

Figure S1

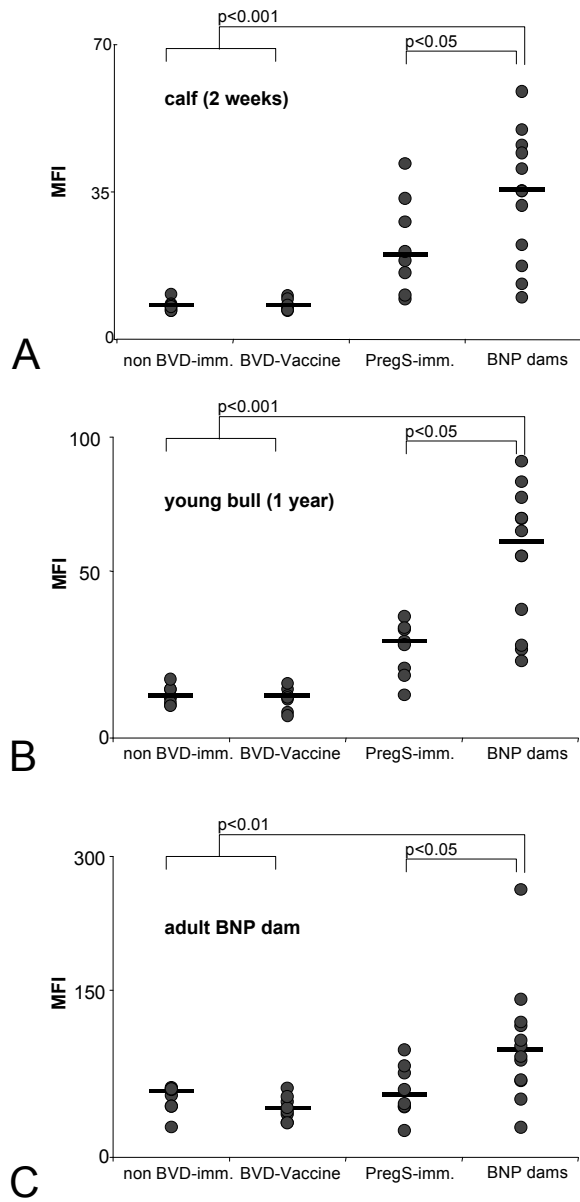


Fig S1 Age related differences have no influence on alloantibody recognition: Leukocytes were isolated from whole blood of three different donors. A panel of eight non-BVD-immunized control sera (non-BVD-imm.), eight sera from alternatively BVD-vaccinated cows (BVD-vaccine), eight sera from PregSure immunized non-BNP dams (PregS-imm.) and from twelve PregSure immunized BNP dams (BNP dams) was tested by flow cytometry. Dots represent the MFI within the lymphocyte gate for individual sera. Black bars indicate the median value for each group. P-values were calculated by simple Student's T-Test. Leukocytes were obtained from a calf of two weeks age (A), from a one-year-old bull (B) and from an adult BNP dam (C).

Figure S2

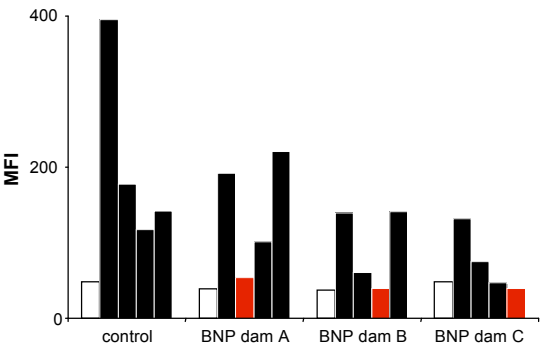


Fig S2 BNP associated alloantibodies do not bind autologous leukocytes: A panel of one control and four BNP dam sera was tested against PHA blasts obtained from one control donor and three BNP dams. Sera are always shown in the same order. Columns indicate the MFI with control (white) or BNP dam sera (black). Red columns represent the autologous combination, i.e. when the serum of a BNP dam is tested on autologous PHA blasts.

Figure S3

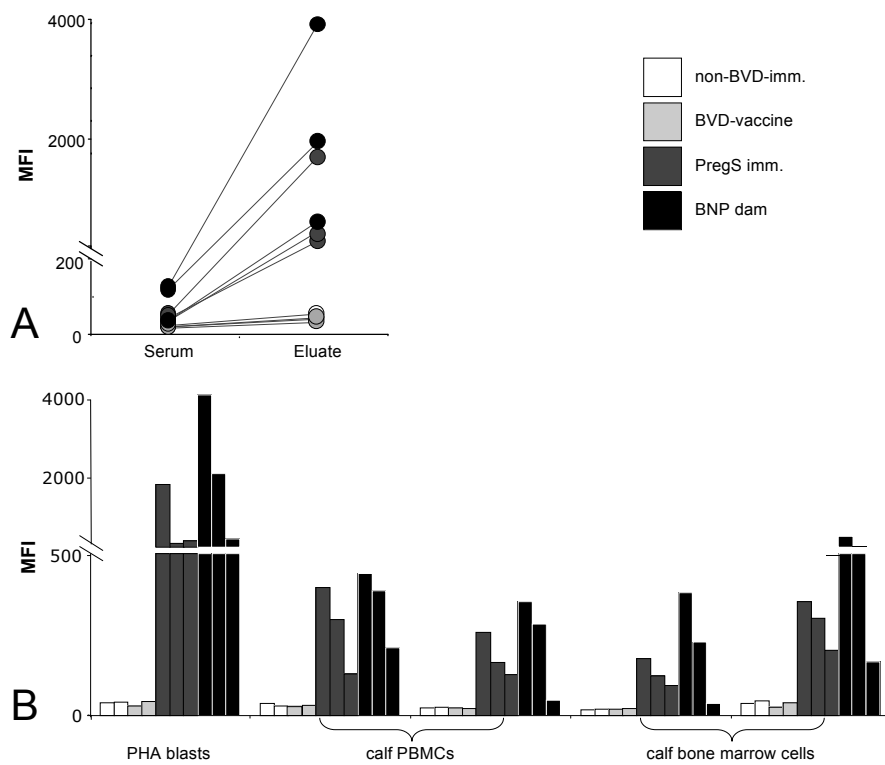


Fig S3 Affinity purified alloantibodies recognize calf PBMCs and bone marrow cells: Alloreactive antibodies were affinity-purified by desorption from the cell surface of the bovine kidney cell line used for vaccine production. (A) Affinity purified alloantibodies and the corresponding sera were analyzed for alloreactive binding to bovine PHA blasts obtained from a one-year-old heifer. Black circles represent three pairs of BNP dam sera and affinity eluates (BNP dam), dark grey circles three PregSure immunized Non-BNP dams (PregS-imm.), light grey alternatively BVD-immunized (BVD-vaccine) and white non-BVD-immunized controls (non-BVD-imm.). Circles show individual MFI values, the grey line indicates corresponding pairs. (B) The same panel of affinity purified antibodies was tested against PHA blasts, PBMCs from two different calves of the age of three and two weeks and bone marrow cells from two calves at the age of sixteen and eighteen days. Columns represent the MFI obtained with individual affinity eluates. The same color code as in (A) is used.