

Supplemental Materials

Title: Non-preferential, but detrimental accumulation of macrophages with clonal hematopoiesis-driver mutations in cardiovascular tissues

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Materials and Methods

CHIP mutation screening

Genomic DNA (gDNA) of 350 μ L citrated whole blood was extracted using DNeasy Blood and Tissue Kit (QIAGEN). Fifty nanogram of total gDNA of each sample was constructed into a targeted genomic library according to the instruction of the TruSight Myeloid Sequencing Panel (Illumina) and sequenced either on a MiSeq instrument with 220 bp paired-end reads or on a NextSeq500 instrument with 150 bp paired-end reads. Each sample was sequenced with a mean depth of 5.2 million reads. Data analysis was performed on the Galaxy Europe platform (usegalaxy.eu). In brief, we first applied Trim Galore! (v0.4.3.1) to remove adapters, low-quality reads (Phred score < 28), and 22 bp from the 5' end of both read 1 and read 2. The trimmed reads were mapped to the human genome assembly GRCh37 (hg19) using bowtie2 (v2.3.4.3). After converting bam files to pileup using Generate pileup (v.1.1.3), we identified variants at positions having a base quality of Phred score > 28 with VarScan (v2.4.2) according to the consensus genotype. The variants were subsequently annotated with SnpEff (v4.3) and GEMINI (v0.20.1). Confirmed CHIP variants should be covered with a read depth (DP) > 2500 and carry a variant allele frequency (FREQ) above 2% on exonic regions. We ruled out variants with a FREQ of 40% to 55% or above 90%, or those occur in more than 0.5% of non-Finnish European population to exclude germline mutations. Variants fulfilling the following criteria were considered technical artifacts and thus excluded: 1. Having a discrepancy of FREQ above 50% between forward and reverse reads. 2. Occurring in more than 10% of the samples of the same library. 3. Locating on the oligo positions of the TruSight Myeloid Sequencing Panel. 4. Occurring only in a specific amplicon at the position where multiple amplicons overlap.

Blood and tissue sample preparation

Ten mL citrated whole blood from each patient first underwent red blood cell lysis for 10 minutes and washed with FACS buffer (Ca/Mg²⁺ free phosphate buffered saline (PBS) + 5% fetal calf serum). Aortic plaque or heart samples were shredded into fine pieces by mechanical forces and incubated in digestion mixes at 37 °C (**Details listed in Table S4**). The digested tissue were filtered through strainers and washed with FACS buffer to retrieve single cell suspensions. Thereafter, the cells were stained with the corresponding FACS panels (**Table S5**) at 4 °C for 30 minutes and sorted using BD Aria III (BD Biosciences) or MoFlo Astrios EQ (Beckman Coulter) following the gating strategy presented in **Figure S2**. The cells were directly deposited in DNA LoBind tubes (Eppendorf) containing buffer RLT Plus (QIAGEN) with 1% β -Mercaptoethanol (PanReac AppliChem) and stored at -20 °C. DNA and RNA of sorted cells were extracted using AllPrep DNA/RNA micro kit (QIAGEN) according to the manufacturer's instruction. DNA amount of blood cells was measured using high-sensitivity Qubit dsDNA assay (ThermoFisher), while DNA amount of tissue cells were extrapolated based on the sorted cell counts.

Fragment analysis of 6-bp insertion

For Carrier 7, who harbored a 6-bp insertion on the CEBPA gene, we quantified VAF of sorted cells by fragment analysis. One ng of isolated DNA from the sorted cells were amplified using the high-fidelity Platinum SuperFi polymerase (ThermoFisher) by 40 cycles. Fragment analysis was performed using Fragment Analyzer (Agilent Technologies), following the instructor's guidance. The data were analyzed with PROSize (Agilent Technologies) and the VAF was computed using the relative fluorescence units (RFU) of the 205 bp (wild-type) and 211 bp (mutated) fragments (**Figure S4**).

Bulk RNA-seq data analysis

The fastq files were first processed into count matrices on the Galaxy Europe platform. In short, after assuring data quality with FASTQC (v 0.74), we ~~first~~ applied Trim Galore! (v0.4.3.1) to remove adapters and low-quality reads (Phred score < 28) and mapped the trimmed reads to human genome assembly GRCh38 (hg38) using RNA STAR (v2.7.10). The gene expression was quantified using featureCounts (v2.0.3). We then combined the count table of each sample into gene expression matrices and further performed the analysis in R (v4.1). The data of patients undergoing carotids endarterectomy and those undergoing heart surgery were analyzed separately. After removal of ribosome genes, the rest of genes with total counts between 5,000 to 500,000 among all samples were used to compute DEGs using DESeq2 (v1.40.1). Normalized count matrices generated by DESeq2 were used for making heatmaps with pheatmap (v1.0.12). The normalized counts were also used for comparing CCR2+ and CCR2– heart macrophages from the same carriers. The biological process enrichment was analyzed using clusterProfiler (v4.8.1). The Volcano plots were generated using EnhancedVolcano (v1.18.0). Module scores were calculated using all genes categorized in the specific gene ontology (GO) terms (**Table S6**).

Statistical analysis

Patient clinical characteristics were analyzed using GraphPad Prism (v10.0.2) to compute the descriptive statistics. Continuous variables were shown as mean with standard deviations and the statistical significance between carriers and matched non-carriers were tested using two-sided Mann-Whitney test. Categorical variables were shown as frequency (%) and the statistical significance between carriers and matched non-carriers were tested using Fisher's exact test.

The ddPCR readouts were analyzed using QuantaSoft (v1.7) to acquire the number of droplets containing mutant and non-mutant amplicons. The VAF was acquired by calculating the proportion of droplets containing mutant amplicons among all droplets that contain the targeted amplicons (**Figure S3B**). The correlation of VAF between blood monocytes and other cell types were analyzed using GraphPad Prism to compute Pearson's correlation coefficients (r) and to visualize the fit lines with 95% confidence bands with simple linear regression. The comparison of next generation sequencing and ddPCR methods was conducted using a paired non-parametric Friedman test.

DEGs of the bulk RNA-seq data were computed with DESeq2 (v1.40.1) using Wald test with Benjamini and Hochberg (BH) method to adjust p values. The enriched GO terms by upregulated DEGs were analyzed using clusterProfiler (v4.8.1) with BH method to adjust p values. The statistical analysis of module scores were computed using GraphPad Prism with one-tailed Mann-Whitney test. The difference of transcriptomic features between CCR2+ and CCR2- heart macrophages was analyzed by calculating the mean of gene expression ratio between the two cell types from the same individuals. The p value was computed with a two-tailed student's t -test.

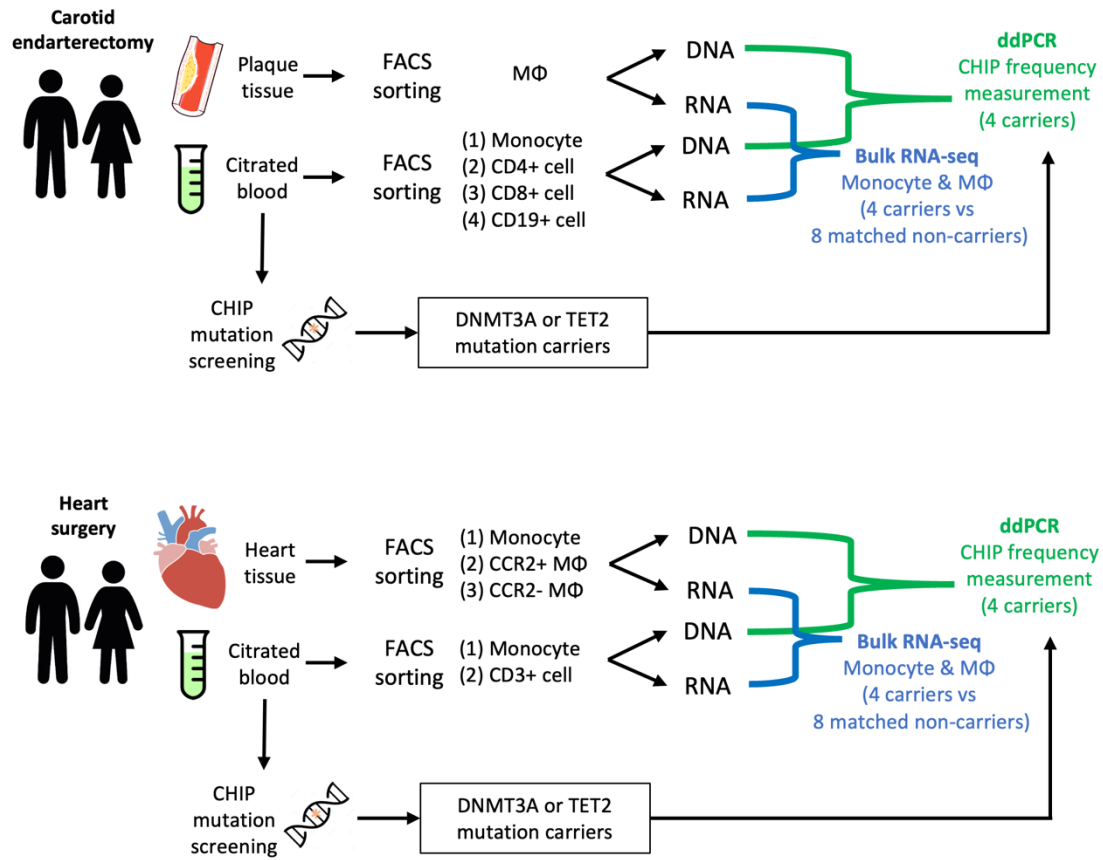
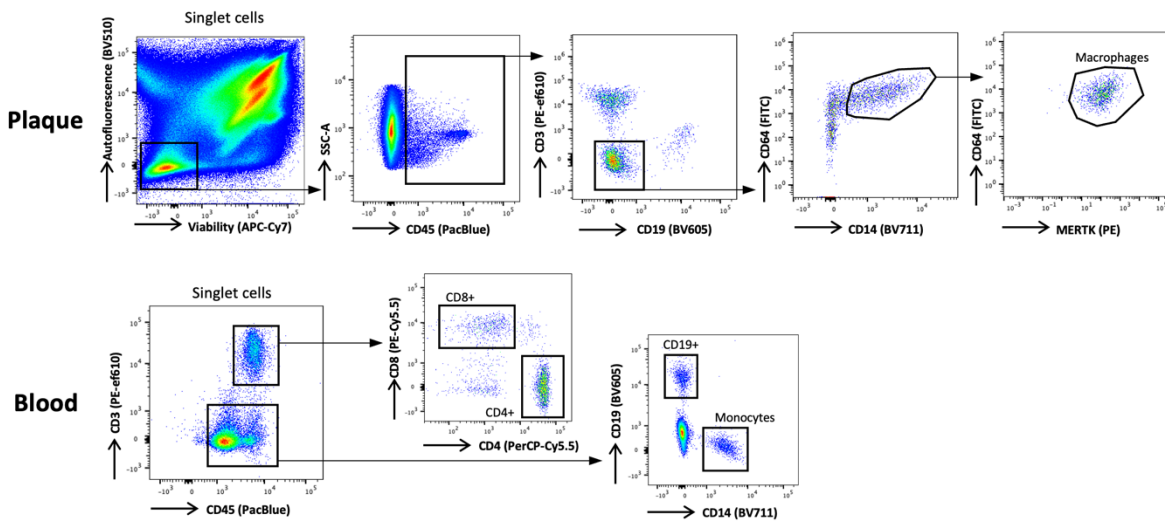


Figure S1. Study design and workflow

Schematic illustration of the study workflow. MΦ, macrophages.

Carotid endarterectomy



Heart surgery

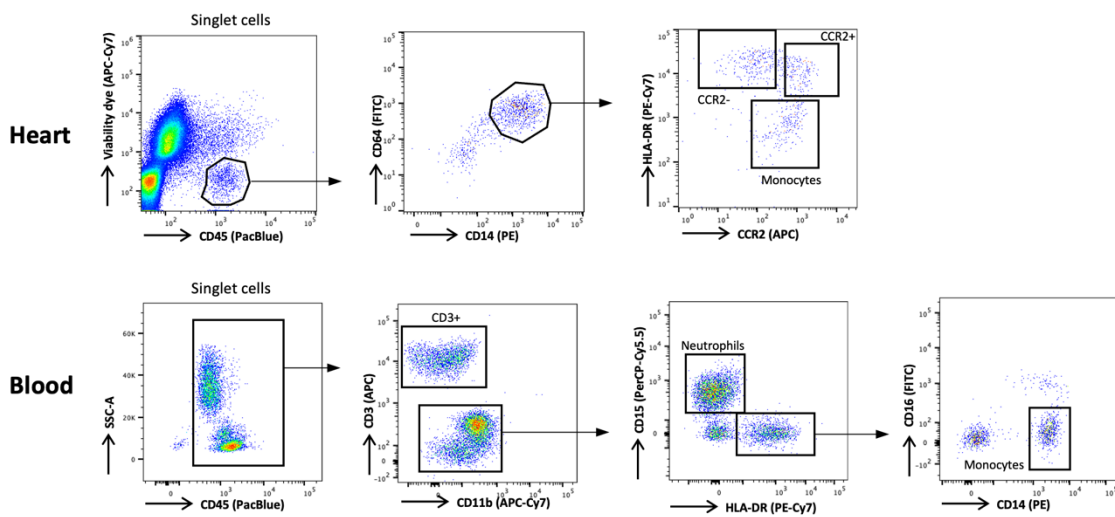


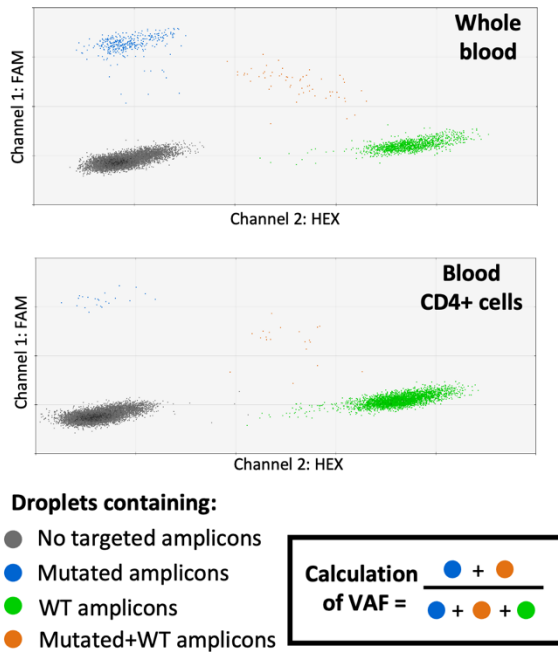
Figure S2. FACS gating strategy of blood and tissue samples

FACS dot plots illustrating the gating strategy of myeloid cells and lymphocytes from blood and tissue samples in the two patient cohorts. Macrophages in the plaque were identified as CD45+ CD3- CD19- CD14+ CD64+ MerTK+. Macrophages in the heart were identified as CD45+ CD14+ CD64+ HLA-DR+ and further subdivided into CCR2+ and CCR2- populations. Monocytes, Neutrophils and lymphocytes were identified as depicted by the gating strategy.

(A)

ddPCR primers and probes used for each individual carriers	
Carrier 1 (DNMT3A G800S) Primer-Forward: AGTCCCAGCCACAG Primer-Reverse: CAGCTGCACACAGG Probe-Mutated: CCTGTTCA+TAC+ <u>CGG</u> +GAA+GGTTAC Probe-WT: CCTGTTCA+TAC+ <u>TGG</u> +GAA+GGTTAC	Carrier 6 (IDH1 R132H) Primer-Forward: GCCAACATGACTTACTTG Primer-Reverse: TGTGGAAATCACCAATG Probe-Mutated: TAA+GCA+TGA+ <u>TGA</u> +CCTATGA Probe-WT: TAA+GCA+TGA+ <u>CGA</u> +CCTATGA
Carrier 2 (DNMT3A C557*) Primer-Forward: AGCGAAGAACATCTGGAG Primer-Reverse: GAGAGTCTCTCTGCTCA Probe-Mutated: CTTC+CAG+GTG+ <u>CTT</u> +TTGCGT Probe-WT: CTTC+CAG+GTG+ <u>ATT</u> +TTGCGT	Carrier 7 (CEBPA P232HPP) Primer-Forward: CGATCTGGAAGTGCAGGTG Primer-Reverse: CTGGAGCCCCTGTACGAG (*The 6-bp insertion was quantified by fragment analysis.)
Carrier 3 (TET2 W954*) Primer-Forward: CCAGAAAGGACACTCAAAA Primer-Reverse: CTGACTATGGCAAGACTC Probe-Mutated: C+TAA+GGT+ <u>GGC</u> +ATCTCTT Probe-WT: C+TAA+GGT+ <u>AGC</u> +ATCTCTT	Carrier 8 (TET2 K1197*) Primer-Forward: TGGGTTTCTTTAAGGTTTG Primer-Reverse: AGGCTTTATCAAGTCACA Probe-Mutated: TTACCC+ACT+ <u>TAG</u> +CAA+TAGG Probe-WT: TTACCC+ACT+ <u>AAG</u> +CAA+TAGG
Carrier 4 (DNMT3A W297G) Primer-Forward: CTGGCCACCAGGAGAAGC Primer-Reverse: CACCAAAGCAGGCGATGA Probe-Mutated: AGTT+TCC+CCC+ <u>ACA</u> +CCAGC Probe-WT: AGTT+TCC+CCC+ <u>CCA</u> +CCAGC	Carrier 9 (DNMT3A V563M) Primer-Forward: GGTCTTCCTTAATGGCTG Primer-Reverse: GGTCTTTCCATTCCAGGTA Probe-Mutated: ACCAAGAGGTCCA <u>T</u> ACTCCACG Probe-WT: ACCAAGAGGTCCA <u>C</u> ACTCCACG
Carrier 5 (IDH1 R132H) Primer-Forward: GCCAACATGACTTACTTG Primer-Reverse: TGTGGAAATCACCAATG Probe-Mutated: TAA+GCA+TGA+ <u>TGA</u> +CCTATGA Probe-WT: TAA+GCA+TGA+ <u>CGA</u> +CCTATGA	Carrier 10 (DNMT3A R736L) Primer-Forward: CTTGTCTACTAACGCCCATGG Primer-Reverse: GAGCCACACCACTGTCCTAT Probe-Mutated: TTC+TAC+ <u>CGC</u> +CTC+CTGC Probe-WT: TTC+TAC+ <u>CTC</u> +CTC+CTGC
Carrier 11 (DNMT3A R729W) Primer-Forward: CTTGTCTACTAACGCCCATGG Probe-Mutated: TCAAAGAAGAGCC <u>A</u> GCCAGTGCCTC	Primer-Reverse: GAGCCACACCACTGTCCTAT Probe-WT: TCAAAGAAGAGCC <u>G</u> CAGTGCCTC

(B)



(C)

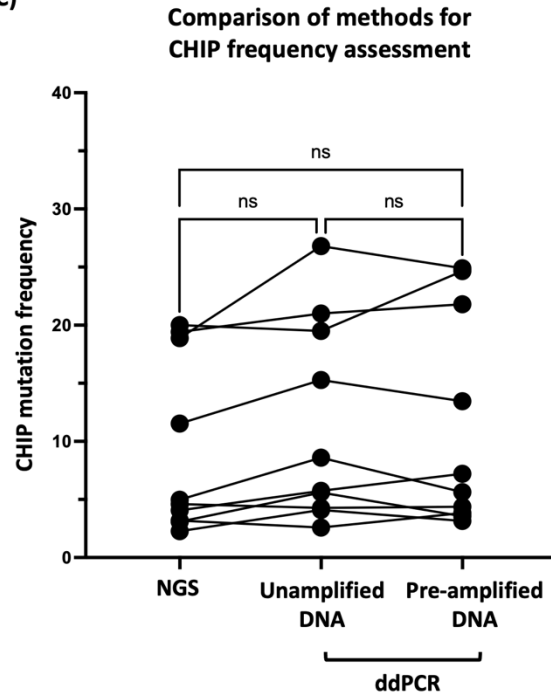


Figure S3. ddPCR method establishment and validation

(A) Oligonucleotide sequence of primers and probes targeting specific mutated site of each carrier. Probes targeting mutated and wild-type (WT) gDNA were labeled with FAM and HEX, respectively. + denotes locked nucleic acids. Underlined nucleotide in probes indicates the single nucleotide polymorphism of the carriers. **(B)** Schematic illustration of how VAF was calculated based on the ddPCR results. Adequate ddPCR results with at least one double-negative cluster and one single-positive cluster in the two-dimensional figure were selected for gating and droplet counting. Each sample had at least three technical replicates. **(C)** A dot plot comparing the CHIP mutation frequency in the whole blood measured by three methods, namely next generation sequencing (NGS) ddPCR using unamplified DNA, and pre-amplified DNA. The lines connect dots of the same carriers. ns denotes no significant differences between the three methods evaluated by paired non-parametric Friedman test.

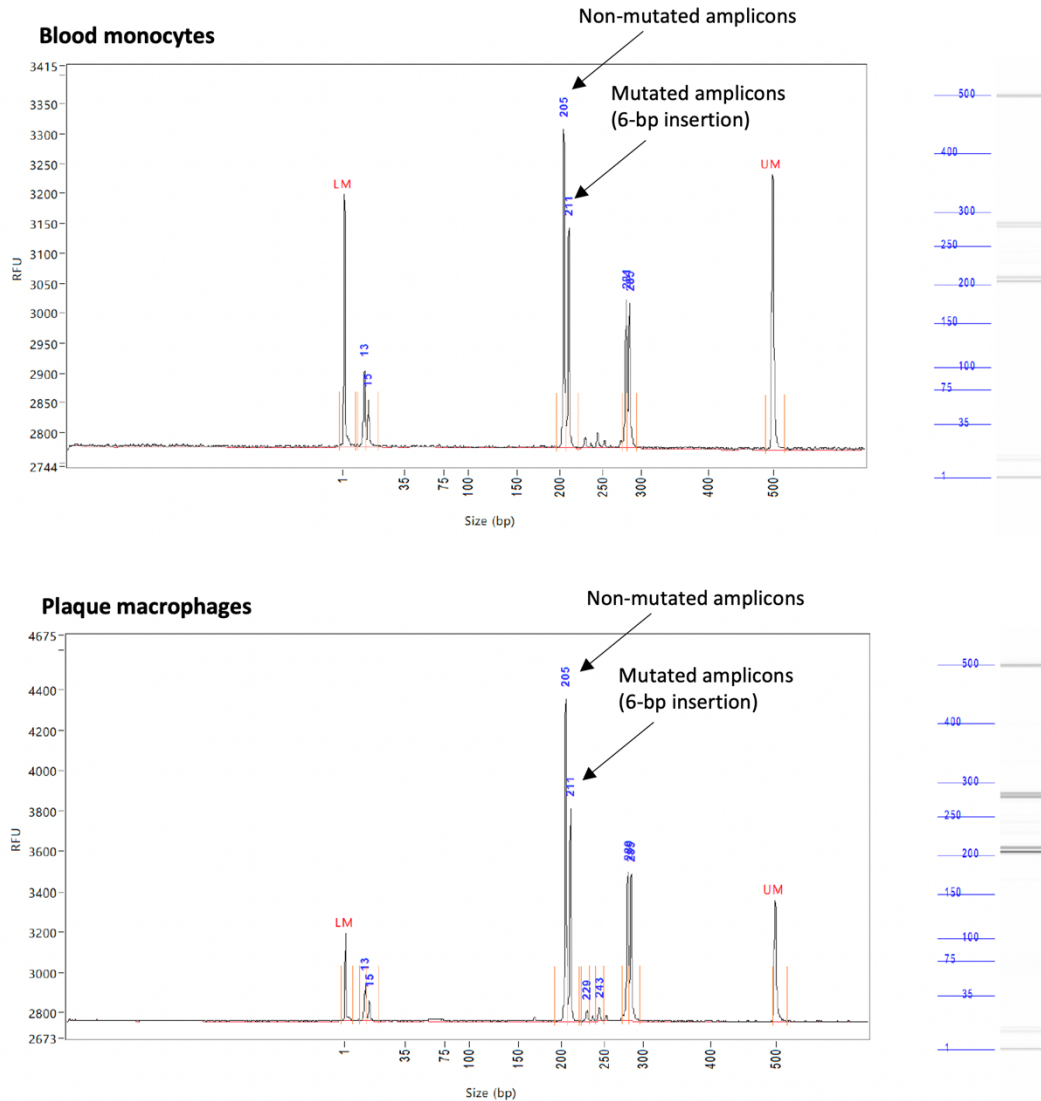


Figure S4. Fragment Analysis to quantify CEBPA^{p.P232HPP} mutation

Reports of DNA fragment size of CEBPA from blood monocytes and plaque macrophages retrieved from Carrier 7. The designed primers aimed to produce a 205-bp amplicon of wild-type CEBPA. The 211-bp fragment indicates amplicons encompassing a 6-bp insertion. The relative fluorescence unit (RFU) indicates the quantity of each DNA fragments and was used to calculate VAF ($VAF = RFU_{211bp} \div (RFU_{211bp} + RFU_{205bp})$).

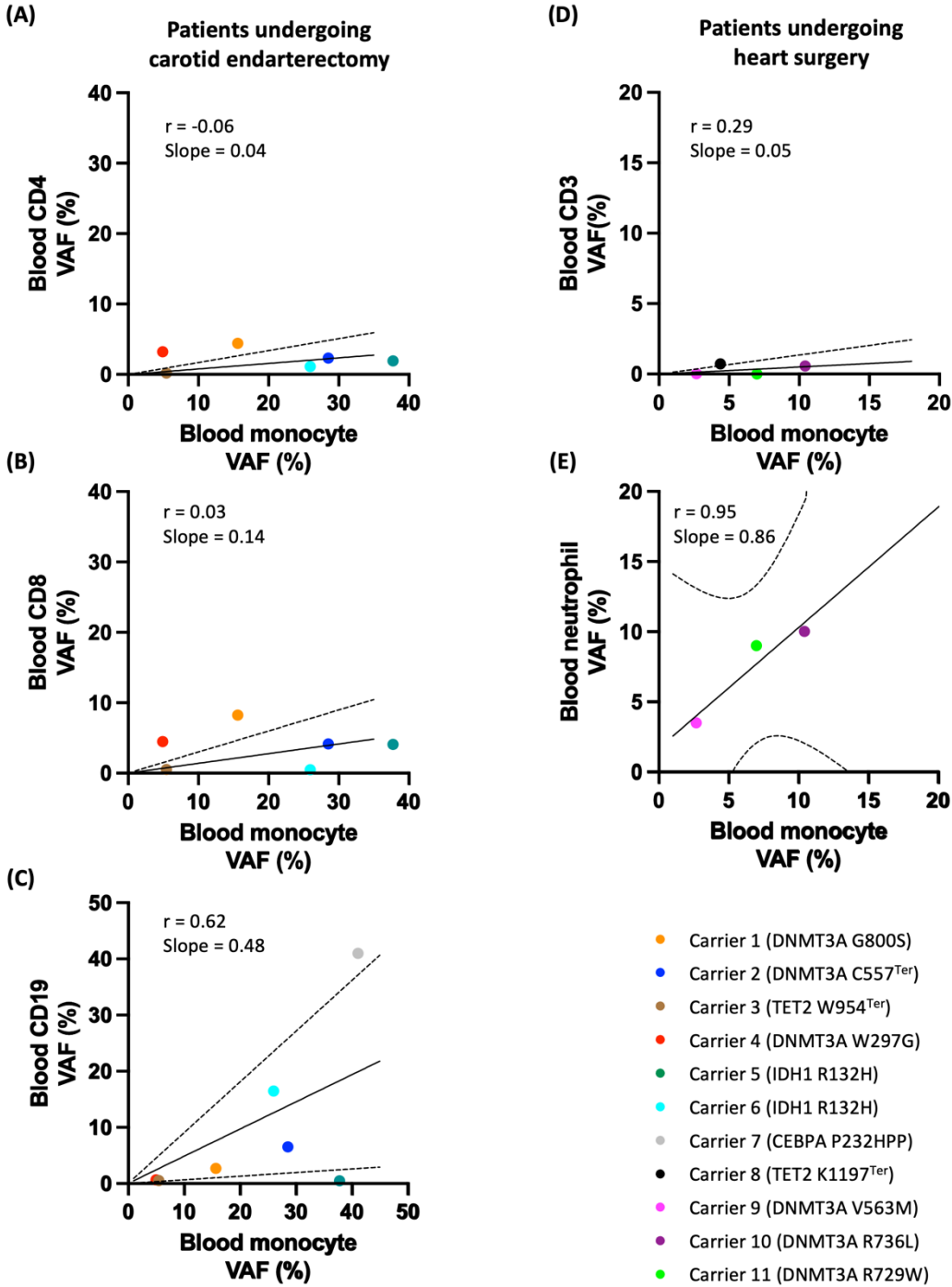


Figure S5. Correlation of CHIP clone size between blood monocytes and other cell types

Dot plots depicting VAF of blood monocytes and other blood cells retrieved from patients undergoing (A-C) carotid endarterectomy or (D-E) heart surgery. Simple linear regression was

performed to compute a fit line (solid line) and the 95% confidence bands (dotted lines) for each plot. The Pearson's correlation coefficient (r) was computed to evaluate correlation.

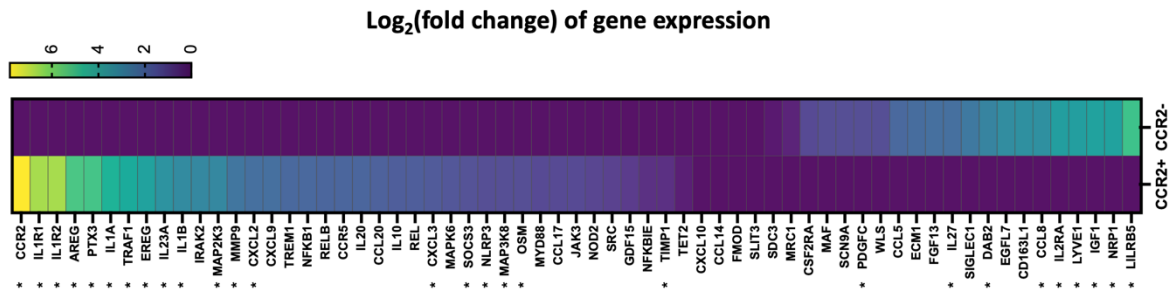


Figure S6. Transcriptomic features of CCR2+ and CCR2- heart macrophages

A heatmap showing the mean of logarithmic fold change of intra-individual gene expression ratio between the CCR2+ and CCR2- heart macrophages. * denotes a p -value < 0.05 of two-tailed student's t -test.

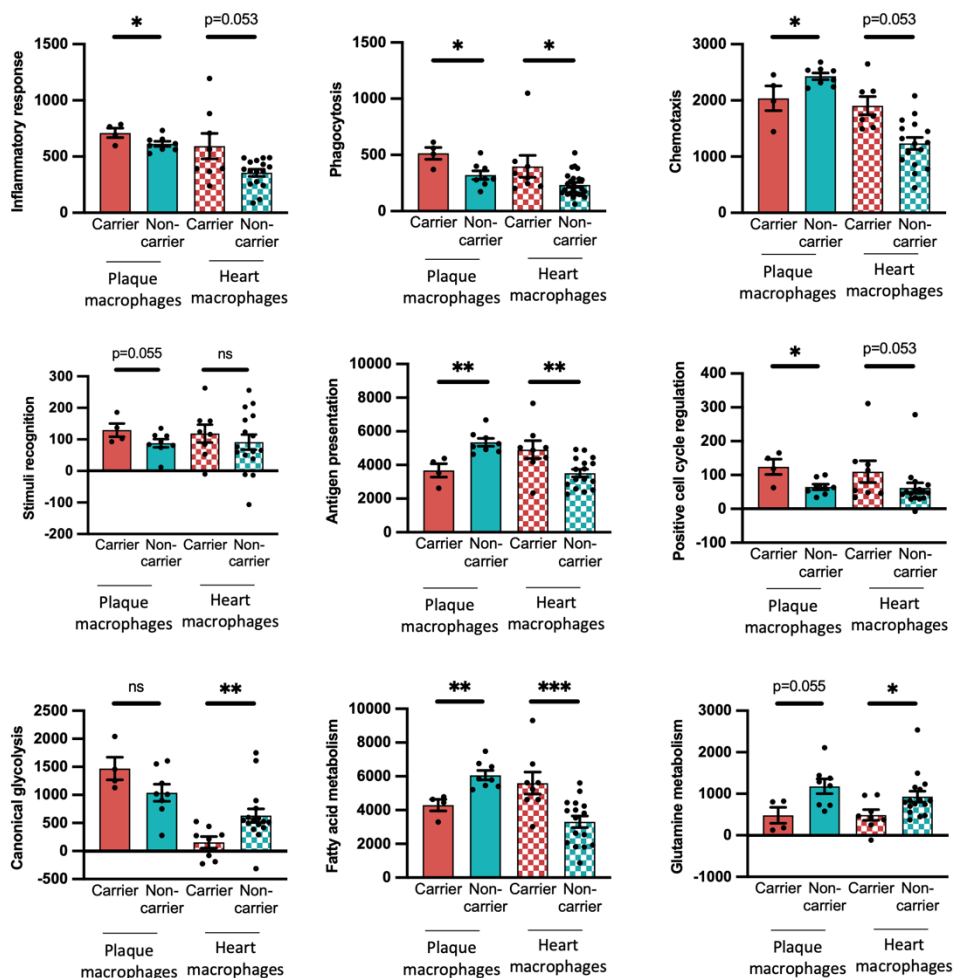


Figure S7. Module scores of biological processes associated with immune functions

Bar charts showing module scores of selected biological processes of tissue macrophages from carriers and non-carriers. Genes comprising the specific GO pathways (**Table S6**) were used for module score calculation. The p -values were calculated using one-tailed Mann-Whitney test. ns denotes no significant differences, * denotes p -value ≤ 0.05 , ** denotes p -value ≤ 0.01 , *** denotes p -value ≤ 0.001 .

Table S1. Clinical characteristics of patients undergoing carotid endarterectomy

	Carrier (n=4)	Non-carrier (n=8)	<i>p</i> -value
Sex [no.] (male:female)	3:1	5:3	>0.99
Age [yrs] Mean (SD)	78.75 (11.90)	67.13 (12.02)	0.28
Body mass index [kg/m ²] Mean (SD)	26.67 (4.41)	31.17 (10.32)	0.83
Side of endarterectomy [no.] (left:right)	2:2	5:3	>0.99
Degree of carotid artery stenosis [%] Mean (SD)	75 (7.07)	76 (6.9)	>0.99
Blood work			
Leukocyte [10 ³ /μL] Mean (SD)	8.65 (4.04)	8.85 (3.80)	0.81
Neutrophil [10 ³ /μL] Mean (SD)	6.29 (3.41)	5.97 (2.62)	0.78
Lymphocyte [10 ³ /μL] Mean (SD)	1.6 (0.57)	1.87 (0.79)	0.54
Monocyte [10 ³ /μL] Mean (SD)	0.66 (0.25)	0.70 (0.19)	0.82
Thrombocyte [10 ³ /μL] Mean (SD)	238.3 (58.07)	278.5 (85.11)	0.57
Hemoglobin [g/dL] Mean (SD)	11.53 (2.46)	12.28 (1.19)	0.77
Hematocrit [%] Mean (SD)	33.18 (6.75)	35.16 (3.71)	0.81
Hemoglobin A1c [%] Mean (SD)	5.98 (0.21)	6.08 (0.58)	0.69
Cholesterol [mg/dL] Mean (SD)	115.3 (22.32)	150.5 (38.49)	0.05
Homologous LDL [mg/dL] Mean (SD)	52.50 (20.98)	85.75 (29.73)	0.07
Homologous HDL [mg/dL] Mean (SD)	51.25 (7.4)	51.75 (14.0)	0.90
Lipoprotein(a) [mg/dL] Mean (SD)	42.50 (34.65)	63.33 (52.06)	0.85
Triglyceride [mg/dL] Mean (SD)	105.8 (36.34)	119.3 (53.95)	0.71
Medical history			
Coronary artery disease [no.] (%)	1 (25)	3 (37.5)	>0.99
Hypertension [no.] (%)	3 (75)	6 (75)	>0.99
Diabetes mellitus [no.] (%)	0 (0)	2 (25)	0.52

Transient ischemic attack [no.] (%)	1 (25)	1 (12.5)	>0.99
Stroke [no.] (%)	3 (75)	4 (50)	0.58
Use of statins [no.] (%)	4 (100)	8 (100)	>0.99

Continuous variables are shown as mean (SD); significance tested using two-sided Mann-Whitney test.

Categorical variables are shown as frequency (%); significance tested using Fisher's exact test.

HDL, high-density lipoprotein; LDL, low-density lipoprotein.

Table S2. Clinical characteristics of patients undergoing heart surgery

	Carrier (n=4)	Non-carrier (n=8)	<i>p</i> -value
Sex [no.] (male: female)	3:1	4:4	0.58
Age [yrs] Mean (SD)	69.50 (5.97)	61.50 (11.77)	0.25
Body mass index [kg/m ²] Mean (SD)	27.78 (6.20)	30.07 (4.54)	0.56
Left ventricular ejection fraction [%] Mean (SD)	0.50 (0.10)	0.52 (0.13)	0.37
Tissue source [no.] (left ventricle: right atrium)	1:3	2:6	>0.99
Blood work			
Leukocyte [10 ³ /μL] Mean (SD)	6.81 (0.57)	8.14 (1.95)	0.21
Thrombocyte [10 ³ /μL] Mean (SD)	204.5 (39.23)	262.1 (98.19)	0.23
Hemoglobin [g/dL] Mean (SD)	13.23 (1.31)	13.83 (1.48)	0.43
Hematocrit [%] Mean (SD)	39.50 (3.67)	40.83 (4.88)	0.79
Medical history			
Coronary artery disease [no.] (%)	1 (25)	3 (37.5)	>0.99
Hypertension [no.] (%)	1 (25)	3 (37.5)	>0.99
Diabetes mellitus [no.] (%)	2 (50)	1 (12.5)	0.23
Transient ischemic attack [no.] (%)	0 (0)	0 (0)	>0.99
Stroke [no.] (%)	0 (0)	0 (0)	>0.99
Use of statins [no.] (%)	2 (50)	4 (50)	>0.99

Continuous variables are shown as mean (SD); significance tested using two-sided Mann-Whitney test.

Categorical variables are shown as frequency (%); significance tested using Fisher's exact test.

Comparison of lipid profiles and Hemoglobin A1c was not conducted in the patients undergoing aortic valve replacement, because not more than three among the eight patients had these data available.

1 **Table S3. CHIP mutations of patients undergoing carotid endarterectomy or heart surgery**

2 **CHIP mutations identified among patients who underwent carotid endarterectomy**

POS	VAF	chrom	ref	alt	rs_ids	gene	exon	aa_change	impact	sex
209113112	19.42%	chr2	C	T	rs121913500	IDH1	4	R132H	missense_variant	female
25462009	11.54%	chr2	C	T	rs757083492	DNMT3A	20	G800S	missense_variant	male
25467204	20%	chr2	G	T	None	DNMT3A	15	C557*	stop_gained	female
209113112	18.88%	chr2	C	T	rs121913500	IDH1	4	R132H	missense_variant	male
33792731	37.02%	chr19	G	GGCGGGT	rs762459325	CEBPA	1	P232HPP	inframe_insertion	female
106157960	2.27%	chr4	G	A	None	TET2	3	W954*	stop_gained	male
25470585	4.61%	chr2	A	C	rs751916447	DNMT3A	8	W297G	missense_variant	male

3

4 **CHIP mutations identified among patients who underwent heart surgery**

POS	VAF	chrom	ref	alt	rs_ids	gene	exon	aa_change	impact	sex
106164079	3.19%	chr4	A	T	None	TET2	5	K1197*	stop_gained	male
25467188	3.09%	chr2	C	T	None	DNMT3A	15	V563M	missense_variant	male
25463286	4.06%	chr2	C	A	None	DNMT3A	19	R736L	missense_variant	female
25463308	4.98%	chr2	G	A	rs200018028	DNMT3A	19	R729W	missense_variant	male

5

6 POS, position; VAF, variant allele frequency; chrom, chromosome; ref, reference nucleotide; alt, alternative nucleotide; rs_ids, Single Nucleotide Polymorphism

7 identification number; aa_change, change of amino acid.

Table S4. Digestion mixes for human CV tissues

Digestion mixes for human aortic plaque

Digestion mix I 1 mL for 200 mg tissue Incubate at 37 °C, 300 rpm for 70 min		
Reagents	Amount	Supplier
PBS with Ca/Mg ²⁺	905.3 mL	ThermoFisher
Collagenase I (675 U/mL)	67.5 mL	Sigma-Aldrich
Collagenase XI (187,5 U/mL)	15 mL	Sigma-Aldrich
Hyaluronidase (90 U/mL)	7.5 mL	Sigma-Aldrich
Dnase I (90 U/mL)	4.7 mL	Sigma-Aldrich

Digestion mix II 1 mL for 200 mg tissue Incubate at 37 °C, 300 rpm for 8 min		
Reagents	Amount	Supplier
PBS with Ca/Mg ²⁺	952.5 mL	ThermoFisher
Collagenase II (400 U/mL)	40 mL	Sigma-Aldrich
Hyaluronidase (90 U/mL)	7.5 mL	Sigma-Aldrich

Digestion mixes for human heart tissue

Digestion mix 1 mL for 200 mg tissue Incubate at 37 °C, 300 rpm for 30 min		
Reagents	Amount	Supplier
DMEM	953.3 mL	ThermoFisher
Collagenase I (675 U/mL)	38.7 mL	Sigma-Aldrich
Hyaluronidase (90 U/mL)	3 mL	Sigma-Aldrich
Dnase I (90 U/mL)	5 mL	Sigma-Aldrich

16 **Table S5. FACS panels for human CV tissues and blood**
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FACS panel for human aortic plaque			
Target	Fluorochrome	Manufacturer	Dilution
Viability dye	APC-Cy7	ThermoFisher	1:1000
CD45	PacBlue	BioLegend	1:300
CD3	PE-ef610	ThermoFisher	1:300
CD19	BV605	BioLegend	1:300
CD14	BV711	BioLegend	1:300
CD64	FITC	BioLegend	1:300
MERTK	PE	BioLegend	1:300

FACS panel for human blood (patients undergoing carotid endarterectomy)			
Target	Fluorochrome	Manufacturer	Dilution
CD45	PacBlue	BioLegend	1:300
CD3	PE-ef610	BioLegend	1:300
CD4	PerCP-Cy5.5	BioLegend	1:300
CD8	PE-Cy5	BioLegend	1:300
CD19	BV605	BioLegend	1:300
CD14	BV711	BioLegend	1:300

FACS panel for human heart			
Target	Fluorochrome	Manufacturer	Dilution
Viability dye	APC-Cy7	ThermoFisher	1:1000
CD45	PacBlue	BioLegend	1:300
CD14	PE	BioLegend	1:300
CD64	FITC	BioLegend	1:300
CCR2	APC	BioLegend	1:300
HLADR	PE-Cy7	BioLegend	1:300

FACS panel for human blood (patients undergoing heart surgery)			
Target	Fluorochrome	Manufacturer	Dilution
CD45	PacBlue	BioLegend	1:300
CD3	APC	BioLegend	1:300
CD11b	APC-Cy7	BioLegend	1:300
HLA-DR	PE-Cy7	BioLegend	1:300
CD15	PerCP-Cy5.5	BioLegend	1:300
CD16	FITC	BioLegend	1:300
CD14	PE	BioLegend	1:300

Table S6. GO pathways comprising genes for the calculation of module scores

Module scores	GO pathways	Number of genes
Inflammatory response	GO:0050729 positive regulation of inflammatory response	167
Phagocytosis	GO:0050766 positive regulation of phagocytosis	94
Stimuli recognition	GO:0062208 positive regulation of pattern recognition receptor signaling pathway	69
Chemotaxis	GO:0002690 positive regulation of leukocyte chemotaxis	112
Antigen presentation	GO:0019882 antigen processing and presentation	135
Positive cell cycle regulation	GO:0045787 positive regulation of cell cycle	367
Canonical glycolysis	GO:0061621 canonical glycolysis	17
Fatty acid metabolism	GO:0045923 positive regulation of fatty acid metabolic process	50
Glutamine metabolism	GO:0006541 glutamine metabolic process	22

Major Resources Table

In order to allow validation and replication of experiments, all essential research materials listed in the Methods should be included in the Major Resources Table below. Authors are encouraged to use public repositories for protocols, data, code, and other materials and provide persistent identifiers and/or links to repositories when available. Authors may add or delete rows as needed.

Antibodies

Target antigen	Vendor or Source	Catalog #	Working concentration	Persistent ID / URL
Viability dye (APC-Cy7)	ThermoFisher	65-0865-14	1:1000	https://reurl.cc/QednpM
CD45 (PacBlue)	BioLegend	368540	1:300	https://www.biolegend.com/fr-lu/products/pacific-blue-anti-human-cd45-antibody-14908?GroupID=BLG14850
CD3 (PE-ef610)	ThermoFisher	61-0037-42	1:300	https://reurl.cc/zl8g86
CD19 (BV605)	BioLegend	302244	1:300	https://www.biolegend.com/fr-lu/products/brilliant-violet-605-anti-human-cd19-antibody-8483
CD14 (BV711)	BioLegend	367140	1:300	https://www.biolegend.com/fr-lu/products/brilliant-violet-711-anti-human-cd14-antibody-17966
CD64 (FITC)	BioLegend	399506	1:300	https://www.biolegend.com/fr-lu/products/fits-anti-human-cd64-antibody-19189
MERTK (PE)	BioLegend	367608	1:300	https://www.biolegend.com/fr-lu/products/pe-anti-human-mertk-antibody-12249
CD4 (PerCP-Cy5.5)	BioLegend	300530	1:300	https://www.biolegend.com/fr-lu/products/percp-cyanine5-5-anti-human-cd4-antibody-4216
CD8 (PE-Cy5)	BioLegend	344770	1:300	https://www.biolegend.com/fr-lu/products/pe-cyanine5-anti-human-cd8-antibody-21385
CD14 (PE)	BioLegend	325606	1:300	https://www.biolegend.com/fr-lu/products/pe-anti-human-cd14-antibody-3952
CCR2 (APC)	BioLegend	357208	1:300	https://www.biolegend.com/fr-lu/products/apc-anti-human-cd192-ccr2-antibody-8652
HLADR (PE-Cy7)	BioLegend	260158	1:300	https://www.biolegend.com/fr-lu/products/gmp-pecyanine7-anti-human-hla-dr-22369
CD3 (APC)	BioLegend	300312	1:300	https://www.biolegend.com/fr-lu/products/apc-anti-human-cd3-antibody-749
CD11b (APC-Cy7)	BioLegend	301342	1:300	https://www.biolegend.com/fr-lu/products/apc-cyanine7-anti-human-cd11b-antibody-9611
CD15 (PerCP-Cy5.5)	BioLegend	301922	1:300	https://www.biolegend.com/fr-lu/products/percp-cyanine5-5-anti-human-cd15-ssca-1-antibody-17072
CD16 (FITC)	BioLegend	302006	1:300	https://www.biolegend.com/fr-lu/products/fits-anti-human-cd16-antibody-567

Data & Code Availability

Description	Source / Repository	Persistent ID / URL
RNA-seq data of monocytes and tissue macrophages	GSE226642/ GEO	https://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE226642