

*Supplementary Information*

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

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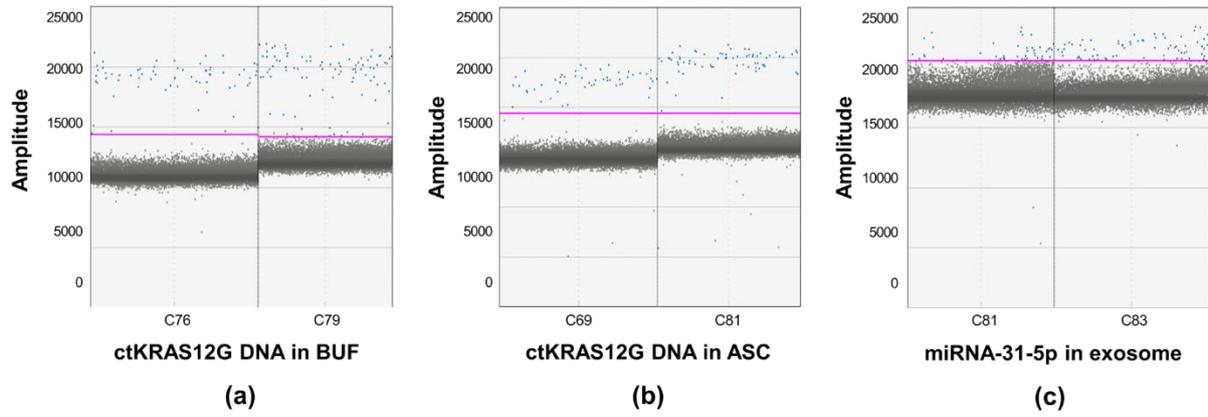
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## **Statistics**

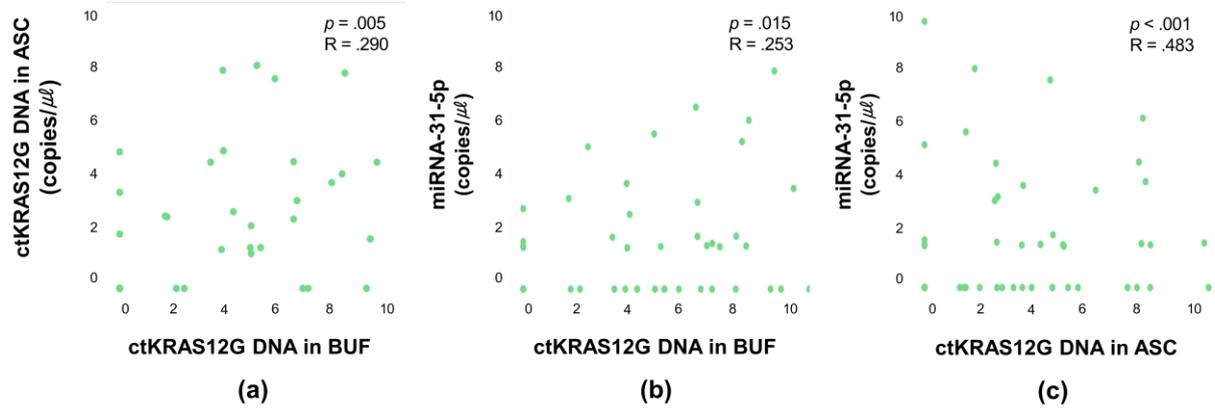
Various associations were determined by calculating Spearman correlation coefficient ( $r$ ) between ctDNA and exosome. A p-value  $<0.05$  was considered significant. All statistical analyses were made through SPSS Statistics 26 (SPSS, IL).

Supplementary Figure.



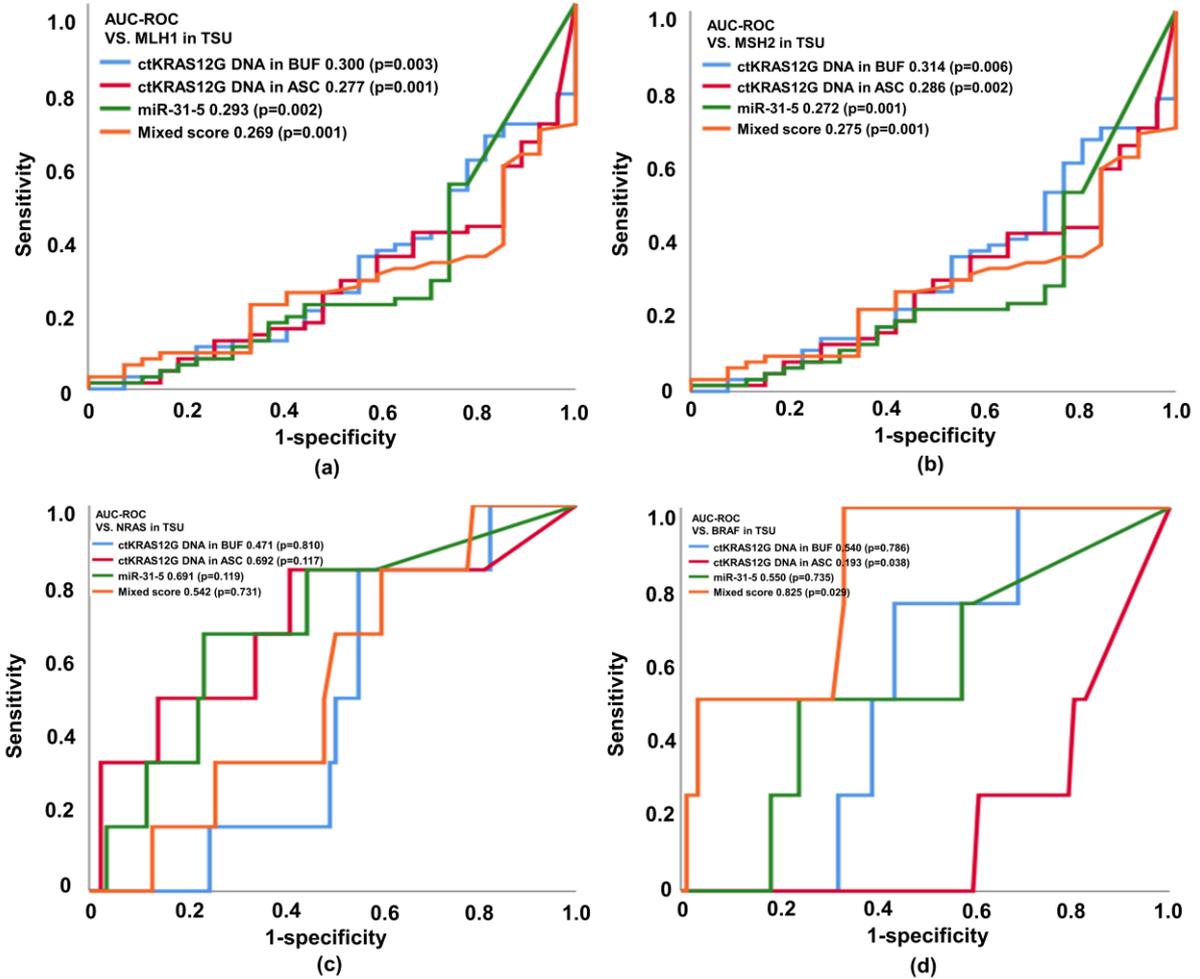
**Figure S1.** ddPCR analysis of (a) *KRAS* G12D mutation in buffy coat (blue: positive droplet), (b) *KRAS* G12D mutation in ascites (blue: positive droplet), and (c) miR-31-5 in exosome.

Supplementary Figure.



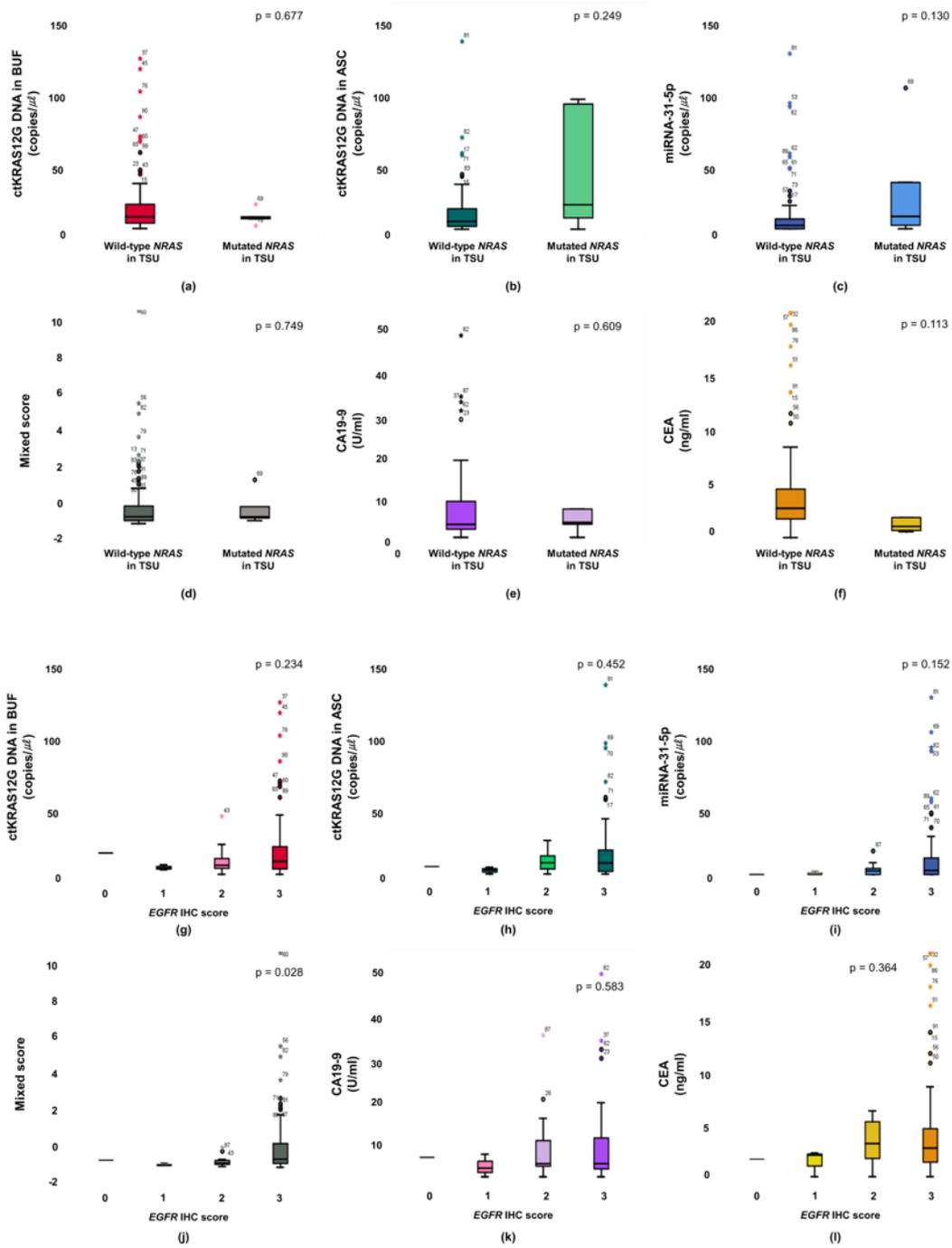
**Figure S2.** Spearman correlation analysis (a) between ascites and buffy coat of ctKRAS, (b) between ctKRAS in buffy coat and miR-31-5 in exosome, (c) between ctKRAS in ascites and miR-31-5 in exosome.

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.