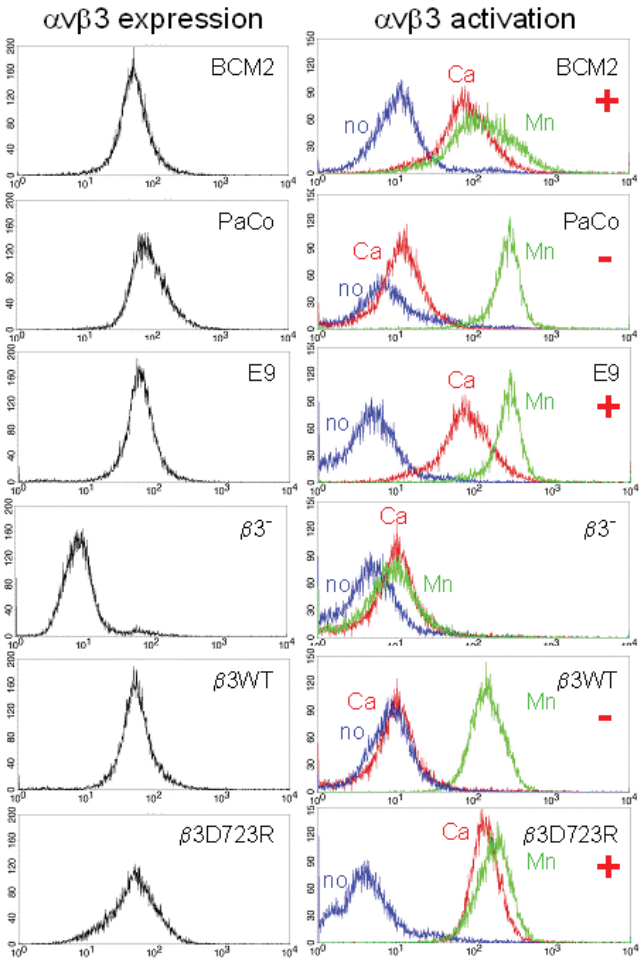
Supplementary Information

Supplemental Figure S1.

Tumor cell model for integrin αvβ3 activation

Flow cytometry analysis of tumor cell variants expressing defined levels and affinity states of integrin αvβ3. The cell variants were derived from MDA-MB 435 human breast cancer cells. Left panels: Overall integrin αvβ3 heterodimer expression (Mab VNR1, does not distinguish between high and low affinity αvβ3). Right panels: integrin αvβ3 activation state measured by binding of soluble RGD-ligand mimetic scFv antibody Bc-15 which behaves like a natural ligand and is specific for αvβ3. Lack of scFv binding in the absence of divalent cations (blue lines = no cations) indicates cation dependence. Intrinsic activation of αvβ3 (high affinity) allows scFv binding in the presence of calcium (1mM) (red line = Ca^{2+}), while low or no binding in calcium indicates low affinity αvβ3 in cells that express the receptor. ScFv binding in the presence manganese (50 μM) (green line = Mn^{2+}) is maximized, regardless of the intrinsic αvβ3 activation state, as long as the cells are αvβ3 positive. Bold red +/- signs on the right indicate the αvβ3 activation state.

Figure S1


αvβ3 expression
αvβ3 activation
BCM2
PaCo
E9
β3-
β3WT
β3D723R
Ca
Mn
no
+
-
+
-
+
+