

Supplemental information

**Vaccine-induced spike- and nucleocapsid-specific
cellular responses maintain potent cross-reactivity
to SARS-CoV-2 Delta and Omicron variants**

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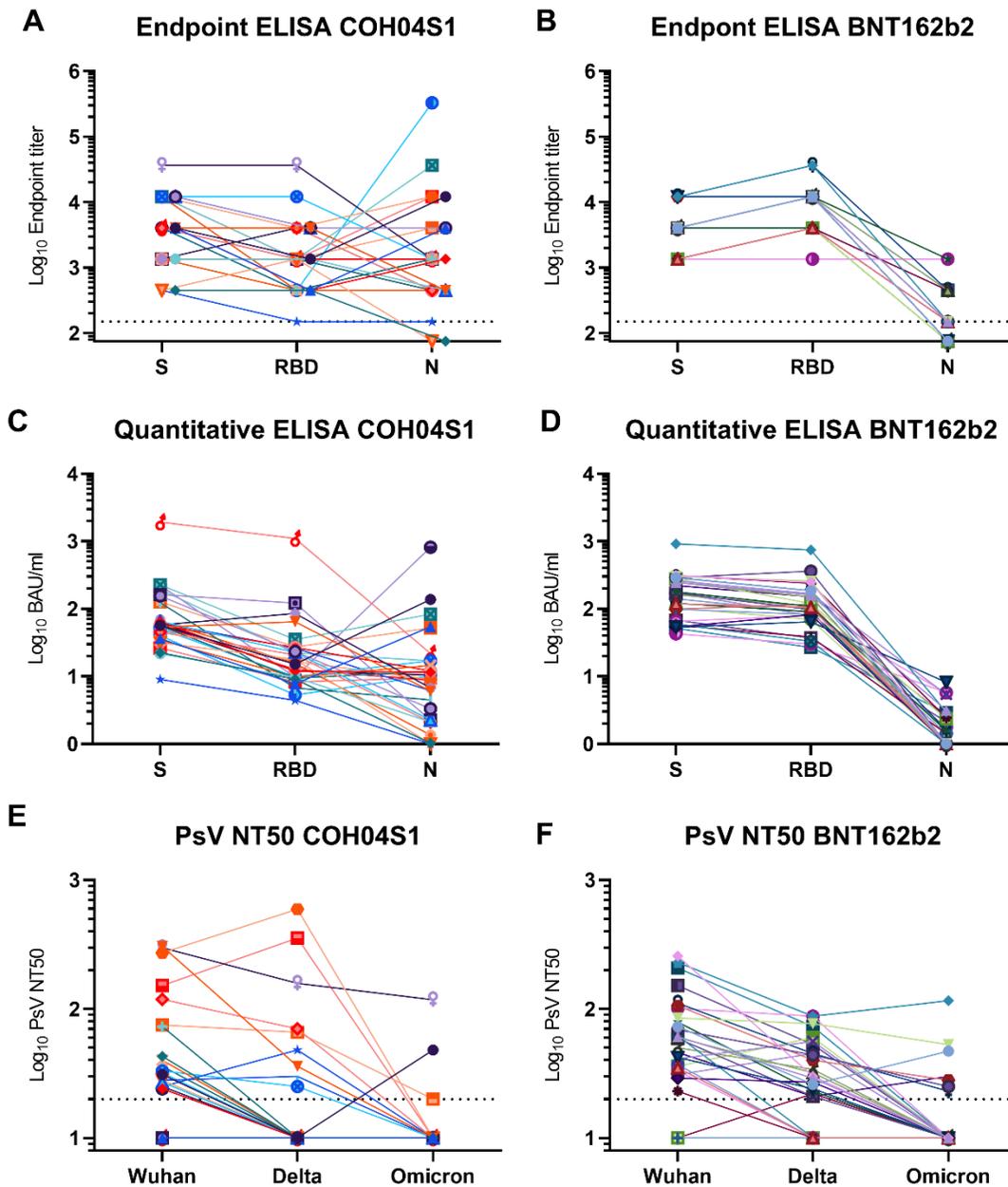


Figure S1. Related to Figure 1. SARS-CoV-2 antigen-specific binding antibodies and VOC-specific neutralizing antibodies in COH04S1 and BNT162b2 vaccinees six months or more after primary vaccination. a-d. Binding antibody titers to spike (S), receptor binding domain (RBD), and nucleocapsid (N) were measured by endpoint (A-B) and quantitative (C-D) ELISA in serum samples from COH04S1- (A, C) and BNT162b2- (B, D) vaccinated subjects six months or more after the primary vaccination series. Shown are the individual measurements spanning across SARS-CoV-2 antigens. Dotted lines in A-B represent the lower limit of quantifications. Samples with titers below the limit of quantification were indicated with an endpoint titer of 75. **E-F.** 50% neutralizing antibody titers (NT50) against ancestral SARS-CoV-2 (Wuhan-Hu-1), Delta and Omicron variants were measured using a pseudovirus (PsV) assay in serum samples from

COH04S1- (E) and BNT162b2- (F) vaccinated subjects six months or more after the primary vaccination series. Shown are the individual measurements spanning across SARS-CoV-2 variants. Dotted lines represent the lower limit of quantification. Samples with titers below the limit of quantification were indicated with an NT50 titer of 10.

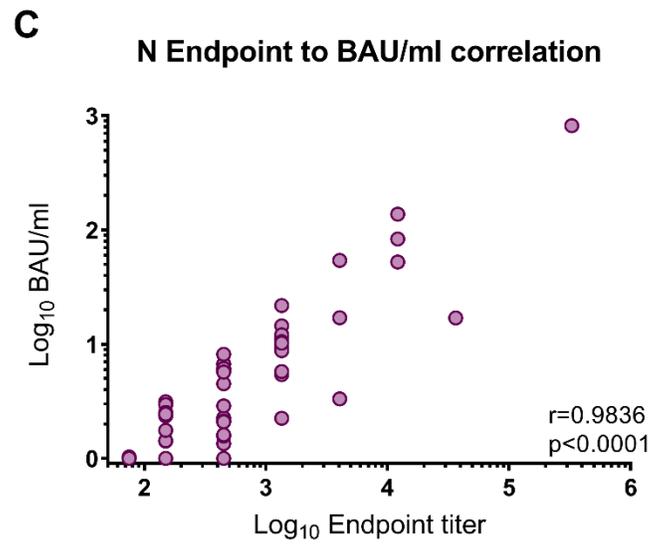
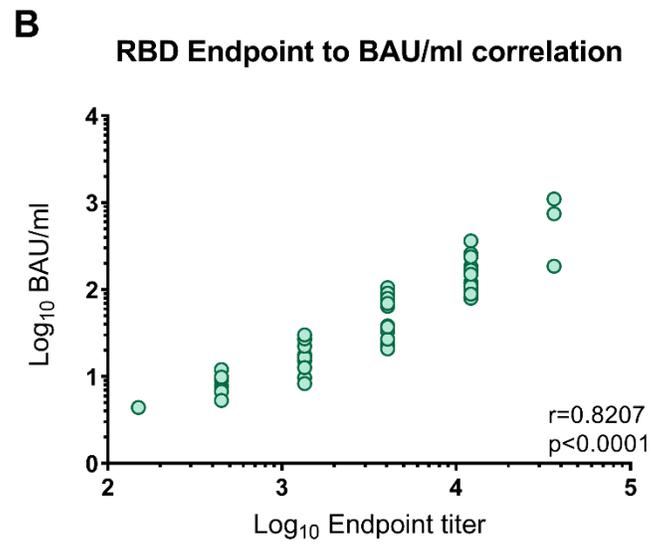
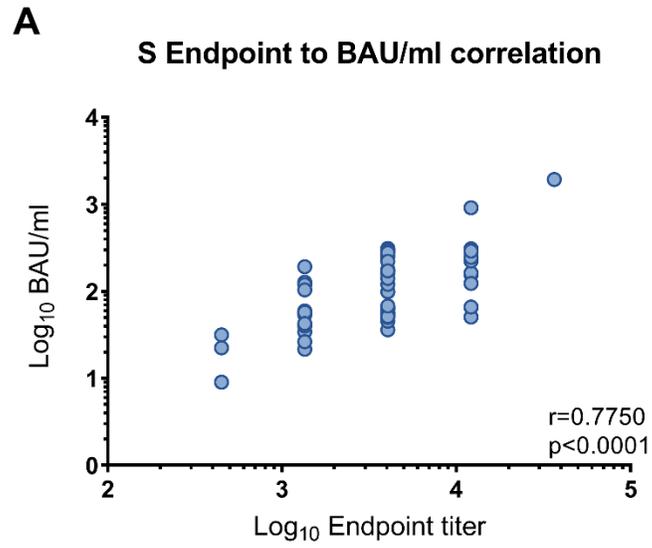


Figure S2. Related to Figure 1. Correlation of SARS-CoV-2 antigen-specific endpoint titers to binding antibody units in COH04S1 and BNT162b2 vaccinees six months or more after primary vaccination. Binding antibody titers to spike (S), receptor binding domain (RBD), and nucleocapsid (N) were measured by endpoint and quantitative ELISA in serum samples from COH04S1- and BNT162b2-vaccinated subjects six months or more after the primary vaccination series. Shown is the correlation analysis of S (A), RBD (B), and N (C) IgG endpoint titers to binding antibody units (BAU/ml). Shown in each figure is two-tailed Pearson's correlation coefficient (r) and its significance (p).

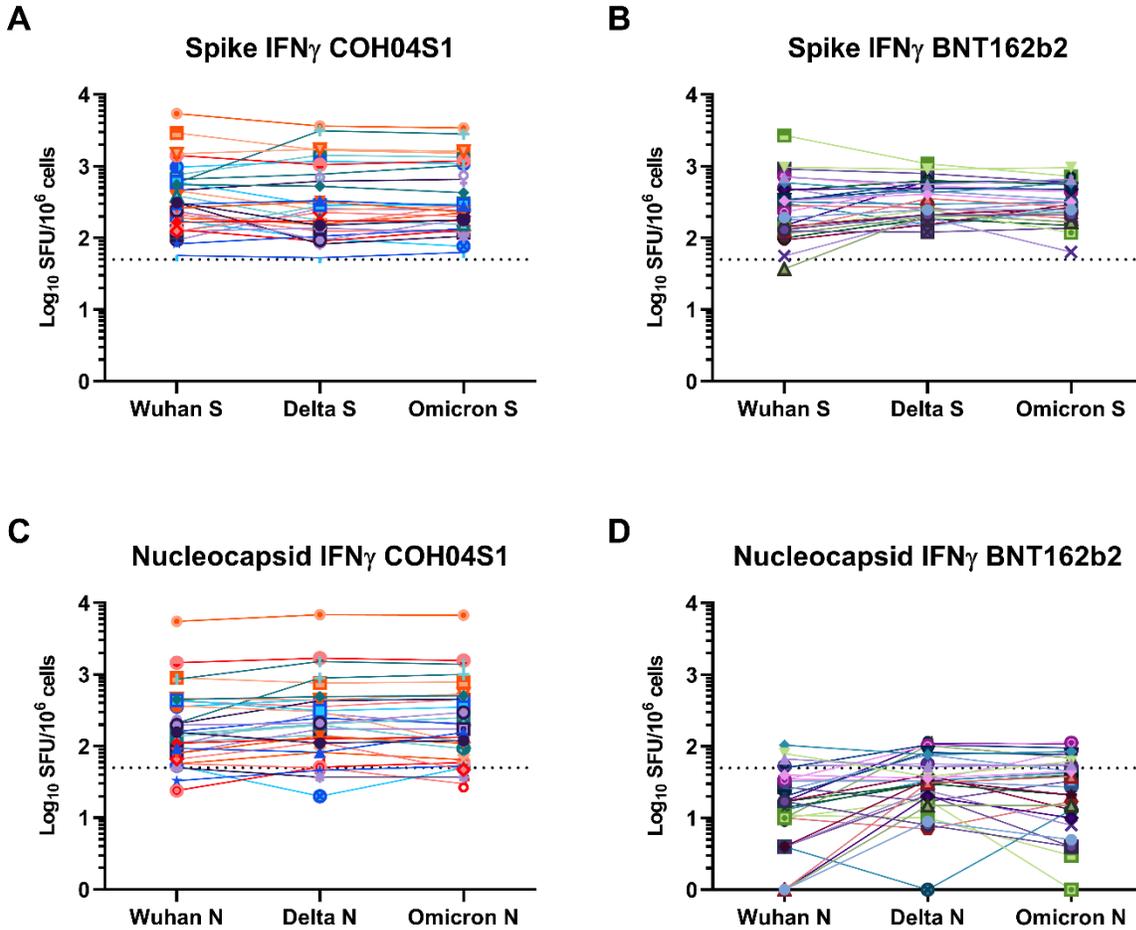


Figure S3. Related to Figure 2. SARS-CoV-2 ancestral and variant-specific IFN γ T cell responses in COH04S1 and BNT162b2 vaccinees six months or more after primary vaccination. Ancestral (Wuhan-Hu-1), Delta, and Omicron-specific spike (A, B) and nucleocapsid (C, D) IFN γ T cell responses were quantified by IFN γ ELISPOT upon PBMCs stimulation with S and N peptide libraries in subjects vaccinated with COH04S1 (A, C) or BNT162b2 (B, D) six months or more after the primary vaccination series. Shown are the individual IFN γ spot forming units (SFU) measured in 10^6 PBMCs and spanning across SARS-CoV-2 variants. Dotted lines represent the arbitrary threshold for positive response (50 spots/ 10^6 PBMCs).

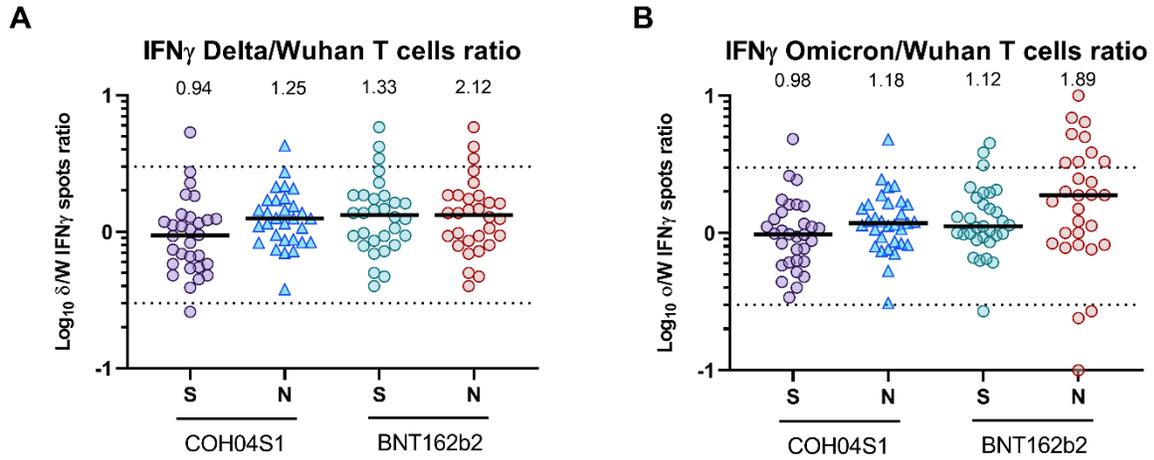


Figure S4. Related to Figure 2. Ratios of variant-specific to ancestral-specific IFN γ T cell responses in COH04S1 and BNT162b2 vaccinees six months or more after primary vaccination. Delta to Ancestral (Wuhan-Hu-1, A), and Omicron to ancestral (B) spike and nucleocapsid IFN γ T cell responses were quantified by IFN γ ELISPOT upon PBMCs stimulation with S and N peptide libraries in subjects vaccinated with COH04S1 or BNT162b2 six months or more after the primary vaccination series. Shown are IFN γ spot forming units (SFU) ratios. Bars represent medians with median values indicated above each group. Top dotted line represents a three-fold increase and the bottom dotted line represents a three-fold decrease in the ratio.

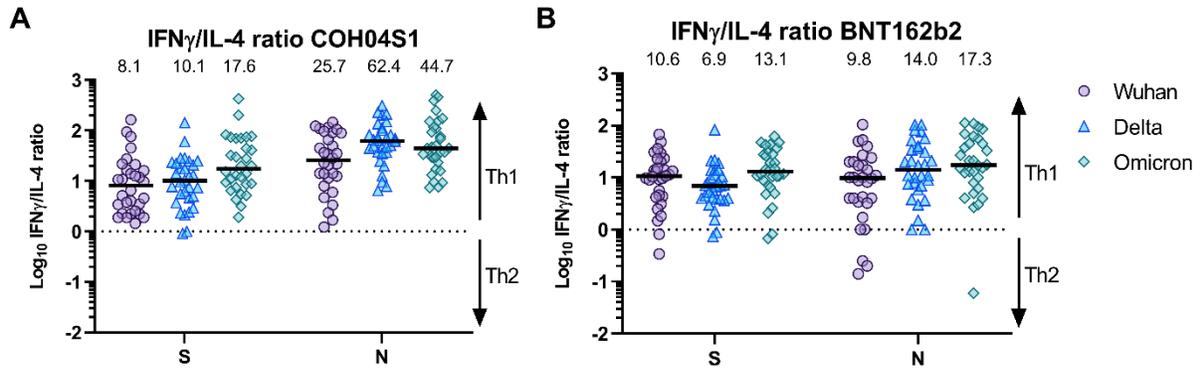


Figure S5. Related to Figure 2. Ratios of ancestral-specific IFN γ to IL-4 T cell responses in COH04S1 and BNT162b2 vaccinees six months or more after primary vaccination. Ancestral (Wuhan-Hu-1) specific spike and nucleocapsid IFN γ to IL-4 T cell ratios were quantified by IFN γ ELISPOT upon PBMCs stimulation with S and N peptide libraries in subjects vaccinated with COH04S1 (A) or BNT162b2 (B) six months or more after the primary vaccination series. Shown are IFN γ spot forming units (SFU) ratios. Bars represent medians with median values indicated above each group. Dotted line represents a ratio of 1. A ratio >1 is indicative of a Th1-biased response.

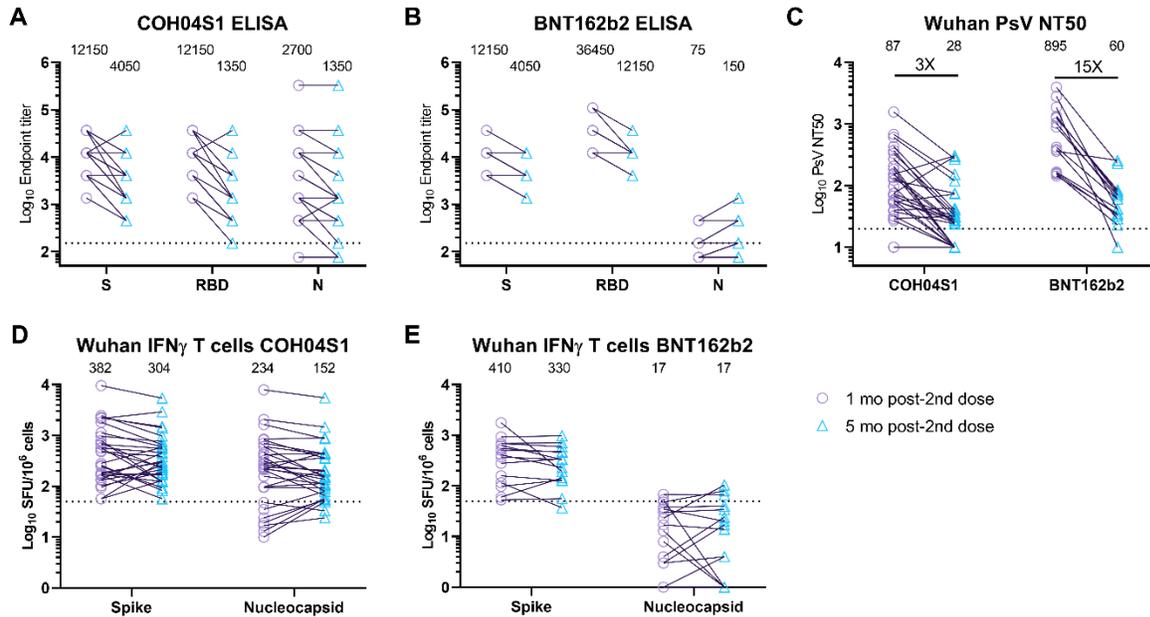


Figure S6. Related to Figures 1 and 2. Comparison of SARS-CoV-2-specific humoral and cellular responses in COH04S1 and BNT162b2 vaccinees one month and five months after the second vaccination. A-B. Binding antibodies. Binding antibody titers to Wuhan-Hu-1-specific spike (S), receptor binding domain (RBD), and nucleocapsid (N) antigens were measured by endpoint ELISA in serum samples of COH04S1- (A) and BNT162b2- (B) vaccinated subjects one month and five months after the second vaccination. **C.** NAb responses. 50% neutralizing antibody titers (NT50) against ancestral SARS-CoV-2 (Wuhan-Hu-1) were measured using a pseudovirus (PsV) assay in serum samples of COH04S1- and BNT162b2-vaccinated subjects one month and five months after the second dose. Samples with titers below the limit of quantification were indicated with an NT50 titer of 10. Dotted lines in A-C represents the lower limit of quantification. **D-E.** Cellular responses. Ancestral (Wuhan-Hu-1)-specific S and N IFN_γ T cell responses were quantified by IFN_γ upon PBMCs stimulation with S and N peptide libraries in subjects vaccinated with COH04S1 (D) or BNT162b2 (E) one month after and five months after the second vaccination. Shown are the IFN_γ spot forming units (SFU) measured in 10⁶ PBMCs. Dotted lines represent the arbitrary threshold for positive response (50 spots/10⁶ PBMCs). For BNT162b2, only the 15 subjects that had available samples from both timepoints are included. Median values are indicated above each group.

Table S1. Related to Figures 1-3. Volunteers characteristics

Characteristic	COH04S1, N = 30	DL1, N = 18	DL2, N = 6	DL3, N = 6	BNT162b2, N = 30
Age ¹	38 (22, 54)	44 (22, 50)	36 (28, 54)	29 (25, 41)	56 (24, 75)
Gender					
Female ²	16 (53%)	10 (56%)	3 (50%)	2 (33%)	22 (73%)
Male ²	14 (47%)	8 (44%)	3 (50%)	4 (67%)	8 (27%)
Days from first dose ¹	180 (180-242)	180 (180-242)	180 (180-180)	180 (180-180)	209 (180-405)

¹Median (Range); ²n (%). DL= dose level

Table S2. Related to Figures 1-3. Individual characteristics of subjects vaccinated with COH04S1

ID#	Age (y)	Sex at birth	Days post dose 1	COH04S1 DL
COH001	48	F	180	DL1
COH002	42	M	180	DL1
COH003	45	F	180	DL1
COH004	28	F	180	DL1
COH006	42	M	180	DL1
COH007	28	M	180	DL2
COH013	38	F	180	DL2
COH014	54	F	180	DL2
COH016	48	F	180	DL1
COH017	29	M	180	DL2
COH018	48	M	180	DL1
COH019	34	F	180	DL2
COH021	50	M	180	DL1
COH022	41	M	180	DL3
COH023	30	F	242	DL1
COH027	26	F	180	DL3
COH028	34	M	180	DL3
COH030	25	M	180	DL1
COH033	22	F	208	DL1
COH034	44	F	208	DL1
COH036	28	M	180	DL3
COH037	29	F	180	DL3
COH039	50	F	180	DL1
COH044	44	F	208	DL1
COH045	30	F	180	DL1
COH046	43	F	180	DL1
COH047	38	M	208	DL1
COH051	47	M	180	DL1
COH058	38	M	180	DL2
COH060	25	M	180	DL3

y=years; DL=dose level

Table S3. Related to Figures 1-3. Individual characteristics of subjects vaccinated with BNT162b2

ID#	Age (y)	Sex at birth	Days post dose 1
EUA001	34	F	180
EUA002	38	F	180
EUA003	53	M	180
EUA004	48	M	180
EUA006	54	F	180
EUA007	24	M	180
EUA009	56	F	180
EUA010	66	F	180
EUA011	37	M	180
EUA012	61	F	180
EUA014	52	F	180
EUA016	42	F	180
EUA017	50	F	180
EUA018	40	F	180
EUA019	50	F	180
EUA021	60	F	263
EUA023	56	F	289
EUA024	60	F	281
EUA027	56	F	257
EUA028	57	F	280
EUA029	56	F	265
EUA030	64	M	238
EUA035	61	M	281
EUA038	61	F	312
EUA041	62	M	241
EUA044	59	F	405
EUA048	75	F	283
EUA049	56	F	399
EUA050	57	M	380
EUA054	63	F	378

y=years