# Supplementary Materials

**Material S1. Methods**

In our study four molecular biomarkers were used in patients with invasive breast carcinoma (IBC): estrogen receptors (ER), progesterone receptors (PR), HER2 and Ki-67.

Antibodies for ER , PR, Ki-67 (Daco Omnis company, according protocol Dako company, nuclear reaction) were used. For HER2 antibodies ( Ventana company, staining on Benchmark machine, membrane reaction) were used. ER, PR were considered positive if tumors have 1% or more than nuclear-stained cells. HER2 staining was scored on a scale 0 to 3+according to the guidelines. The cut off for Ki-67 ≥14%was considered as high proliferation index (range from 0-to 100).

ER-is activated by the estrogen hormone, present in normal and IBC cells. Estrogen hormone stimulates the growth of normal breast epithelial cells and in IBC. ER are measured in BC tissue in our study using immunohistochemistry methods. It is performed on routinely prepared histological samples, primarily formalin-fixed paraffin-embedded (FFPE) tissue sections. Then slides are assessed microscopically to determine the intensity and proportions of positive cells. ER positive tumors are when ≥1%positive -staining tumor cells.

PR-is also routinely assessed similar to ER by immunohistochemistry methods, and is inactivated by progesterone hormone and stimulate the growth of tumor cells. PR positive tumors are positive, when ≥1% staining tumor cells is observed. The phenotype ER/PR positive is most frequent type of BC [1]

Expression of ER and PR determines the responsiveness of tumors to hormone therapy.

HER2 is a growth factor receptor, which is associated with cell proliferation, development and survival .The over-expression of HER2 status indicates a poorer prognosis. Today number of HER2-targeted therapies are available improve the overall survival. Is located on the surface at low levels in normal breast epithelium [2].

**Table S1.** Inferential results of univariate logistic regression explaining individual molecular cancer subtypes with US features.

| | **LA** | **LB HER2−** | **LB HER2+** | **HER2+** | **TNBC** |
|---|---|---|---|---|---|
| Shape | | | | | |
| Regular | OR = 4.387<br>*p* = 0.2345<br>CI: [0.4057–96.5053] | NA * | NA * | OR = 2.8<br>*p* = 0.4125<br>CI: [0.1255–31.0646] | NA * |
| Irregular | Ref | Ref | Ref | Ref | Ref |
| Orientation | | | | | |
| Parallel | OR = 0.35<br>*p* = 0.0672<br>CI: [0.1123–1.0816] | OR = 3.155<br>*p* = 0.2836<br>CI: [0.5677–59.2076] | OR = 3.155<br>*p* = 0.2836<br>CI: [0.5677–59.2076] | OR = 2.917<br>*p* = 0.3186<br>CI: [0.5217–54.833] | OR = 0.573<br>*p* = 0.3909<br>CI: [0.1688–2.283] |
| Non-parallel | Ref | Ref | Ref | Ref | Ref |
| Margin | | | | | |
| Circumscribed | NA * | NA * | NA * | NA * | NA * |
| Indistinct | Ref | Ref | Ref | Ref | Ref |
| Angular/ spiculated | OR = 0.977<br>*p* = 0.9615<br>CI: [0.3806–2.6144] | OR = 1.466<br>*p* = 0.5404<br>CI: [0.4603–5.64] | OR = 1.193<br>*p* = 0.7807<br>CI: [0.3644–4.6599] | OR = 2.315<br>*p* = 0.301<br>CI: [0.557–15.791] | OR = 0.389<br>*p* = 0.088<br>CI: [0.1298–1.1689] |
| Micro/macrolobulated | OR = 1.000<br>*p* = 1<br>CI: [0.1785–4.7934] | NA * | OR = 1.643<br>*p* = 0.6079<br>CI: [0.1978–10.4917] | OR = 3.571<br>*p* = 0.2418<br>CI: [0.3746–34.5553] | OR = 0.679<br>*p* = 0.6686<br>CI: [0.0881–3.6026] |
| Echo pattern | | | | | |
| Complex/hypoechoic | Ref | Ref | Ref | Ref | Ref |
| Hyper/isoechoic | OR = 2.241<br>*p* = 0.2764<br>CI: [0.4992–10.0823] | OR = 0.696<br>*p* = 0.7431<br>CI: [0.036–4.312] | OR = 0.696<br>*p* = 0.7431<br>CI: [0.036–4.312] | OR = 1.905<br>*p* = 0.457<br>CI: [0.2613–9.2842] | NA * |
| Posterior features | | | | | |
| No Posterior Features | Ref | Ref | Ref | Ref | Ref |
| Enhancement | NA * | OR = 0.323<br>*p* = 0.3262<br>CI: [0.0159–2.3083] | OR = 2.091<br>*p* = 0.4099<br>CI: [0.3393–12.9903] | OR = 5.750<br>*p* = 0.0324<br>CI: [1.2257–32.8005] | OR = 4.800<br>*p* = 0.0967<br>CI: [0.8049–38.8888] |
| Shadowing | OR = 0.368<br>*p* = 0.0911<br>CI: [0.1108–1.1472 | OR = 0.764<br>*p* = 0.7144<br>CI: [0.1687–3.2667] | OR = 3.407<br>*p* = 0.1006<br>CI: [0.8497–17.3023] | OR = 0.639<br>*p* = 0.6402<br>CI: [0.0786–4.1934] | OR = 2.857<br>*p* = 0.2373<br>CI: [0.5519–21.4375] |
| Combined Pattern | OR = 0.565<br>*p* = 0.2759<br>CI: [0.1994–1.5725] | OR = 1.014<br>*p* = 0.9832<br>CI: [0.284–3.8394] | OR = 0.697<br>*p* = 0.6748<br>CI: [0.1196–4.0566] | OR = 1.237<br>*p* = 0.7856<br>CI: [0.2747–6.5233] | OR = 3.429<br>*p* = 0.1415<br>CI: [0.7685–24.1953] |
| Calcifications | | | | | |
| Present | OR = 0.444<br>*p* = 0.0604<br>CI: [0.1866–1.0261] | OR = 2.457<br>*p* = 0.1178<br>CI: [0.8332–8.2791] | OR = 3.488<br>*p* = 0.0411<br>CI: [1.1305–13.1713] | OR = 3.143<br>*p* = 0.0632<br>CI: [1.0054–11.9458] | OR = 0.176<br>*p* = 0.0041<br>CI: [0.0469–0.5335] |
| Absent | Ref | Ref | Ref | Ref | Ref |

| | | | | | |
|---|---|---|---|---|---|
| Additional features | | | | | |
| Skin changes | OR = 0.422<br>*p* = 0.1518<br>CI: [0.1132–1.2689] | OR = 2.545<br>*p* = 0.1083<br>CI: [0.776–7.853] | OR = 2.545<br>*p* = 0.1083<br>CI: [0.776–7.853] | OR = 0.504<br>*p* = 0.391<br>CI: [0.0749–2.0193] | OR = 0.677<br>*p* = 0.5681<br>CI: [0.1461–2.3215] |
| Edema | OR = 1.422<br>*p* = 0.4492<br>CI: [0.583–3.6727] | OR = 0.63<br>*p* = 0.3968<br>CI: [0.2171–1.9027] | OR = 1.179<br>*p* = 0.7766<br>CI: [0.3944–4.0026] | OR = 0.557<br>*p* = 0.2928<br>CI: [0.1872–1.7084] | OR = 1.425<br>*p* = 0.5343<br>CI: [0.4893–4.7721] |
| Vascularity | OR = 0.255<br>*p* = 0.0734<br>CI: [0.0494–1.1078] | NA * | OR = 1.436<br>*p* = 0.7431<br>CI: [0.2319–27.8049] | OR = 1.329<br>*p* = 0.7969<br>CI: [0.2135–25.7841] | OR = 1.658<br>*p* = 0.646<br>CI: [0.2701–32.007] |
| Elastography | | | | | |
| Soft | Ref | Ref | Ref | Ref | Ref |
| Intermediate | OR = 0.522<br>*p* = 0.3111<br>CI: [0.1448–1.8523] | OR = 2.167<br>*p* = 0.4994<br>CI: [0.3077–43.6893] | OR = 2.690<br>*p* = 0.3815<br>CI: [0.4013–53.5133] | OR = 0.222<br>*p* = 0.1237<br>CI: [0.0265–1.5039] | OR = 2.400<br>*p* = 0.3033<br>CI: [0.5239–17.2288] |
| Hard | OR = 0.359<br>*p* = 0.0977<br>CI: [0.1043–1.2188] | OR = 3.405<br>*p* = 0.2617<br>CI: [0.5758–65.1953] | OR = 3.023<br>*p* = 0.3126<br>CI: [0.5047–58.1122] | OR = 0.960<br>*p* = 0.956<br>CI: [0.2459–4.7758] | OR = 0.913<br>*p* = 0.9162<br>CI: [0.1897–6.6557] |

LA= luminal A; LB HER2- = luminal B without HER2 overexpression; LB HER2+ = luminal B with HER2 overexpression; HER2+ = human epidermal growth factor receptor 2 positive; TNBC = triple negative breast cancer. OR=Odds ratio, CI= Confidence interval, Ref=reference, NA*= not applicable

**Supplementary References:**


1. WHO Classification of tumours, 5th edition Breast tumours. Invasive breast carcinoma. *Gen. Overv. IARC Fr.* **2019**,*2*, 82–101.
2. O Nielsen, T.; Leung, S.C.Y.; Rimm, D.L.; Dodson, A.; Acs, B.; Badve, S.; Denkert, C.; Ellis, M.J.; Fineberg, S.; Flowers, M.; et al. Assessment of Ki67 in Breast Cancer: Updated Recommendations From the International Ki67 in Breast Cancer Working Group. *J. Natl. Cancer Inst.* **2021**, *113*, 808–819, doi:10.1093/jnci/djaa201.