Genetic Analysis of Biopsy Tissues from Colorectal Tumors in Patients with Ulcerative Colitis

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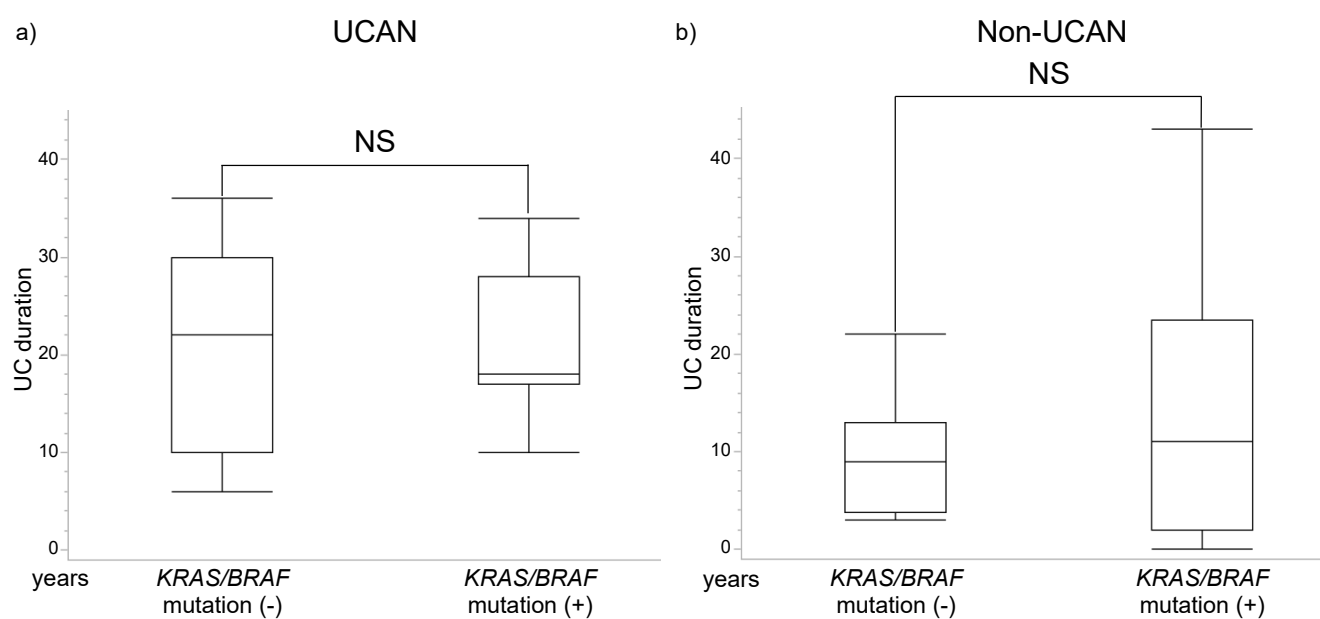* Correspondence: beyan13@hiroshima-u.ac.jp; Tel.: +81-82-257-5193

Figure S1: Correlation between *KRAS*/*BRAF* mutation status of biopsy specimens and UC duration in the 45 CRNUC cases in step 1 and step 2. The vertical axis indicates the duration of UC. a) Correlation between *KRAS*/*BRAF* mutation of the biopsy specimens and UC duration in UCANs. b) Correlation between *KRAS*/*BRAF* mutation of the biopsy specimens and UC duration in non-UCANs. CRNUC, colorectal neoplasia developing from ulcerative colitis mucosa; UCAN, ulcerative colitis-associated neoplasia; NS, not significant.
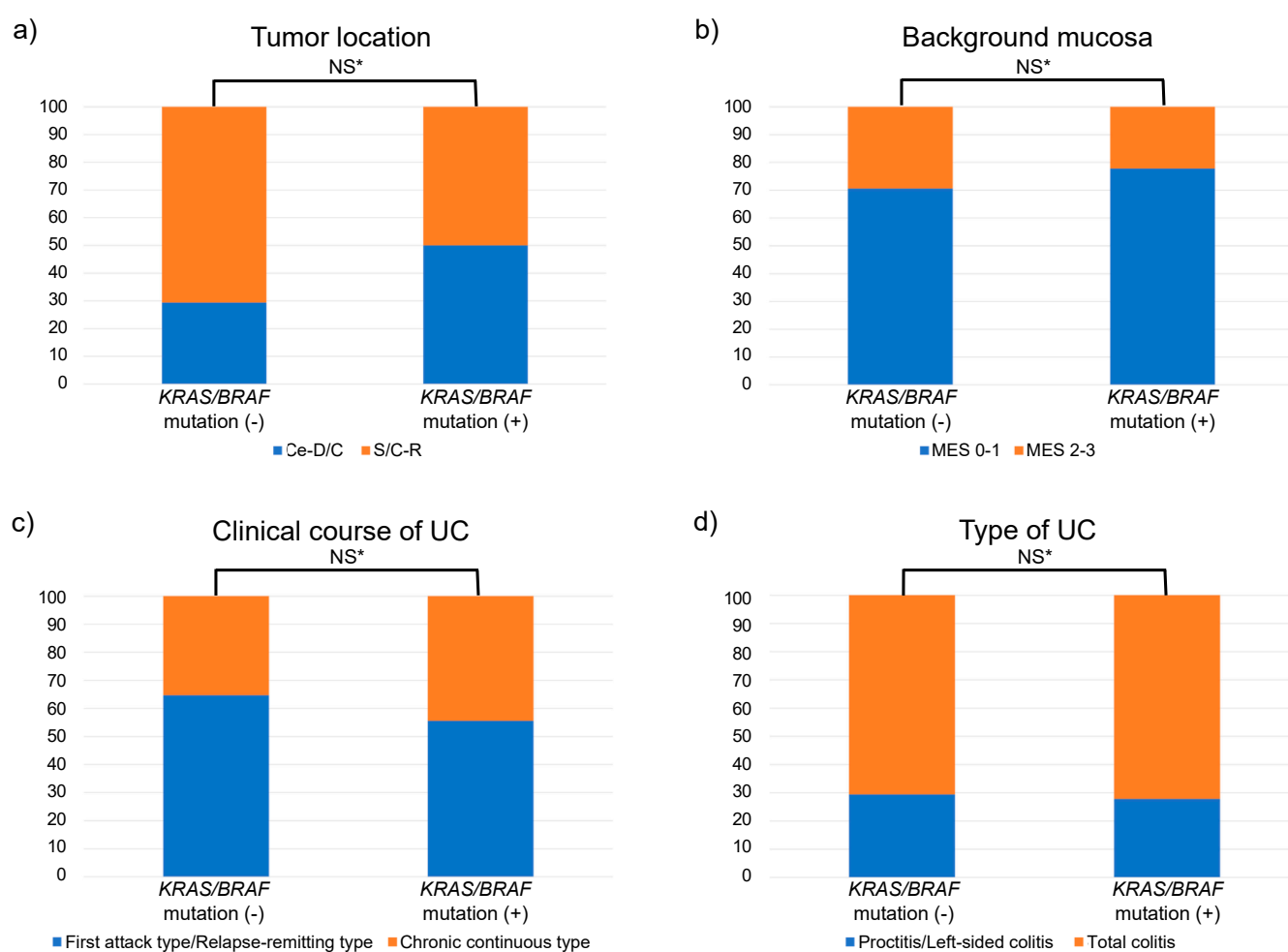
Figure S2: Correlation between *KRAS/BRAF* mutation status in biopsy specimens and the clinical features of 35 early CRNUCs. The bar graphs show the correlation between *KRAS/BRAF* mutation in biopsy specimens and a) tumor location (Ce–D/C versus [vs.] S/C–R), b) MES of background mucosa (MES0–1 vs. MES2–3), c) clinical course of UC (first attack type vs. chronic continuous type), and d) type of UC (proctitis/left-sided colitis vs. total colitis). CRNUC, colorectal neoplasia developing from ulcerative colitis mucosa; UC, ulcerative colitis; Ce, cecum; D/C, descending colon; S/C, sigmoid colon; R, rectum; MES, Mayo endoscopic subscore; NS, not significant. *Categorical variables were compared using Fisher's exact tests.
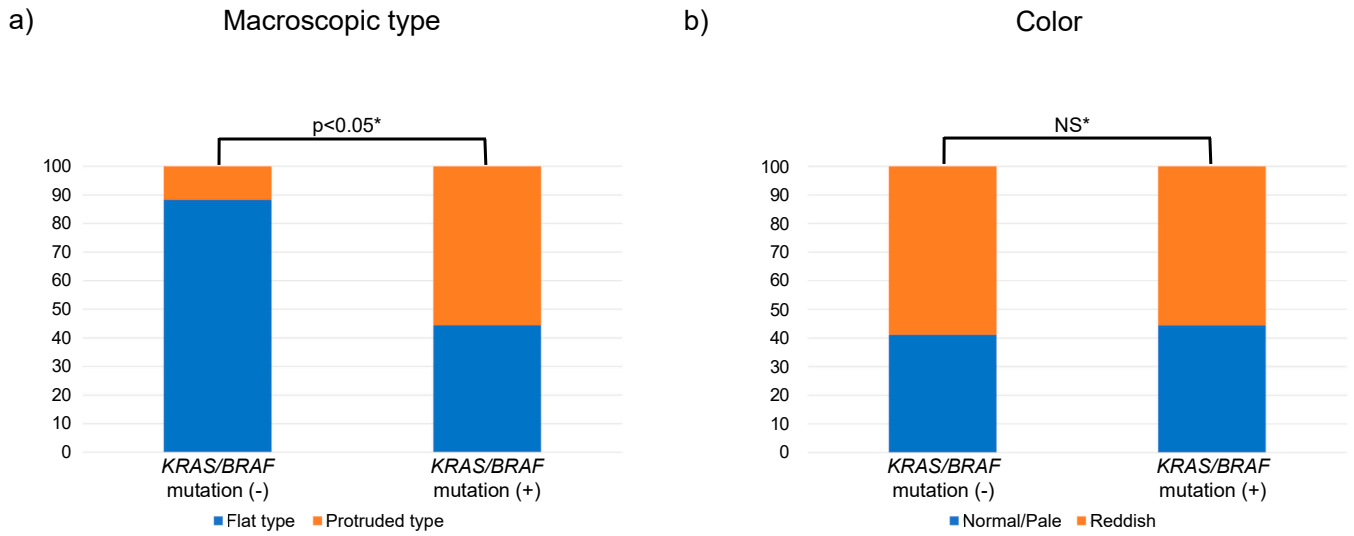
a) **Macroscopic type**   b) **Color**

Figure S3: Correlation between *KRAS/BRAF* mutation status in biopsy specimens and endoscopic findings of the 40 early CRNUCs. The bar graphs show the correlation between *KRAS/BRAF* mutation in biopsy specimens and a) macroscopic type of tumor (flat type versus [vs.] protruded type), and b) color of tumor (normal/pale vs. reddish). CRNUC, colorectal neoplasia developing from ulcerative colitis mucosa; NS, not significant. *Categorical variables were compared using Fisher's exact tests.

Table S1: Details of the 40 genes included in the multi-gene panel in this study.

APC	*TCF7L2*	*NRG1*
TP53	*NWD1*	*MAP2K3*
KRAS	*ACVR1B*	*TPO*
PRKDC	*BRAF*	*TGFBR2*
CSMD3	*RNF43*	*MDM2*
CREBBP	*SMAD2*	*PIK3CA*
KIF26B	*ZNF536*	*SOX9*
LTBP4	*IL16*	
EP300	*IL17RD*	
ATM	*FBXW7*	

The multi-gene panel used in this study includes the same set of genes as the panel used in previous research.

Table S2: Clinicopathological features of colorectal neoplasia in step 1 (n = 14).

Variables	(%)
Age (years old, median [range])	57 [45–85]
Sex	
Male	5 (36)
UC duration (years, median [range])	19 [4–43]
Type of UC	
Total colitis	11 (79)
Left-sided colitis	3 (21)
Clinical course of UC	
Relapse-remitting type	5 (36)
Chronic continuous type	9 (64)
Location	
Right-side colon	2 (14)
Left-side colon	6 (43)
Rectum	6 (43)
Background mucosa	
MES 0/1	8 (57)
MES 2/3	6 (43)
Resected method	
Endoscopic resection	12 (86)
Surgical resection	2 (14)
Histology	
High-grade dysplasia	8 (57)
Carcinoma	6 (43)

UC: ulcerative colitis, MES: mayo endoscopic subscore

Table S3: Clinicopathological features of colorectal neoplasia in step 2 (n = 26).

Variables	(%)
Age (years old, median [range])	61 [38–78]
Sex	
Male	17 (65)
UC duration (years, median [range])	16 [0–36]
Type of UC	
Total colitis	17 (65)
Left-sided colitis	6 (23)
Proctitis	3 (12)
Clinical course of UC	
Relapse-remitting type	19 (73)
Chronic continuous type	6 (23)
First attack type	1 (4)
Location	
Right-side colon	7 (27)
Left-side colon	10 (38)
Rectum	9 (35)
Background mucosa	
MES 0/1	20 (77)
MES 2/3	6 (23)
Resected method	
Endoscopic resection	14 (54)
Surgical resection	12 (46)
Histology	
Low-grade dysplasia	6 (23)
High-grade dysplasia	9 (35)
Carcinoma	8 (30)
Serrated lesion	3 (12)

UC: ulcerative colitis, MES: mayo endoscopic subscore

Table S4: Correlation between the pathological diagnosis of resected specimens and the pathological diagnosis of   biopsy specimens in 26 CRNUCs from step 2.

Pathological diagnosis of biopsy specimen [a]	Pathological diagnosis of resection specimen [b]		Total (%)
	UCAN (%)	Non-UCAN (%)	
UCAN	5 (100)	0 (0)	5 (100)
Non-UCAN	0 (0)	5 (100)	5 (100)
Unclassified	10 (63)	6 (37)	16 (100)

[a] The pathological diagnosis of biopsy specimens in this table refers to the pathological diagnosis by hematoxylin and eosin (HE) staining.

[b] The pathological diagnosis of resection specimens in this table refers to the final pathological diagnosis by HE staining with p53 and Ki67 immunostaining assessment.

CRNUC, colorectal neoplasia developing from ulcerative colitis mucosa; UCAN, ulcerative colitis-associated neoplasia.

Table S5: Correlation between the pathological diagnosis of resected specimens and p53 staining of biopsy specimens in the 26 CRNUCs in step 2.

p53 staining of biopsy specimen	Pathological diagnosis of resection specimen [a]		Total (%)	
	UCAN (%)	Non-UCAN (%)		
Strongly positive	8 (100)	0 (0)	8 (100)	
				$p<0.01$ [bc]
Weakly positive	4 (29)	10 (71)	14 (100)	
Negative	3 (75)	1 (25)	4 (100)	

[a] The pathological diagnosis of biopsy specimens and resection specimens in this table refers to the pathological diagnosis by hematoxylin and eosin staining with p53 and Ki67 immunostaining assessment.

[b] Categorical variables were compared using Fisher's exact tests.

[c] We compared a group with strongly positive p53 staining in biopsy specimens and a group with weakly positive/negative p53 staining.

CRNUC, colorectal neoplasia developing from ulcerative colitis mucosa; UCAN, ulcerative colitis-associated neoplasia.

Table S6: Correlation between the pathological diagnosis of resected specimens and the *TP53* mutation status of the biopsy specimens from the 26 CRNUCs in step 2.

TP53 mutation	Pathological diagnosis of resection specimen [a]		Total (%)	
	UCAN (%)	Non-UCAN (%)		
+	10 (91)	1 (9)	11 (100)	
				$p<0.01$ [b]
-	5 (33)	10 (67)	15 (100)	

[a] The pathological diagnosis of biopsy specimens and resection specimens in this table refers to the pathological diagnosis by hematoxylin and eosin staining with p53 and Ki67 immunostaining assessment.

[b] Categorical variables were compared using Fisher's exact tests.

CRNUC, colorectal neoplasia developing from ulcerative colitis mucosa; UCAN, ulcerative colitis-associated neoplasia.