

Supplementary Table S1. The determination of the binding epitope of C₄₄Mab-1 by ELISA.

Peptide	Coding exon*	Sequence	C ₄₄ Mab-1
CD44p21–40	2	QIDLNITCRFAGVFHVEKNG	–
CD44p31–50	2	AGVFHVEKNGRYSISRTEAA	–
CD44p41–60	2	RYSISRTEAADLCKAFNSTL	–
CD44p51–70	2	DLCKAFNSTLPTMAQMEKAL	–
CD44p61–80	2/3	PTMAQMEKALSIGFETCRYG	–
CD44p71–90	2/3	SIGFETCRYGFIEGHVVIPR	–
CD44p81–100	3	FIEGHVVIPRIHPNSICAAN	–
CD44p91–110	3	IHPNSICAANNTGVYILTSN	–
CD44p101–120	3	NTGVYILTSNTSQYDTYCFN	–
CD44p111–130	3/4	TSQYDTYCFNASAPPEEDCT	–
CD44p121–140	3/4	ASAPPEEDCTSVTDLPNAFD	–
CD44p131–150	4/5	SVTDLPNAFDGPITITIVNR	–
CD44p141–160	4/5	GPITITIVNRDGTTRYVQKGE	–
CD44p151–170	5	DGTTRYVQKGEYRTNPEDIYP	–
CD44p161–180	5	YRTNPEDIYPSNPTDDDVSS	–
CD44p171–190	5	SNPTDDDVSSGSSSERSSTS	–
CD44p181–200	5	GSSSERSSTSGGYIFYTFST	–
CD44p191–210	5	GGYIFYTFSTVHPIPEDDSP	–
CD44p201–220	5	VHPIPEDSPWITDSTDRIIP	–
CD44p211–230	5/v3	WITDSTDRIIPATSTSSNTIS	–
CD44p221–240	5/v3	ATSTSSNTISAGWEPNEENE	–
CD44p231–250	v3	AGWEPNEENEDERDRHLSFS	–
CD44p241–260	v3	DERDRHLSFSGSGIDDEDF	–
CD44p251–270	v3/v4	GSGIDDEDFISSTISTTPR	–
CD44p261–280	v3/v4	ISSTISTTPRAFDHTKQNQD	–
CD44p271–290	v4	AFDHTKQNQDWTQWNPSHSN	–
CD44p281–300	v4	WTQWNPSHSNPEVLLQTTR	–
CD44p291–310	v4/v5	PEVLLQTTRMTDVRNGTT	–
CD44p301–320	v4/v5	MTDVRNGTTAYEGNWNPEA	–
CD44p311–330	v5	AYEGNWNPEAHPPLIHHEHH	–
CD44p321–340	v5	HPPLIHHEHHHEEEETPHSTS	–
CD44p331–350	v5/v6	EEEETPHSTSTIQATPSSTT	–

CD44p341–360	v5/v6	TIQATPSSTTEETATQKEQW	–
CD44p351–370	v6	EETATQKEQWFGNRWHEGYR	–
CD44p361–380	v6	FGNRWHEGYRQTPREDSHST	–
CD44p371–390	v6/v7	QTPREDSHSTTGTAASAHT	–
CD44p381–400	v6/v7	TGTAASAHTSHPMQGRTP	–
CD44p391–410	v7	SHPMQGRTPSPEDSSWTF	–
CD44p401–420	v7	SPEDSSWTFDFNPISHPMGR	–
CD44p411–430	v7/v8	FNPIHPMGRGHQAGRMDM	–
CD44p421–440	v7/v8	GHQAGRMDMDSSHSTTLQP	–
CD44p431–450	v8	DSSHSTTLQPTANPNTGLVE	–
CD44p441–460	v8	TANPNTGLVEDLRTGPLSM	–
CD44p451–470	v8/v9	DLDRTGPLSMTTQQSNSQSF	–
CD44p461–480	v8/v9	TTQQSNSQSFSTSHEGLEED	–
CD44p471–490	v9	STSHEGLEEDKDHPTTSTLT	+
CD44p481–500	v9/v10	KDHPTTSTLTSSNRNDVTGG	–
CD44p491–510	v9/v10	SSNRNDVTGGRRDPNHSEGS	–
CD44p501–520	v10	RRDPNHSEGSTTLLEGYTS	–
CD44p511–530	v10	TTLLEGYTSHPHTKESRTF	–
CD44p521–540	v10	YPHTKESRTFIPVTSAGTGS	–
CD44p531–550	v10	IPVTSAGTGSFGVTAVTVGD	–
CD44p541–560	v10	FGVTAVTVGDSNSNVNRSLS	–
CD44p551–570	v10/16	SNSNVNRSLSGDQDTFHPSG	–
CD44p561–580	v10/16	GDQDTFHPSGGSHTTHGSES	–
CD44p571–590	16	GSHTTHGSESDGSHGSEQEG	–
CD44p581–600	16/17	DGSHGSEQEGGANTTSGPIR	–
CD44p591–606	17	GANTTSGPIRTPQIPEAAAA	–

+, OD655 $\geq$ 0.3; –, OD655<0.1

*The CD44 exon-coding regions are illustrated in Figure 1.

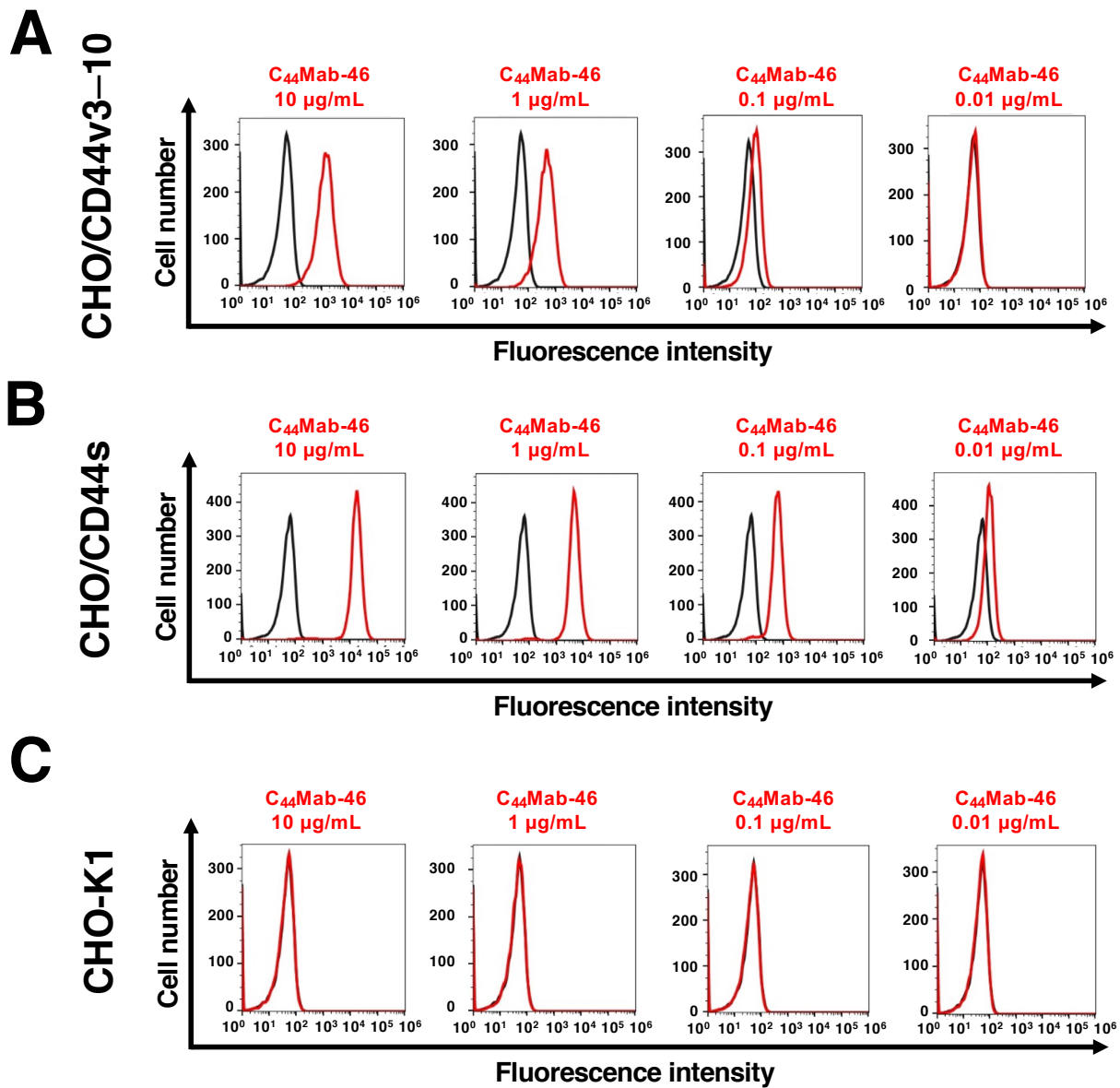
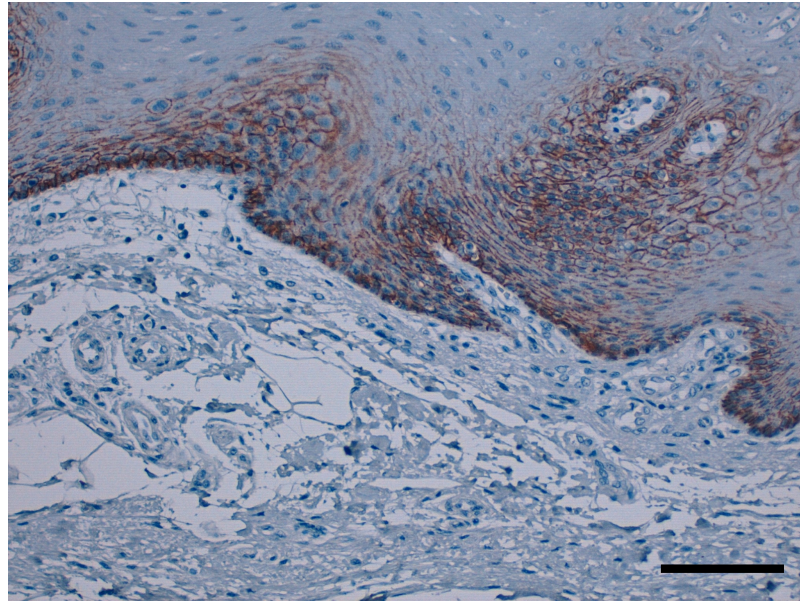
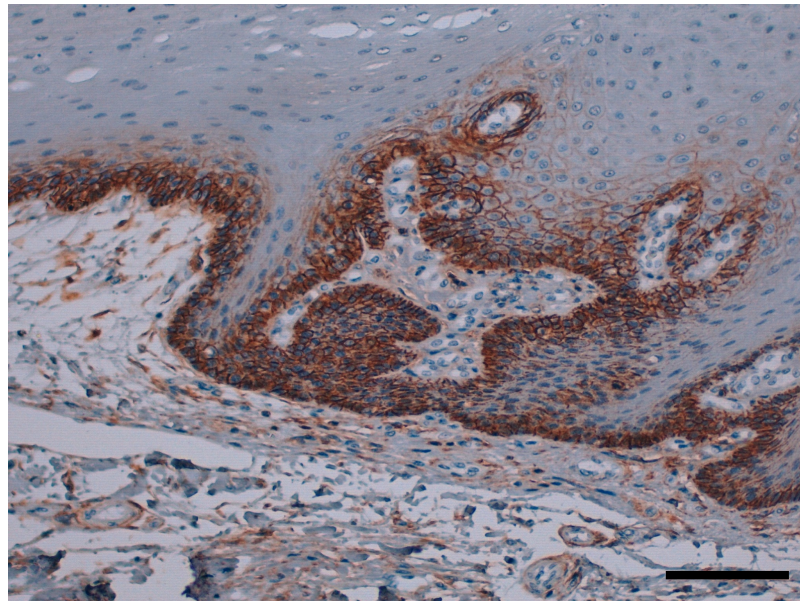


Figure S1 Conformation of the recognition of CHO/CD44s and CHO/CD44v3-10 by C₄₄Mab-46 by flow cytometry. CHO/CD44v3-10 (A), CHO/CD44s (B), and CHO-K1 (C) were treated with 0.01-10 $\mu\text{g/mL}$ of C₄₄Mab-46, followed by treatment with Alexa Fluor 488-conjugated anti-mouse IgG (Red line). The black line represents the negative control (blocking buffer).

C₄₄Mab-1



C₄₄Mab-46



Supplementary Figure S2 Immunohistochemical analysis using C₄₄Mab-1 and C₄₄Mab-46 against oral squamous cell carcinoma tissues. After antigen retrieval, the sections were incubated with 1 $\mu\text{g}/\text{mL}$ of C₄₄Mab-1 (A) and 1 $\mu\text{g}/\text{mL}$ of C₄₄Mab-46 (B), followed by treatment with the Envision+ kit. The color was developed using 3,3'-diaminobenzidine tetrahydrochloride (DAB), and the sections were counterstained with hematoxylin. Scale bar = 100 μm .