

Supplementary Figure 2. Chromosomal preparation from cell culture of female dog mammary gland carcinoma (A). Graphic representation of chromosomal alterations in different cultures of canine mammary gland cancer and its metastases (B).

Table 1: Mammary gland tumors information used to obtain neoplastic cells cultured *in vitro.*

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| **Identification** | **Histologic Classification** | **Graduation** | **Acquisition technique** | **Collagenase/explant time** | **Actual culture passage** |
|
| CM1 | Solid Carcinoma | Grade II | Collagenase | 4 hours | P10 |
| MM1 | Solid Carcinoma | - | Collagenase | 4 hours | P10 |
| CM2 | Comedocarcinoma | Grade II | Collagenase | Overnight | - |
| CM3 | Comedocarcinoma | Grade II | Collagenase | Overnight | - |
| CM4 | Tubulopapillary | Grade II | Collagenase | 4 hours | P10 |
| CM5 | Tubulopapillary | Grade II | Collagenase | 4 hours | P10 |
| CM6 | Carcinoma–mixed type | Grade I | Explant | 15 days | - |
| CM7 | Carcinoma–mixed type | Grade II | Explant | 7 dayss | - |
| CM8 | Comedocarcinoma | Grade II | Collagenase | 4 hours | - |
| CM9 | Tubulopapillary | Grade II | Collagenase | 4 hours | P10 |
| CM10 | Carcinoma–mixed type | Grade I | Collagenase | 4 hours | - |
| CM11 | Tubulopapillary | Grade I | Collagenase | 3 hours | P10 |
| CM12 | Carcinoma–mixed type | Grade II | Collagenase | 4 hours | - |
| CM13 | Carcinoma–mixed type | Grade II | Collagenase | 4 hours | - |
| CM14 | Carcinoma–mixed type | Grade I | Collagenase | 4 hours | - |
| CM15 | Carcinoma–mixed type | Grade II | Collagenase | 4 hours | - |
| CM60 | Adenosquamous carcinoma | Grade II | Collagenase IV | 3 hours | P10 |
| CM61 | Comedocarcinoma | Grade III | Collagenase IV | 3 hours | P10 |
| MM3 | CM60 Metastasis | - | Collagenase IV | 3 hours | P10 |
| MM4 | CM61 Metastasis | - | Collagenase IV | 3 hours | P10 |
| Neoplasms were classified following Goldschmidt et al., 2011. |

Table 2. Primary antibodies used in immunochemistry to characterize the molecular phenotype from mammary gland tumors.

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| **Antibody** | **Manufacture** | **Clone** | **Dilution** | **Imunolocalization** |
| P63 | Dako, Agilent Technologies, Santa Clara, CA, USA | 4A4 | 1:100 | Nuclei |
| HER2 | Roche Diagnostics, Risch-Rotkreuz, Suíça | 4B5 | 1:400 | Membrane |
| ERα | Santa Cruz Biotechnology®, Santa Cruz, CA, USA | C311 | 1:50 | Nuclei |
| PR | Roche Diagnostics, Risch-Rotkreuz, Suíça | 1E2 | Prediluted | Nuclei |
| Ki-67 | Dako, Agilent Technologies, Santa Clara, CA, USA | MIB1 | 1:50 | Nuclei |
| CK5/6 | Dako, Agilent Technologies, Santa Clara, CA, USA | D5/16B4 | 1:10 | Citoplasm |
| EGFR | Invitrogen, Thermo Fisher Scientific Corporation, Carlsbad, CA, USA | 31G7 | 1:20 | Citoplasm |

Table 3. Primary antibodies used in cell immunofluorescence to characterize the cell clone (cell origin) that was expanded in each culture.

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| **Antibody** | **Manufacture** | **Clone** | **Dilution** | **Imunolocalization** |
| Pan-citoqueratin | Invitrogen, Thermo Fisher Scientific Corporation, Carlsbad, CA, USA | AE1/AE3 | 1:300 | Citoplasm |
| Vimentin | Invitrogen, Thermo Fisher Scientific Corporation, Carlsbad, CA, USA | V9 | 1:300 | Citoplasm |
| CK8/18 | Novocastra, Vision BioSystems Ltd, Newcastle, UK, Europe | 5D3 | 1:600 | Citoplasm |