

Supplementary figure 1: Single cell sorting strategy for haemagglutinin specific B cells and abundance of haemagglutinin specific B cells

- (B) Example flow sorting gating strategy on B cells. B cells were negatively separated from PBMC using magnetic sorting prior to flow sorting.
- (C) The proportion of haemagglutinin binding B cells (as % of live B cells), is not significantly different between age groups either before, or 6 weeks after TIV immunization. (D-E) The number of haemagglutinin binding B cells per  $10^7$  PBMC analysed by age and days post vaccination.
- (D) The number of haemagglutinin binding B cells is higher in 22-36 year olds at day 42. The number of haemagglutinin binding B cells was calculated as in (C). In (C) and (E, the P values shown are from an unpaired two-tailed Mann-Whitney test. In (D), samples from the same individual are indicated with a grey line. The P values shown are from a paired two-tailed Mann-Whitney test.

## SessionInfo

```
## R version 3.6.1 (2019-07-05)
## Platform: x86_64-pc-linux-gnu (64-bit)
## Running under: CentOS Linux 7 (Core)
## Matrix products: default
           /bi/apps/R/3.6.1/lib64/R/lib/libRblas.so
## LAPACK: /bi/apps/R/3.6.1/lib64/R/lib/libRlapack.so
##
## locale:
## [1] LC_CTYPE=en_GB.UTF-8
                                   LC NUMERIC=C
                                   LC_COLLATE=en_GB.UTF-8
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##
   [5] LC_MONETARY=en_GB.UTF-8
                                   LC_MESSAGES=en_GB.UTF-8
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                                   LC_NAME=C
   [9] LC_ADDRESS=C
                                   LC_TELEPHONE=C
## [11] LC_MEASUREMENT=en_GB.UTF-8 LC_IDENTIFICATION=C
##
## attached base packages:
## [1] parallel stats4
                           stats
                                     graphics grDevices utils
                                                                    datasets
## [8] methods
##
## other attached packages:
##
  [1] ggpubr_0.2.4
                                    magrittr_1.5
   [3] forcats 0.4.0
                                    stringr 1.4.0
##
                                    purrr 0.3.3
## [5] dplyr_1.0.2
## [7] readr_1.3.1
                                    tidyr_1.0.0
## [9] tibble_3.0.4
                                     ggplot2_3.3.2
## [11] tidyverse_1.3.0
                                    SingleCellExperiment_1.8.0
## [13] MultiAssayExperiment_1.12.1 SummarizedExperiment_1.16.0
## [15] DelayedArray_0.12.0
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## [17] matrixStats_0.55.0
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## [19] GenomicRanges_1.38.0
## [21] IRanges_2.20.1
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## [23] BiocGenerics_0.32.0
## loaded via a namespace (and not attached):
  [1] httr 1.4.1
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                                                       modelr_0.1.5
  [4] assertthat_0.2.1
                               GenomeInfoDbData_1.2.2 cellranger_1.1.0
## [7] yaml_2.2.0
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                                                       backports 1.1.5
## [10] lattice_0.20-38
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## [13] gridtext_0.1.4
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                                                       ggsignif_0.6.0
## [16] rvest 0.3.5
                                                       cowplot_1.0.0
                               colorspace_1.4-1
## [19] htmltools_0.5.2
                               Matrix_1.2-17
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## [22] pkgconfig_2.0.3
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## [25] zlibbioc_1.32.0
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## [31] cli_2.0.0
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## [34] ggtext_0.1.1
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## [37] fansi_0.4.0
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## [46] grid_3.6.1
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## [52] gtable_0.3.0	DBI_1.1.0	markdown_1.1	
## [55] reshape2_1.4.3	R6_2.4.1	lubridate_1.7.4	
## [58] knitr_1.26	fastmap_1.1.0	stringi_1.4.3	
## [61] Rcpp_1.0.3	vctrs_0.3.6	dbplyr_1.4.2	
## [64] tidyselect_1.1.0	xfun_0.31		