

Table 1 Clones, suppliers, dilutions and incubation time of the antibodies used in the canine mammary tissue sections

Antibody	Clone	Supplier	Dilutions	Incubation time
CD44 (Monoclonal)	MAC329	Abcam, ref. ab23844	1:10	1h
CD24 (Polyclonal)	-	Abcam, ref. ab202963	1:100	1h

Table 2 Correspondence between morphological diagnosis and histological type

Morphological diagnosis	Histological type
Tubular Tubulopapillary Tubulotrabeular Ductal	Tubular simple
Tubular complex Tubulopapillary complex Tubulotrabeular complex	Tubular complex
Adenosquamous Adenosquamous and tubular Comedocarcinoma Cribriform Solid and tubular/trabeular	Solid
Anaplastic	Anaplastic

Table 3 Histological malignancy grades and histological diagnosis in the cohort

Histological type	Histological malignancy grade				Total
	Grade 1	Grade 2	Grade 3	IMC	
Tubular simple	13	11	7	0	31
Tubular complex	14	1	2	0	17
Solid	3	7	21	3	34
Anaplastic	0	0	4	10	14
Total	30	19	34	13	96

Table 4 Expression of CD44, CD24, CD44⁺/CD24⁻ and CD44⁻/CD24⁺ in relation to the type of component

Type of component	Luminal (n=75)	Diffuse (n=68)	Myoepithelial (n=37)	p-value
Expression of CD44	60 (80.0) ^{M D}	40 (58.8) ^{L M}	12 (32.4) ^{L D}	< 0.001
Expression of CD24	39 (52.0) ^M	31 (45.6) ^M	2 (5.6) ^{LD}	< 0.001
Expression of CD44 ⁺ /CD24 ⁻	49 (65.3) ^M	31 (45.6)	12 (32.4) ^L	0.002
Expression of CD44 ⁻ /CD24 ⁺	25 (33.8) ^M	24 (35.3) ^M	1 (2.7) ^{LD}	0.023

Values are presented as frequency (percentage)

^L Group significantly different from Luminal; ^D Group significantly different from Diffuse; ^M Group significantly different from Myoepithelial

Table 5 Expression of markers and phenotypes in relation to clinicopathological parameters

		Expression of CD44	Expression of CD24	Expression of CD44 ⁺ /CD24 ⁻	Expression of CD44 ⁻ /CD24 ⁺
Grade	Grade 1 (n=30)	28 (93.3)	12 (40.0)	26 (86.7)*	9 (30.0)
	Grade 2 (n=19)	13 (68.4)	7 (36.8)	13 (68.4)	3 (15.8)
	Grade 3 (n=34)	24 (70.6)	23 (67.6)	18 (52.9)	19 (55.9)
	IMC (n=13) ^{REF}	10 (76.9)	10 (76.9)	6 (46.2)	8 (61.5)
	pvalue	0.071	0.019	0.011	0.008
Histological type	Tubular simple (n=31)	24 (77.4)	11 (35.5)*	20 (64.5)	9 (29.0)
	Tubular complex (n=17)	17 (100.0)*	7 (41.2)	16 (94.1)*	6 (35.3)
	Solid (n=34) ^{REF}	23 (67.6)	23 (67.6)	17 (50.0)	17 (50.0)
	Anaplastic (n=14)	11 (78.6)	11 (78.6)	10 (71.4)	7 (50.0)
	pvalue	0.040	0.010	0.014	0.30
Emboli	Presence (n=29)	23 (79.3)	21 (72.4)	15 (51.7)	19 (65.5)
	Absence (n=67)	52 (77.6)	31 (46.3)	48 (71.6)	20 (29.9)
	pvalue	0.85	0.018	0.059	0.001
Infiltration of LN	Presence (n=17)	12 (70.6)	11 (64.7)	9 (52.9)	9 (52.9)
	Absence (n=32)	26 (81.3)	12 (37.5)	24 (75.0)	7 (21.9)
	pvalue	0.48	0.069	0.12	0.027
Tumor recurrence	Presence (n=13)	12 (92.3)	6 (46.2)	7 (53.8)	5 (38.5)
	Absence (n=71)	52 (73.2)	40 (56.3)	46 (64.8)	29 (40.8)
	pvalue	0.18	0.50	0.54	0.87
Distant metastasis	Presence (n=24)	16 (66.7)	16 (66.7)	9 (37.5)	14 (58.3)
	Absence (n=60)	48 (80.0)	30 (50.0)	44 (73.3)	20 (33.3)
	pvalue	0.19	0.17	0.002	0.035

Value are presented as frequencies (percentage); ^{REF} Reference group for post-hoc comparisons; * Group significantly different from Reference group

Table 6 Staining score of CD44, CD24, CD44⁺/CD24⁻ and CD44⁻/CD24⁺ in relation to clinicopathological parameters

Positive tumors		Expression of CD44	Expression of CD24	Expression of CD44 ⁺ /CD24 ⁻	Expression of CD44 ⁻ /CD24 ⁺
Grade	Grade 1 (n=30)	43.5 (28.0 ; 56.5)	10.0 (7.0 ; 23.0)	25.5 (16.0 ; 28.0)	7.0 (3.0 ; 14.0)
	Grade 2 (n=19)	42.0 (9.0 ; 54.0)	7.0 (2.0 ; 18.0)	12.0 (6.0 ; 27.0)	1.0 (1.0 ; 5.0)
	Grade 3 (n=34)	18.0 (6.5 ; 54.0)	19.0 (8.0 ; 30.0)	13.0 (7.0 ; 21.0)	6.0 (4.0 ; 24.0)
	IMC (n=13) ^{REF}	51.5 (23.0 ; 60.0)	13.5 (9.0 ; 32.0)	24.0 (11.0 ; 30.0)	8.5 (4.0 ; 10.0)
	pvalue	0.19	NA (n<8)	NA (n<8)	NA (n<8)
Histological type	Tubular simple (n=31)	43.5 (11.0 ; 55.0)	22.0 (9.0 ; 27.0)	25.0 (10.0 ; 27.0)	7.0 (5.0 ; 9.0)
	Tubular complex (n=17)	54.0 (40.0 ; 60.0)*	17.0 (9.0 ; 28.0)	27.0 (18.0 ; 29.5)*	5.0 (2.0 ; 18.0)
	Solid (n=34) ^{REF}	15.0 (7.0 ; 43.0)	11.0 (6.0 ; 24.0)	9.0 (6.0 ; 17.0)	6.0 (5.0 ; 12.0)
	Anaplastic (n=14)	58.0 (23.0 ; 63.0)*	15.0 (9.0 ; 30.0)	20.0 (11.0 ; 29.0)	8.0 (2.0 ; 17.0)
	pvalue	0.002	NA (n<8)	0.005	NA (n<8)
Emboli	Presence (n=29)	23.0 (8.0 ; 54.0)	23.0 (9.0 ; 32.0)	12.0 (5.0 ; 27.0)	9.0 (6.0 ; 24.0)
	Absence (n=67)	45.5 (16.5 ; 56.0)	11.0 (6.0 ; 22.0)	22.5 (10.0 ; 27.0)	5.0 (2.0 ; 8.0)
	pvalue	0.20	0.019	0.25	0.021
Tumor recurrence	Presence (n=12)	24.5 (5.0 ; 49.0)	8.0 (7.0 ; 13.0)	24.0 (16.0 ; 27.0)	6.0 (6.0 ; 6.0)
	Absence (n=52)	41.0 (9.5 ; 56.0)	17.0 (7.5 ; 25.5)	16.5 (6.0 ; 28.0)	7.0 (3.0 ; 18.0)
	pvalue	0.19	NA (n<8)	NA (n<8)	NA (n<8)
Distant metastasis	Presence (n=16)	45.0 (9.0 ; 60.5)	16.5 (8.5 ; 30.0)	24.0 (15.0 ; 27.0)	6.0 (2.0 ; 17.0)
	Absence (n=48)	35.5 (8.5 ; 54.0)	13.0 (7.0 ; 24.0)	16.5 (7.5 ; 26.5)	7.0 (4.5 ; 16.0)
	pvalue	0.50	0.41	0.35	0.56

Value are presented as median (Lower Quartile; Upper Quartile); ^{REF} Reference group for post-hoc comparisons; NA : Non Applicable ; * Group significantly different from Reference group