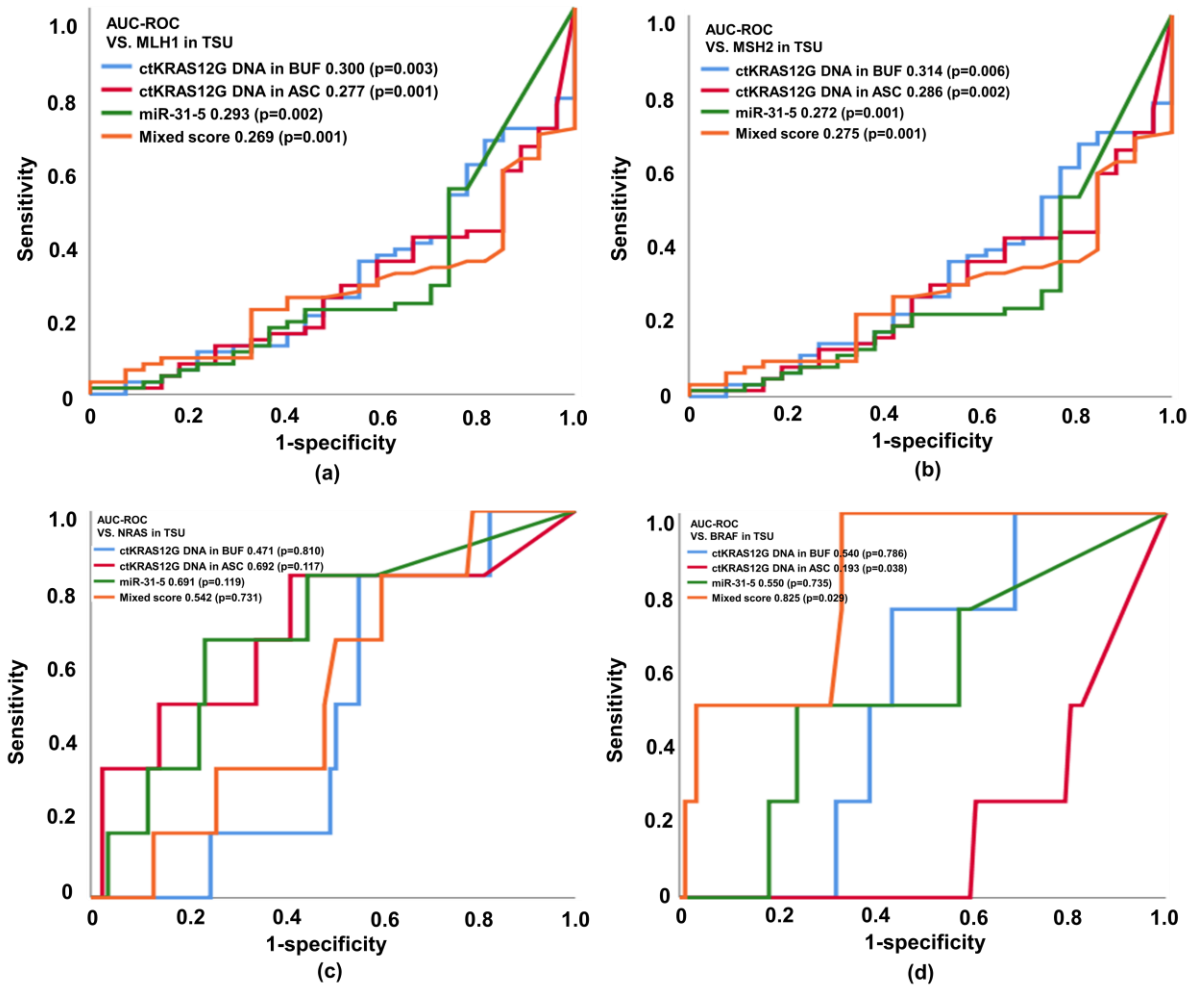
**Figure S1.** ddPCR analysis of (a) *KRAS* G12D mutation in buffy coat (blue: positive droplet), (b) *KRAS* G12D mutation in ascites (blue: positive droplet), and (c) miR-31-5 in exosome.

## Supplementary Figure.



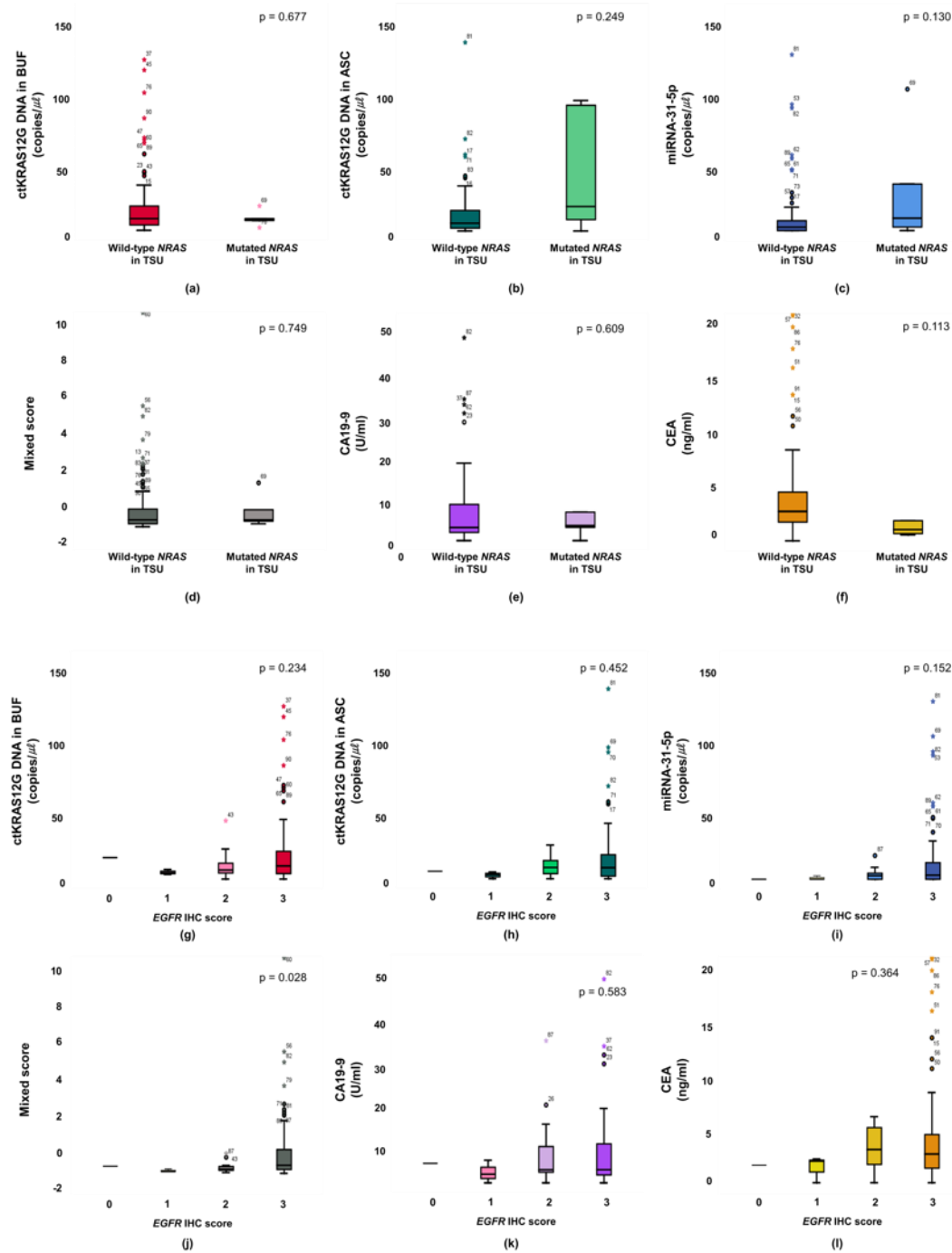
**Figure S2.** Spearman correlation analysis (a) between ascites and buffy coat of ctKRAS, (b) between ctKRAS in buffy coat and miR-31-5 in exosome, (c) between ctKRAS in ascites and miR-31-5 in exosome.

## Supplementary Figure.



**Figure S3.** ctKRAS G12D mutation in buffy coat, ctKRAS G12D mutation in ascites, miR-31-5 in exosome, and mixed score reflects the pathological status of the tumor burden based on MLH1 (a), MSH2 (b), NRAS (c), and BRAF (d).

## Supplementary Figure.



**Figure S4.** The expression levels in buffy coat (a), ascites (b), miR-31-5 (c), mixed score(d), CEA(e), and CA19-9(f) from mutant type and wild type of NRAS. The expression levels in buffy coat (g), ascites (h), miR-31-5 (i), mixed score(j), CEA(k), and CA19-9(l) from mutant type and wild type of EGFR.