

Clonal hematopoiesis, cardiovascular events and treatment benefit in 63,700 individuals from five TIMI randomized trials

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Clonal hematopoiesis of indeterminate potential (CHIP) has been associated with an increased risk of cardiovascular (CV) disease in the general population. Currently, it is unclear whether this association is observed in large clinical trial cohorts with a high burden of existing CV disease or whether CV therapies can mitigate CHIP-associated CV risk. To address these questions, we studied 63,700 patients from five randomized trials that tested established therapies for CV disease, including treatments targeting the proteins PCSK9, SGLT2, P2Y12 and FXa. During a median follow-up of 2.5 years, 7,453 patients had at least one CV event (CV death, myocardial infarction (MI), ischemic stroke or coronary revascularization). The adjusted hazard ratio (aHR) for CV events for CHIP+ patients was 1.07 (95% CI: 0.99–1.16, $P = 0.08$), with consistent risk estimates across each component of CV risk. Significant heterogeneity in the risk of MI was observed, such that CHIP+ patients had a 30% increased risk of first MI (aHR = 1.31 (1.05–1.64), $P = 0.02$) but no increased risk of recurrent MI (aHR = 0.94 (0.79–1.13), $P_{int} = 0.008$), as compared to CHIP– patients. Moreover, no significant heterogeneity in treatment effect between individuals with and without CHIP was observed for any of the therapies studied in the five trials. These results indicate that in clinical trial populations, CHIP is associated with incident but not recurrent coronary events and that the presence of CHIP does not appear to identify patients who will derive greater benefit from commonly used CV therapies.

Clonal hematopoiesis of indeterminate potential (CHIP) is a common age-related process characterized by the clonal expansion of blood stem cells due to the acquisition of a cancer driver mutation¹. CHIP has been associated with increased risk of cardiovascular (CV) disease, prompting interest in CHIP for clinical trial enrichment and as a drug target^{2–6}. However, CHIP has largely been evaluated in population cohorts^{7–10}, which depend on International Classification of Diseases (ICD) billing codes for the diagnosis of complex cardiac conditions. Clinical trial data have the benefit of carefully defined populations enriched for CV events, rigorous follow-up, formally adjudicated events and randomized treatment assignments.

We performed whole-exome sequencing in five international randomized controlled CV outcomes trials conducted by the TIMI Study Group, providing a unique opportunity to advance our understanding of CHIP in this context. In this analysis, we set out to address the following three outstanding questions in the field of CHIP and CV disease: (1) is the presence of CHIP associated with major CV events and CV deaths in well-characterized clinical trial populations? (2) Does CHIP predict recurrent coronary events to a similar degree as incident events? (3) Do PCSK9 inhibitors, SGLT2 inhibitors, P2Y12 inhibitors or factor Xa inhibitors have greater efficacy in patients with CHIP mutations?

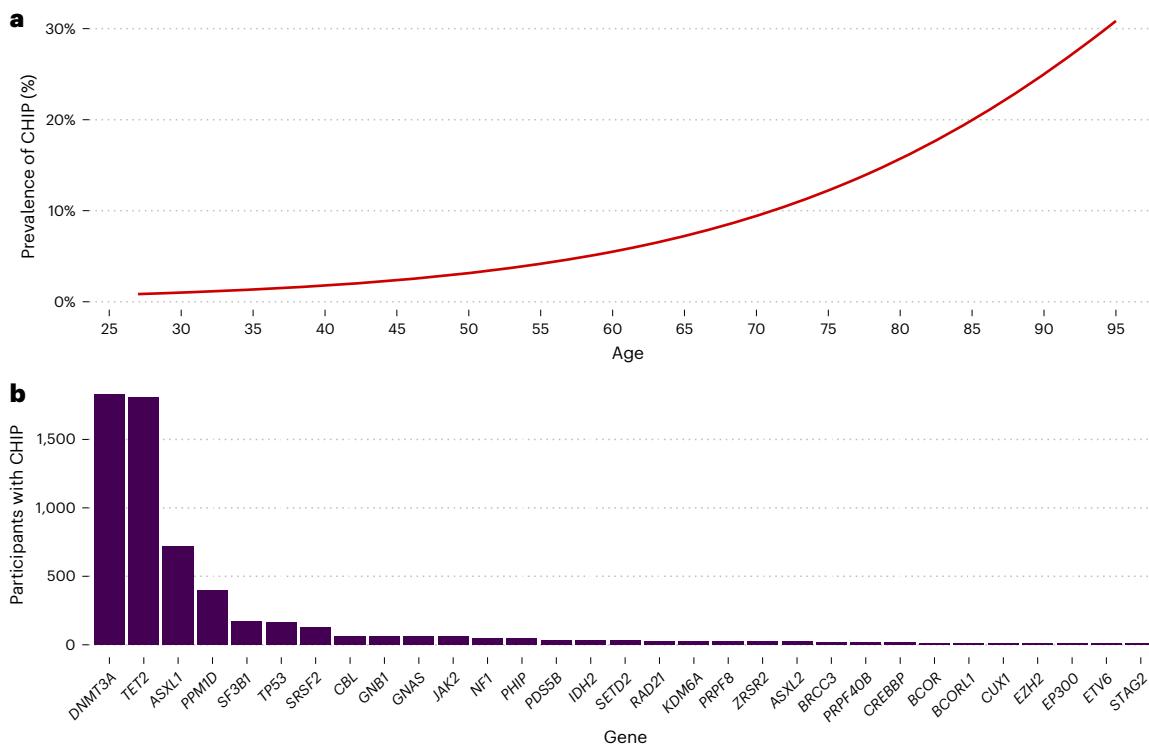


Fig. 1 | Distribution of CHIP over the lifetime and by gene. **a**, The prevalence of CHIP increases with age. **b**, Frequency of CHIP mutations across 31 genes.

Results

A total of 63,700 patients were included in the analysis. The median age was 65 years (interquartile range = 59–72). The cohort consisted of 31% female participants, 56% with diabetes and 74% with baseline atherosclerotic CV disease (ASCVD; including 53% prior myocardial infarction (MI), 11% prior stroke and 8% peripheral artery disease (PAD)). Baseline characteristics for each trial are shown in Extended Data Table 1. CHIP was present in 8.2% of the population and strongly associated with age (Fig. 1a), ranging from <10% in individuals under 70 years to above 20% in those older than 85 years. The median variant allele frequency (VAF) was 13%. The most commonly involved CHIP mutations were DNMT3A ($n = 1,730$, 33%), TET2 ($n = 1,712$, 33%) and ASXL1 ($n = 687$, 13%; Fig. 1b). CHIP+ patients were significantly older (70 versus 65 years old, $P < 0.0001$) and more likely to have a history of cancer (7% versus 5%, $P < 0.0001$), heart failure (28% versus 25%, $P = 0.0002$) and chronic kidney disease (CKD; 22% versus 17%, $p < 0.0001$; Table 1). A total of 7,453 patients had at least one major CV event during follow-up, including 2,201 CV deaths, 2,458 with MI, 1,362 with ischemic stroke and 3,732 with coronary revascularization during the 2.5-year median follow-up.

CHIP and major CV events

The rate of major CV events at 2.5 years was 12.7% in those with a CHIP mutation versus 10.4% in those without (unadjusted HR = 1.18 (1.09–1.27), $P < 0.0001$). However, after adjustment for age, sex, genetic principal components and CV risk factors and comorbidities, the association was attenuated and no longer significant (adjusted hazard ratio (aHR) = 1.07 (0.99–1.16), $P = 0.08$; Fig. 2a and Extended Data Table 2). Consistent risk estimates were seen across each of the MACE components, with no significant associations observed for CV death, MI, ischemic stroke or coronary revascularization (Fig. 2a). Sensitivity analyses removing one trial at a time (Extended Data Table 3) and using Fine–Gray model to address competing risks (Extended Data Table 4a) were consistent, even when considered by trial

(Extended Data Table 4b). Results were also consistent among statin users and non-statin users (Extended Data Table 5).

Neither large CHIP (VAF > 10%) nor multiple CHIP clones (two or more CHIP mutations $\geq 2\%$) provided additional risk stratification beyond that of the conventional VAF threshold of $\geq 2\%$ (MACE rates of 12.8% and 12.9%, respectively, compared with 12.7%). However, the presence of multiple large CHIP clones (two or more CHIP mutations at $> 10\%$) was associated with the greatest absolute risk, with a MACE rate of 15.9%. This risk was most prominent among individuals without a history of MI (Fig. 3), where a nonsignificant 25% increased risk compared to CHIP– patients was observed (aHR = 1.25 (0.84–1.86), $n = 147$).

The association between specific CHIP driver genes and MACE with and without stratification by history of MI is presented in Extended Data Fig. 1. No individual CHIP genes were significantly associated with MACE. Spliceosome genes (SF3B1, SRSF2 or U2AF1) had a nonsignificant 18% increased risk (aHR = 1.18 (0.87–1.60)) and DNMT3A carried a nonsignificant 11% increased risk (aHR = 1.11 (0.97–1.26)). CHIP mutations in TET2 or ASXL1 did not appear to have any association with MACE (aHR = 1.00 (0.87–1.14) and 1.00 (0.81–1.23), respectively). JAK2 variants made up only 1% of CHIP driver gene mutations and did not have any association with MACE (aHR = 0.99 (0.44–2.21)). The stratified results were consistent among those with and without prior MI, with the exception of PPMID for which CHIP appeared to be associated with greater risk in those with a history of MI (aHR = 1.29 (0.93–1.81)) compared to those without prior MI (aHR = 0.81 (0.50–1.31)). Given the relationship between inflammation and CHIP, IL6R Asp358Ala genotypes (rs2228145) were evaluated in relation to the risk of CHIP and TET2 in particular⁴. A sensitivity analysis stratifying by Asp358Ala genotypes showed no heterogeneity of effect of CHIP (Extended Data Fig. 2a) while a small TET2 effect was observed in the two minor alleles group (Extended Data Fig. 2b; aHR = 1.17 (0.81–1.69), $P = 0.4$). We also looked at the CV risk associated with TET2 mutations stratified by statin use (Extended Data Table 5) and found a numerically greater risk from TET2+ among those not on statin (aHR = 1.06 (0.80–1.40)) compared

Table 1 | Baseline characteristics of patients as stratified by CHIP

Characteristic	CHIP-, n=58,470	CHIP+, n=5,230	P value
Age \pm s.d.	65.1 \pm 8.9	69.6 \pm 8.4	<0.0001
Sex: male (%)	40,545 (69.3)	3,570 (68.3)	0.1072
Ethnicity ^a (%)	Asian: 3,313 (5.7) Black: 1,946 (3.3) Other: 2,159 (3.7) White: 51,052 (87.3)	Asian: 220 (4.2) Black: 129 (2.5) Other: 134 (2.6) White: 4,747 (90.8)	<0.0001
BMI (kg m^{-2}) \pm s.d.	30.4 \pm 5.7	29.8 \pm 5.4	<0.0001
ASCVD (%)	43,430 (74.3)	3,882 (74.2)	0.9477
Prior MI (%)	31,006 (53)	2,649 (50.7)	0.0010
Prior stroke (%)	6,443 (11)	630 (12)	0.0250
History of PAD (%)	4,893 (8.4)	511 (9.8)	0.0005
History of hypertension (%)	49,499 (84.7)	4,491 (85.9)	0.0205
History of CHF (%)	14,921 (25.5)	1,458 (27.9)	0.0002
History of diabetes (%)	32,862 (56.2)	2,824 (54)	0.0022
History of hypercholesterolemia (%)	36,811 (63)	3,262 (62.4)	0.4089
History of CKD (%)	9,836 (16.8)	1,154 (22.1)	<0.0001
History of AFIB (%)	15,516 (26.5)	1,674 (32)	<0.0001
History of CAD (%)	37,972 (64.9)	3,392 (64.9)	0.9125
History of coronary revascularization (%)	28,531 (48.8)	2,505 (47.9)	0.2179
History of cancer (%)	2,776 (4.7)	360 (6.9)	<0.0001
Current smoker (%)	9,904 (16.9)	747 (14.3)	<0.0001
Systolic blood pressure (mmHg) \pm s.d.	133.2 \pm 16	133.8 \pm 16.1	0.0318
eGFR ($\text{ml min}^{-1} \text{1.73 m}^{-2}$) \pm s.d.	75.1 \pm 20.5	71 \pm 20.5	<0.0001
Hemoglobin (g dL^{-1}) \pm s.d.	14.1 \pm 1.4	13.9 \pm 1.4	<0.0001
White blood count ^b (count per μl) \pm s.d.	7.2 \pm 1.9	7.3 \pm 2.1	0.2984
Platelets (10^3 per μl) \pm s.d.	217.5 \pm 58	216.4 \pm 71	<0.0001
Lipid-lowering therapy (%)	46,992 (80.4)	4,156 (79.5)	0.1193
Antiplatelet therapy (%)	33,276 (56.9)	2,874 (55)	0.0064
Anticoagulant therapy (%)	15,045 (25.7)	1,611 (30.8)	<0.0001
IL6R Asp358Ala rs2228145 ^a (%)	A-A: 19,393 (39.8) C-A: 22,579 (46.3) C-C: 6,815 (14)	A-A: 1,776 (39.4) C-A: 2,108 (46.7) C-C: 627 (13.9)	0.8418

^aChi-square test evaluating whether the proportion of the four ethnic groups or three IL6R genotypes is different between CHIP- and CHIP+ cohorts. ^bData is available only in the PEGASUS trial. CHF, congestive heart failure. Mean (s.d.) values are reported for numerical values; otherwise percentages are reported.

to no effect seen in those on statins (aHR = 0.99 (0.84–1.16)). When the risk of TET2 mutation was assessed as a function of low-density lipoprotein cholesterol (LDL-C) levels (Extended Data Fig. 3), no interaction between hyperlipidemia and TET2-associated CV risk was observed.

CHIP and death

There were 3,650 deaths in the five trials over the median follow-up period of 2.5 years. Of these, 35% ($n = 1,279$) were non-CV deaths and 60% ($n = 2,201$) were CV deaths, including 23% ($n = 855$) coronary heart disease deaths. CHIP+ was associated with an 11% increase in all-cause mortality (aHR = 1.11 (1.00–1.23), $P = 0.05$). This was strengthened by a 17% increased risk of non-CV deaths (aHR = 1.17 (0.99–1.39), $P = 0.07$), particularly an increase in cancer-related deaths (aHR = 1.20 (0.95–1.53), $P = 0.13$). Conversely, CHIP was not significantly associated with CV deaths (aHR = 1.05 (0.91–1.20), $P = 0.5$) or coronary heart disease deaths (aHR = 1.04 (0.82–1.31), $P = 0.7$; Fig. 2b).

CHIP and incident versus recurrent coronary events

CHIP+ was not significantly associated with MI (aHR = 1.06 (0.93–1.22)); however, when stratified by prior MI, there was significant heterogeneity observed. CHIP+ strongly associated with first MI in individuals with no prior MI (aHR = 1.31 (1.05–1.64), $P = 0.02$), but not with recurrent MI in those with prior MI (aHR = 0.94 (0.79–1.13), $P_{\text{interaction}} (P_{\text{int}}) = 0.008$; Fig. 4a). The same pattern was seen for first (aHR = 1.28 (1.05–1.57)) and recurrent coronary revascularization (aHR = 1.02 (0.88–1.17), $P_{\text{int}} = 0.02$; Fig. 4b). No heterogeneity was observed for first versus recurrent ischemic stroke.

CHIP and CV therapies

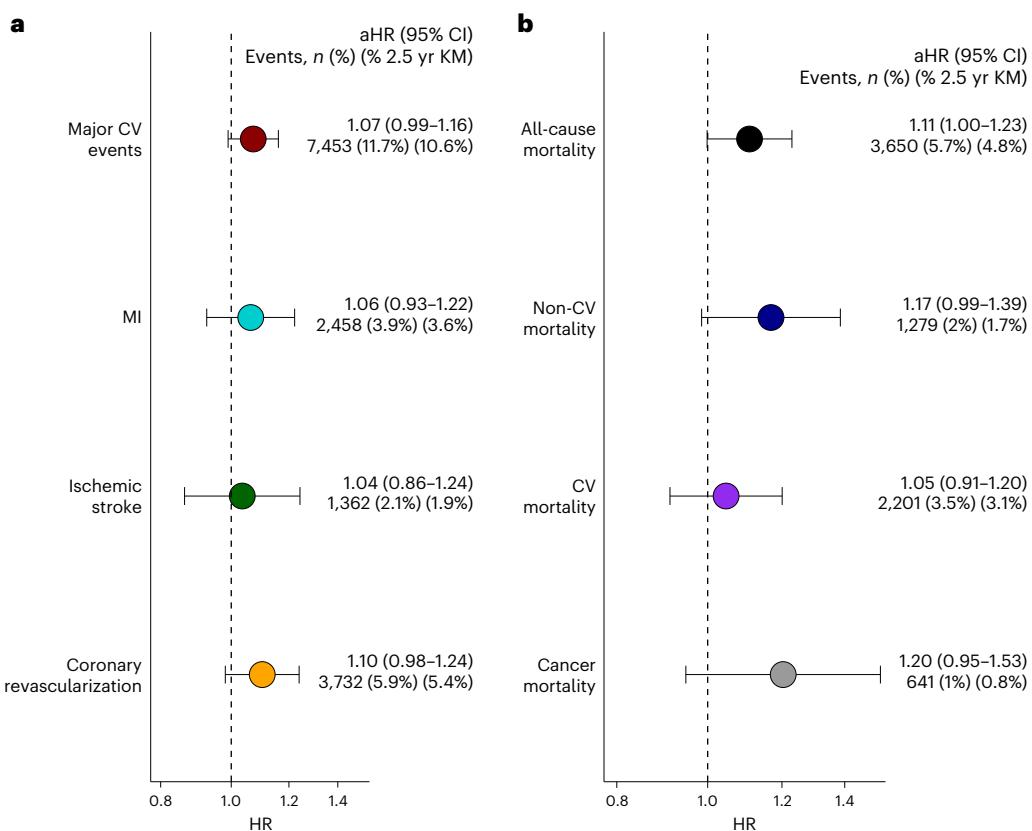
Four of the randomized therapies tested in the five TIMI trials had HRs for MACE <1.0 (trial-wide relative risk reduction (RRR) for MACE of 15% in FOURIER, 16% in PEGASUS, 12% in high-dose regimen of ENGAGE AF and 7% in DECLARE), allowing for CHIP by treatment interaction testing. The absolute risk reductions (ARR) and RRR for MACE in those with and without CHIP are presented in Table 2. There was no statistically significant heterogeneity in the reduction of MACE between CHIP+ patients compared to those without CHIP for the PCSK9 inhibitor evolocumab, SGLT2 inhibitor dapagliflozin, P2Y12 inhibitor ticagrelor and direct factor Xa inhibitor edoxaban. If anything, in FOURIER, there was a trend toward greater benefit from evolocumab in patients without CHIP, rather than with CHIP (HR = 0.80 (0.72–0.88) versus 1.10 (0.79–1.55), $P_{\text{int}} = 0.1$).

Discussion

With over 63,000 patients and approximately 10,000 major CV events across five CV outcomes trials, this study represents the largest analysis of CHIP in the CV clinical trial setting to date. Strengths of this cohort include a broad population of at-risk individuals with and without a history of major CV events, adjudicated cardiac events and cause of death and placebo-controlled randomized treatment assignments. Using this unique dataset, we were able to address multiple unanswered questions in the field and highlight five key findings that contribute to our understanding of the relationship between CHIP and CV disease.

First, the association between CHIP and adjudicated CV events in a broad population of participants with or at-risk for ASCVD is modest, with consistent findings across the different outcomes of CV death, MI, ischemic stroke and coronary revascularization. While the attenuated risk of CHIP for CV events has previously been reported¹¹, our results are in contrast with the majority of prior case-control and population-based epidemiology studies which have shown a more robust relationship between CHIP and prevalence of coronary artery disease (CAD)^{8–10}. Notably, a recent analysis demonstrated no association between CHIP and CVD, but those authors employed a new CHIP definition limiting the ability to directly compare with our results using a traditional approach¹². There are several potential explanations for this, including differences in patient populations, use of background therapies and how composite endpoints were defined and determined. In this CV clinical trial population, the majority of patients had ASCVD, a key difference from most prior studies which have been in unselected cohorts. Even those who did not have overt ASCVD in this analysis had a high burden of risk factors, including diabetes, and therefore a higher prevalence of subclinical disease. Our data suggest CHIP has a stronger relationship in a primary prevention population without a clear association in patients with prior MI. A second possibility is that the majority of patients in these trials were heavily treated with intensive medical management (including high-dose statins) consistent with contemporary practice. Earlier studies included patients who were not as intensively treated, mainly because they were healthier populations, but also due to changes in practice over time.

Supporting the hypothesis that CHIP-related risk may differ by population, our second key finding is that CHIP had the strongest association with first MI and first coronary revascularization but did

**Fig. 2 | Association between CHIP and major CV events and cause of death.**

a, b, aHRs and 95% CI, number of events and Kaplan–Meier (KM) rates at 2.5 years are shown for each endpoint—major CV events, 1.07 (0.99–1.16; $P = 0.08$); CV deaths, 1.05 (0.91–1.20; $P = 0.52$); MI, 1.06 (0.93–1.22; $P = 0.39$); ischemic stroke, 1.04 (0.86–1.24; $P = 0.71$) and coronary revascularization, 1.10 (0.98–1.24; $P = 0.1$; **a**), and for all-cause mortality, 1.11 (1.00–1.23; $P = 0.05$); non-CV deaths, 1.17

(0.99–1.39; $P = 0.07$); CV deaths, 1.05 (0.91–1.20; $P = 0.5$) and cancer deaths, 1.20 (0.95–1.53; $P = 0.13$; **b**). Cox proportional hazards model was adjusted for age, age², sex, BMI, genetic principal components 1–10, lipid-lowering therapy, blood pressure, diabetes, hyperlipidemia, eGFR, prior MI, heart failure, AFIB, peripheral artery disease, prior stroke, history of cancer, trial and smoking. Two-sided P values were obtained via Wald test.

not appear to have any association with recurrent MI or recurrent revascularization. This stronger association for first events is consistent with the strong associations seen in prior studies that were primarily in relatively healthy, primary prevention populations. Additionally, a recent analysis found that hyperlipidemia acts synergistically with *TET2* deficiency to activate the *NLRP3* inflammasome, which could contribute to why CHIP carries greater risk in primary prevention individuals who are not aggressively treated with lipid-lowering therapy¹³. In our study, we observed that *TET2* mutations were associated with a nominally higher risk of MACE among patients who were not on statins compared to no association with risk among statin users. Heterogeneity in CHIP-associated CV risk has important implications for its potential role in clinical trial enrichment, where it appears to be better suited for use in primary prevention CAD trials rather than secondary prevention.

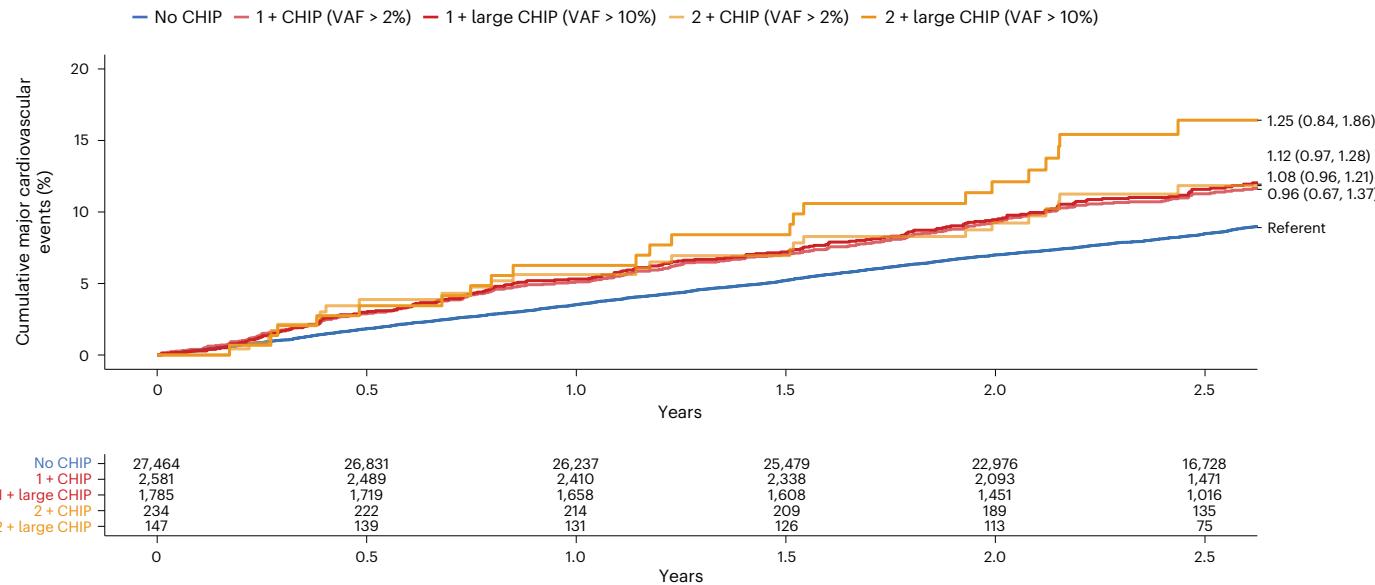
A third key observation in our study relates to CHIP's association with mortality. While the relationship between CHIP and all-cause mortality is established, our adjudicated cause-specific mortality suggests that this increase is driven by an association with non-CV deaths, largely from cancer-associated mortality, and not CV deaths. This is an important consideration as prior CHIP studies have often included all-cause mortality in their 'ASCVD' composite endpoint^{4,10}, and as a result, non-CV deaths such as those from cancer may be contributing to the larger associations between CHIP and ASCVD found in those studies. When only CV deaths are included in the ASCVD composite, a more modest relationship between CHIP and ASCVD events has been seen¹¹.

Fourth, the large number of CHIP patients and events in this clinical trial cohort allowed for a deeper exploration of specific CHIP driver genes of interest. Prior literature has supported *TET2* as having the most

promise for its association with atherosclerotic CV disease^{11,14–16}. However, while *TET2* was one of the most common CHIP mutations in our study, there was no evidence that it carried any added risk of CV events, with a neutral effect. One possible explanation could be the previously described interaction between hyperlipidemia and *TET2*. This suggests that the risk associated with *TET2* requires hyperlipidemia, which was rare in this higher risk, well-treated clinical trial population¹³. However, we did not observe such an interaction in this clinical trial cohort. Similarly, *JAK2* is often considered a likely culprit for increased CV events given its predisposition for thrombosis. While *JAK2* mutations were much less common than *TET2*, there was again no evidence of increased risk in this trial cohort. Conversely, *DNMT3A* gene mutations were the most common (present in 1/3 of the CHIP+ patients), and carried the greatest risk of CV events, with 11% increased risk. Given the prevalence of these mutations and their positive association with events, our data would suggest *DNMT3A* may be a target for further study.

Finally, we were able to assess the treatment benefit of four commonly used CV therapies from four distinct drug classes among those with and without CHIP. This included a lipid-lowering therapy (the PCSK9 inhibitor evolocumab), a cardiometabolic/antiglycemic agent (the SGLT2 inhibitor dapagliflozin), an antiplatelet therapy (the P2Y12 inhibitor ticagrelor) and an anticoagulant (the direct factor Xa inhibitor edoxaban). The benefit of these therapies was observed in both individuals with and without CHIP. While no CHIP by treatment interaction was observed, this represents a clinically important set of analyses as it demonstrates that patients with CHIP will benefit similarly from guideline-based clinical care with respect to lipid-lowering therapy, cardiometabolic, anticoagulant and antiplatelet therapy—four

a No history of MI



b History of MI

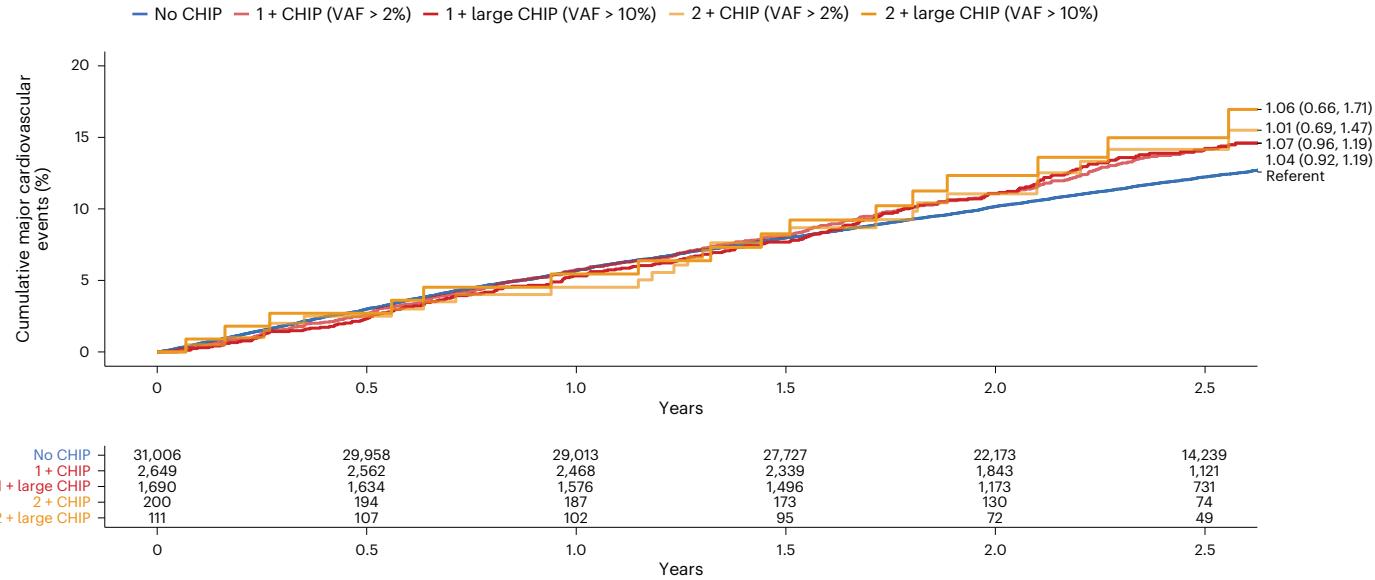


Fig. 3 | Association between CHIP burden and major CV events in patients with and without prior MI. a, b, Kaplan–Meier curves are shown for MACE over 2.5 years in individuals without (a) or with (b) a prior MI stratified by CHIP burden. aHRs and 95% CI are reported adjusted for age, age², sex, BMI, genetic

principal components 1–10, lipid-lowering therapy, blood pressure, diabetes, hyperlipidemia, eGFR, prior MI, heart failure, AFIB, peripheral artery disease, prior stroke, history of cancer, trial and smoking. Two-sided *P* values were obtained via Wald test.

cornerstones of our clinical armamentarium. The search for targeted CV therapies that modify CHIP risk specifically is of great interest. Notably, we did not have an anti-inflammatory drug to test among these trials, which may have the most promise given CHIP's mechanism of action¹⁶.

This study has several limitations. First, while these large international trials have a global footprint, they are still predominantly non-Hispanic white individuals. However, these results do include data from >8,000 nonwhite patients. Second, while these five trials represent a broad range of CV conditions and risk, the trial inclusion criteria do create distinct cohorts from one another. For these reasons, we have adjusted by trial and added N-1 trial and per-trial analyses to Extended Data Tables 3 and 4b. Third, since these data come from CV outcomes trials, the follow-up time is shorter than in prior studies in prospective observational cohorts. Therefore, the impact of CHIP

beyond 3 years cannot be evaluated. Fourth, given CHIP's significant association with older age and cancer, there is a possibility of competing risks. We evaluated this using a Fine–Gray model to account for competing risks and a sensitivity analysis excluding individuals with a history of cancer and found that the results were consistent. Fifth, unlike germline mutations where the genetic variation clearly precedes disease, the temporal relationship between somatic variation and ASCVD is more complicated as ASCVD may promote CHIP¹⁶. In an attempt to address this, we evaluated events that occurred after somatic mutations were identified; however, this does not eliminate the possibility that the underlying ASCVD did not precede the CHIP mutation¹⁷. Sixth, while a median coverage of 60× and a minimum threshold of 5 alternative reads to report mutations is robust, it is still possible that we have missed some low VAF mutations that may be identified with deeper sequencing. However, most published CHIP studies have

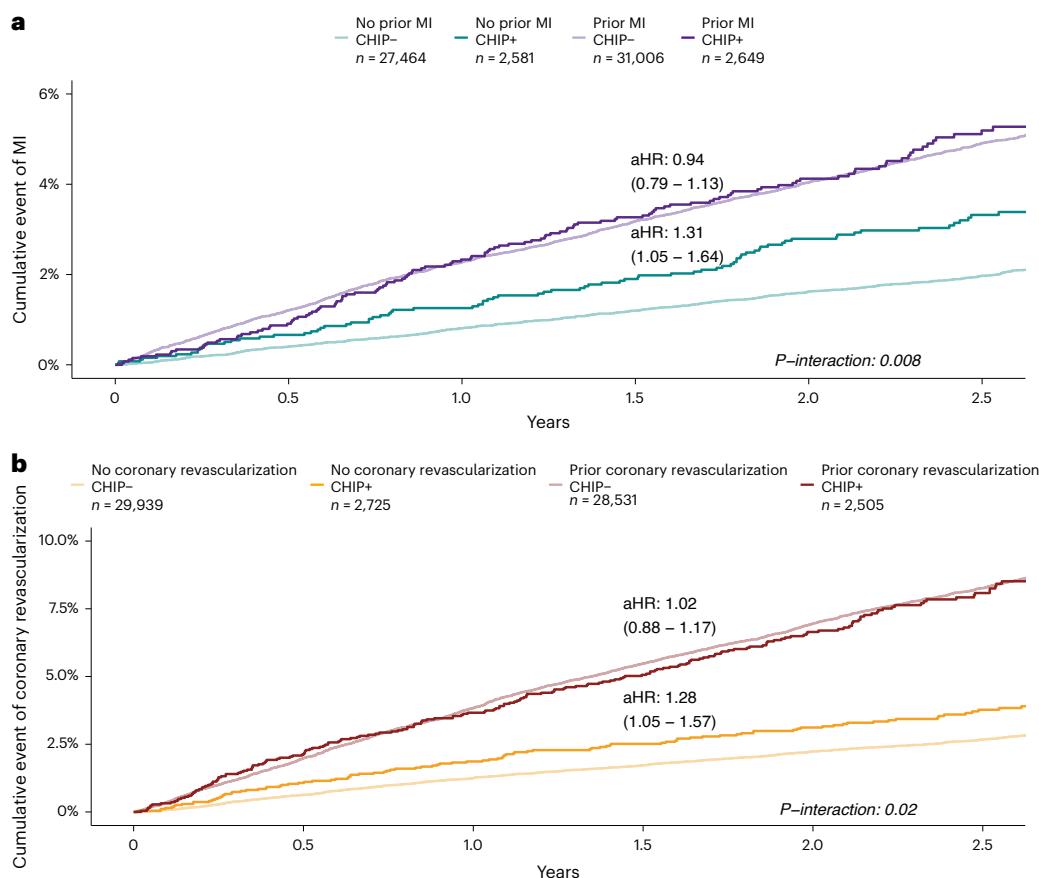


Fig. 4 | Associations between CHIP and the cumulative risk of first versus recurrent MI and coronary revascularization. **a,b,** The association between CHIP and MI (**a**) and coronary revascularization (**b**) is shown for individuals with or without a prior MI. Cox proportional hazards model was adjusted for age, age², sex, BMI, genetic principal components 1–10, lipid-lowering therapy, blood pressure, diabetes, hyperlipidemia, eGFR, prior MI (**b** only, as **a** shows data

already stratified by prior MI), heart failure, AFIB, peripheral artery disease, prior stroke, history of cancer, trial and smoking. HRs and 95% CI are reported adjusted for age, age², sex, BMI, genetic principal components 1–10, lipid-lowering therapy, blood pressure, diabetes, hyperlipidemia, eGFR, prior MI, heart failure, AFIB, peripheral artery disease, prior stroke, history of cancer, trial and smoking. Two-sided P values were obtained via Wald test.

Table 2 | RRR for major CV events with PCSK9 inhibition (evolocumab), SGLT2 inhibition (dapagliflozin), P2Y12 inhibition (ticagrelor) and direct factor Xa inhibition (edoxaban) in the overall population in the trials testing these therapies and across CHIP subgroups

CHIP status	Tx arm	n _{events} /n _{patients}	Event rate (%)	aHR (95% CI)	P _{int} (HR)	ARR (95% CI)	P _{int} (ARR)
Evolocumab (PCSK9i)							
CHIP+	Placebo	73/690	11.1%	1.1 (0.79,1.55)		0.15% (-3.43,3.73)	
	Evolocumab	70/640	10.9%		0.11		0.24
CHIP-	Placebo	846/7,409	12.3%	0.8 (0.72,0.88)		2.44% (1.34,3.54)	
	Evolocumab	688/7,472	9.8%				
Dapagliflozin (SGLT2i)							
CHIP+	Placebo	75/455	11.8%	0.85 (0.61,1.19)		0.04% (-4.19,4.27)	
	Dapagliflozin	69/458	11.8%		0.86		0.68
CHIP-	Placebo	838/5,892	9.3%	0.92 (0.84,1.02)		0.95% (-0.08,1.98)	
	Dapagliflozin	782/5,880	8.3%				
Ticagrelor 60 or 90 mg dose (P2Y12i)							
CHIP+	Placebo	50/347	14.8%	0.72 (0.5,1.05)		3.82% (-0.93,8.57)	
	Ticagrelor (pooled)	71/634	10.9%		0.27		0.29
CHIP-	Placebo	438/3,930	10.7%	0.92 (0.82,1.04)		1.16% (-0.05,2.37)	
	Ticagrelor (pooled)	801/7,880	9.6%				
Edoxaban high dose (direct factor Xa inhibitor)							
CHIP+	Placebo	66/446	13.9%	0.97 (0.68,1.40)		2.57% (-1.97,7.1)	
	Edoxaban (high dose)	54/406	11.3%		0.97		0.39
CHIP-	Placebo	523/4,148	10.5%	0.93 (0.83,1.06)		0.21% (-1.12,1.55)	
	Edoxaban (high dose)	486/4,086	10.2%				

PCSK9i, proprotein convertase subtilisin/kexin type 9 inhibitor; Tx, treatment; n_{events}/n_{patients}, number of events/number of patients in each arm; SGLT2i, sodium-glucose transporter 2 inhibitor; P2Y12i, purinergic receptor P2Y G-protein coupled, 12 protein.

similar depth of coverage and since most effects are seen in high VAF CHIP, the impact of missing a proportion of low VAF mutations should be limited. Seventh, it is possible that some patients with CHIP died before they could be enrolled in one of these trials (survivorship bias) which could attenuate the association with CV death. This is not unique to these trials nor CHIP as a risk factor, and established CV risk factors such as diabetes and heart failure still predict CV death in this cohort.

In clinical trial populations, there was a borderline association between CHIP and the risk of CV events. CHIP was associated with incident but not recurrent coronary events, suggesting its use for prognostic enrichment of clinical trials may be more applicable in primary prevention populations. However, patients with CHIP derived similar benefits from commonly used CV therapies compared to the general trial population, limiting its potential to guide specific management strategies with existing therapies.

Online content

Any methods, additional references, Nature Portfolio reporting summaries, source data, extended data, supplementary information, acknowledgements, peer review information; details of author contributions and competing interests; and statements of data and code availability are available at <https://doi.org/10.1038/s41591-024-03188-z>.

References

- Niroula, A. et al. Distinction of lymphoid and myeloid clonal hematopoiesis. *Nat. Med.* **27**, 1921–1927 (2021).
- Jaiswal, S. et al. Clonal hematopoiesis and risk of atherosclerotic cardiovascular disease. *N. Engl. J. Med.* **377**, 111–121 (2017).
- Bhattacharya, R. et al. Clonal hematopoiesis is associated with higher risk of stroke. *Stroke* **53**, 788–797 (2022).
- Bick, A. G. et al. Genetic Interleukin 6 signaling deficiency attenuates cardiovascular risk in clonal hematopoiesis. *Circulation* **141**, 124–131 (2020).
- Zekavat, S. M. et al. TP53-mediated clonal hematopoiesis confers increased risk for incident atherosclerotic disease. *Nat. Cardiovasc. Res.* **2**, 144–158 (2023).
- Vlasschaert, C., Heimlich, J. B., Rauh, M. J., Natarajan, P. & Bick, A. G. Interleukin-6 receptor polymorphism attenuates clonal hematopoiesis-mediated coronary artery disease risk among 451,180 individuals in the UK Biobank. *Circulation* **147**, 358–360 (2023).
- Vlasschaert, C. et al. A practical approach to curate clonal hematopoiesis of indeterminate potential in human genetic data sets. *Blood* **141**, 2214–2223 (2023).
- Uddin, M. D. M. et al. Clonal hematopoiesis of indeterminate potential, DNA methylation, and risk for coronary artery disease. *Nat. Commun.* **13**, 5350 (2022).
- Bhattacharya, R. et al. Association of diet quality with prevalence of clonal hematopoiesis and adverse cardiovascular events. *JAMA Cardiol.* **6**, 1069–1077 (2021).
- Gumuser, E. D. et al. Clonal hematopoiesis of indeterminate potential predicts adverse outcomes in patients with atherosclerotic cardiovascular disease. *J. Am. Coll. Cardiol.* **81**, 1996–2009 (2023).
- Kessler, M. D. et al. Common and rare variant associations with clonal haematopoiesis phenotypes. *Nature* **612**, 301–309 (2022).
- Stacey, S. N. et al. Genetics and epidemiology of mutational barcode-defined clonal hematopoiesis. *Nat. Genet.* **55**, 2149–2159 (2023).
- Yalcinkaya, M. et al. BRCC3-mediated NLRP3 deubiquitylation promotes inflammasome activation and atherosclerosis in Tet2 clonal hematopoiesis. *Circulation* **148**, 1764–1777 (2023).
- Fuster, J. J. et al. Clonal hematopoiesis associated with TET2 deficiency accelerates atherosclerosis development in mice. *Science* **355**, 842–847 (2017).
- Liu, W. et al. Blockade of IL-6 signaling alleviates atherosclerosis in Tet2-deficient clonal hematopoiesis. *Nat. Cardiovasc. Res.* **2**, 572–586 (2023).
- Svensson, E. C. et al. TET2-driven clonal hematopoiesis and response to canakinumab: an exploratory analysis of the CANTOS randomized clinical trial. *JAMA Cardiol.* **7**, 521–528 (2022).
- Heyde, A. et al. Increased stem cell proliferation in atherosclerosis accelerates clonal hematopoiesis. *Cell* **184**, 1348–1361 e1322 (2021).

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Methods

Study population

We analyzed individuals who underwent whole-exome sequencing in the following five TIMI trials: ENGAGE AF-TIMI 48 (ref. 18), SAVOR-TIMI 53 (ref. 19), PEGASUS-TIMI 54 (ref. 20), DECLARE-TIMI 58 (ref. 21) and FOURIER (TIMI 59)²². Baseline characteristics for each trial are shown in Extended Data Table 1. Patients were considered CHIP+ if they had at least one CHIP clone, defined as a somatic mutation in a CHIP-associated leukemia driver gene with at least five supporting alternative alleles and with a VAF $\geq 2\%$ (10% in dosage and sensitivity analyses). Individuals with more than one CHIP mutation were also evaluated. This multitrial population had global representation from six continents and included nonwhite ethnicities. Information regarding the ethical approval of each trial is available in the primary trial publications.

Clinical endpoints

Study endpoints included major CV events (CV death, MI, ischemic stroke or coronary revascularization). Additional causes of death including coronary heart deaths, non-CV deaths and cancer deaths were also evaluated. All outcomes were adjudicated by an independent blinded clinical endpoints committee. The median follow-up across the five trials was 2.5 years.

Exome sequencing

Exome sequencing was performed at the Broad Institute for ENGAGE AF, SAVOR, PEGASUS and DECLARE on Illumina HiSeq 2500 using TruSeq Rapid Exome Library prep kit. FOURIER samples were sequenced at deCODE on the Illumina HiSeq 2500. Sequence reads from 64,949 exome samples were aligned to GRCh38 with BWA 0.7.15-r1140 and converted to SAM specification 1.6 CRAM files with GATK 4.1.4.1 using the GATK Exome Best Practices Workflow (<https://github.com/gatk-workflows/gatk4-exome-analysis-pipeline>). A total of 63,700 samples were ultimately retained following a QC pipeline which has been previously described²³. GATK version 4.0.10.1 was used in ENGAGE and SAVOR, and version 4.1.8.0 in DECLARE and PEGASUS. The exomes were sequenced to a median depth of 59 \times across trials.

CHIP calling

We applied a previously validated, widely used approach to detect CHIP in exome sequencing data⁷. Briefly, somatic mutations were detected using the GATK Mutect2 (ref. 24) somatic mutation calling algorithm (v4.2.5.0 for ENGAGE AF, SAVOR, PEGASUS and DECLARE, and v4.1.4.1 for FOURIER) applied to a set of 74 canonical CHIP genes. For each cohort separately, 120 randomly chosen samples were called in tumor-only mode using Mutect2; any sample with a putative CHIP variant was discarded, and those without putative CHIP were used as the panel of normals (PON) for tumor-normal calling of the remainder of the cohort. The PON and the TOPMed Freeze 5 (ref. 25) germline variant resource were used for artifact reduction when calling somatic variation for each of the remaining samples within the cohort with Mutect2. Using the GATK FilterMutectCalls tool, variants were then further filtered using population-scale filtering approaches to remove recurrent artifacts and germline genetic variants. The remaining putative somatic mutations were annotated with ANNOVAR (version 24 October 2019)²⁶ and then further filtered using a combination of sequence-based filtering metrics (total allele depth > 20 , minimum number of alternative reads ≥ 5 , support for a variant on both forward and reverse reads) and the prespecified list of putative CHIP driver mutations. Data analysis was performed using basic statistical regression techniques on R v3.6.

Statistical analysis

Individual patient-level data were pooled from the five clinical trials. Risk of MACE during trial follow-up in the CHIP+ group was compared with the CHIP- group using Cox proportional hazards model

(assumptions met) adjusting for age, age 2 , sex, BMI, genetical principal components 1–10, lipid-lowering therapy, blood pressure, diabetes, hyperlipidemia, smoking, estimated glomerular filtration rate (eGFR), prior MI, heart failure, atrial fibrillation (AFIB), peripheral artery disease, prior stroke, history of cancer and trial. Minimally adjusted models by age and sex were also evaluated. Time-to-event data were used to create Kaplan–Meier curves. Additional stratification was performed in patients with and without prior MI, coronary revascularization or ischemic stroke (in which case these baseline variables were not included in the model). Dosage and sensitivity analyses included testing higher CHIP thresholds, including $>10\%$ CHIP and/or two or more CHIP (a leave-one-out approach), where each trial was removed from the analysis, and a competing risk analysis using the Fine–Gray model. The interactionRCS R package (<https://CRAN.R-project.org/package=interactionRCS>) was used to evaluate the effect of *TET2* CHIP over level of LDL-C using a restricted cubic spline interaction model with three knots at first quartile, median and third quartile. LDL-C values in PEGASUS were not available and were calculated using ApoB levels with the formula $1.28 \times \text{ApoB} - 29$ (ref. 27). All *P* values were two sided and assessed at a threshold of 0.05.

Reporting summary

Further information on research design is available in the Nature Portfolio Reporting Summary linked to this article.

Data availability

Due to contractual agreements with the sponsors of the clinical trials, trial data cannot be made publicly available.

References

18. Giugliano, R. P. et al. Edoxaban versus warfarin in patients with atrial fibrillation. *N. Engl. J. Med.* **369**, 2093–2104 (2013).
19. Scirica, B. M. et al. Saxagliptin and cardiovascular outcomes in patients with type 2 diabetes mellitus. *N. Engl. J. Med.* **369**, 1317–1326 (2013).
20. Bonaca, M. P. et al. Long-term use of ticagrelor in patients with prior myocardial infarction. *N. Engl. J. Med.* **372**, 1791–1800 (2015).
21. Wiviott, S. D. et al. Dapagliflozin and cardiovascular outcomes in type 2 diabetes. *N. Engl. J. Med.* **380**, 347–357 (2019).
22. Sabatine, M. S. et al. Evolocumab and clinical outcomes in patients with cardiovascular disease. *N. Engl. J. Med.* **376**, 1713–1722 (2017).
23. Jurgens, S. J. et al. Sequencing in over 50,000 cases identifies coding and structural variation underlying atrial fibrillation risk. *Circulation* **146**, A13496 (2022).
24. Benjamin, D. et al. Calling somatic SNVs and Indels with Mutect2. Preprint at bioRxiv <https://doi.org/10.1101/861054> (2019).
25. Taliun, D. et al. Sequencing of 53,831 diverse genomes from the NHLBI TOPMed program. *Nature* **590**, 290–299 (2021).
26. Wang, K., Li, M. & Hakonarson, H. ANNOVAR: functional annotation of genetic variants from high-throughput sequencing data. *Nucleic Acids Res.* **38**, e164 (2010).
27. Cole, J., Otvos, J. D. & Remaley, A. T. A translational tool to facilitate use of apolipoprotein B for clinical decision-making. *Clin. Chem.* **69**, 41–47 (2023).

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Author contributions

N.A.M. developed the analysis plan, oversaw the analyses, interpreted the results and drafted the paper. J.P.P. led the CHIP calling and data preparation and contributed to the initial draft of the paper. G.E.M.M. led the statistical analyses and contributed to the initial and subsequent drafts of the paper. F.K. provided additional statistical and analytical support and provided critical review of the paper. M.P.B., R.P.G., B.M.S., S.D.W., D.L.B., P.G.S., I.R. and E.B. contributed to data collection and reviewed the paper. P.L., P.T.E., A.G.B., M.S.S. and C.T.R. provided oversight and guidance throughout the analysis, including critical review and revisions of the paper.

Competing interests

P.L. has a financial interest in TenSixteen Bio (a company targeting somatic mosaicism and CHIP to discover and develop new therapeutics to treat age-related diseases). A.G.B. is on the scientific advisory board of TenSixteen Bio. The other authors declare no competing interests.

Additional information

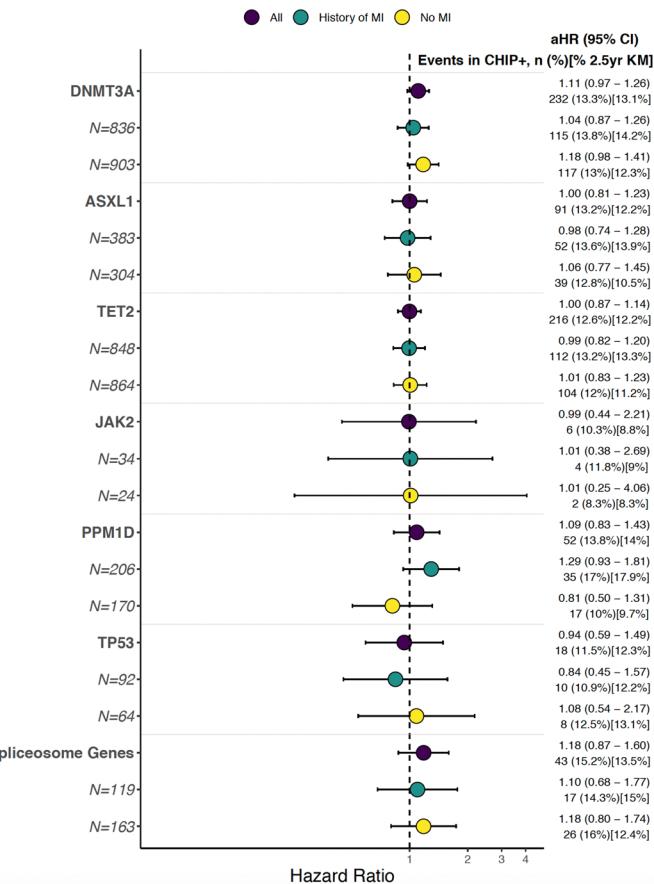
Extended data is available for this paper at
<https://doi.org/10.1038/s41591-024-03188-z>.

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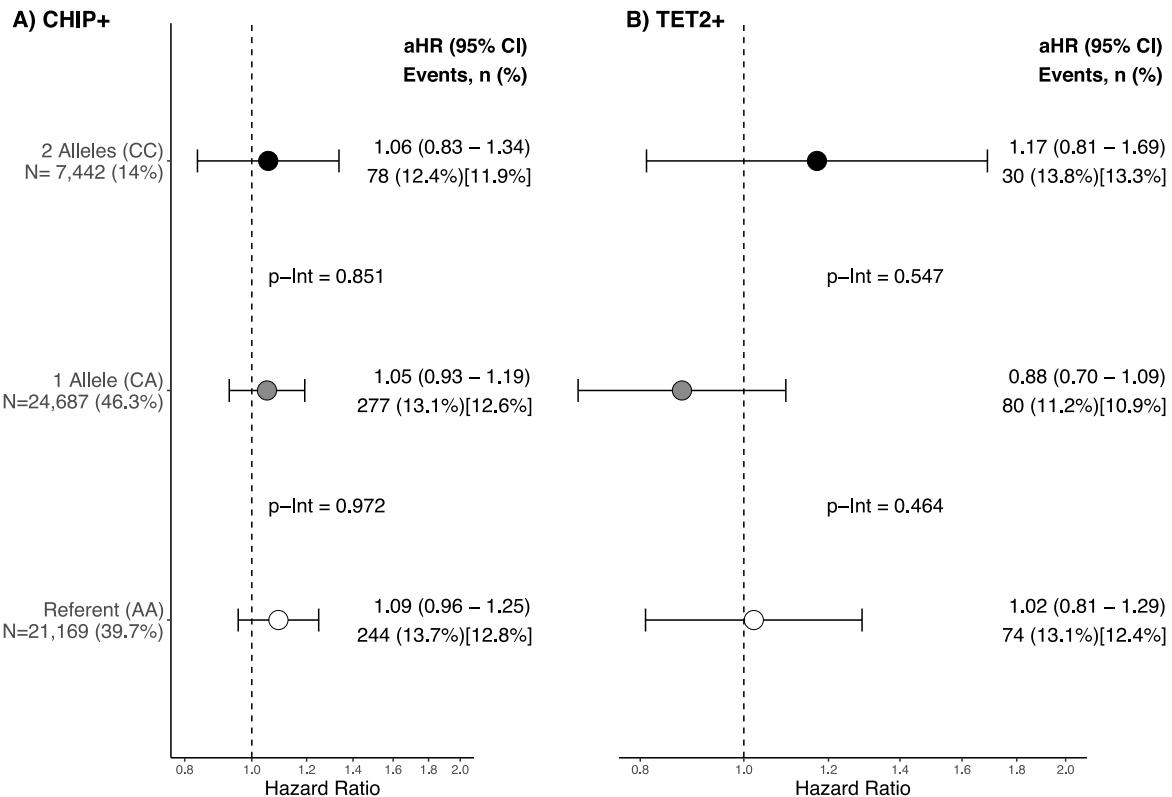
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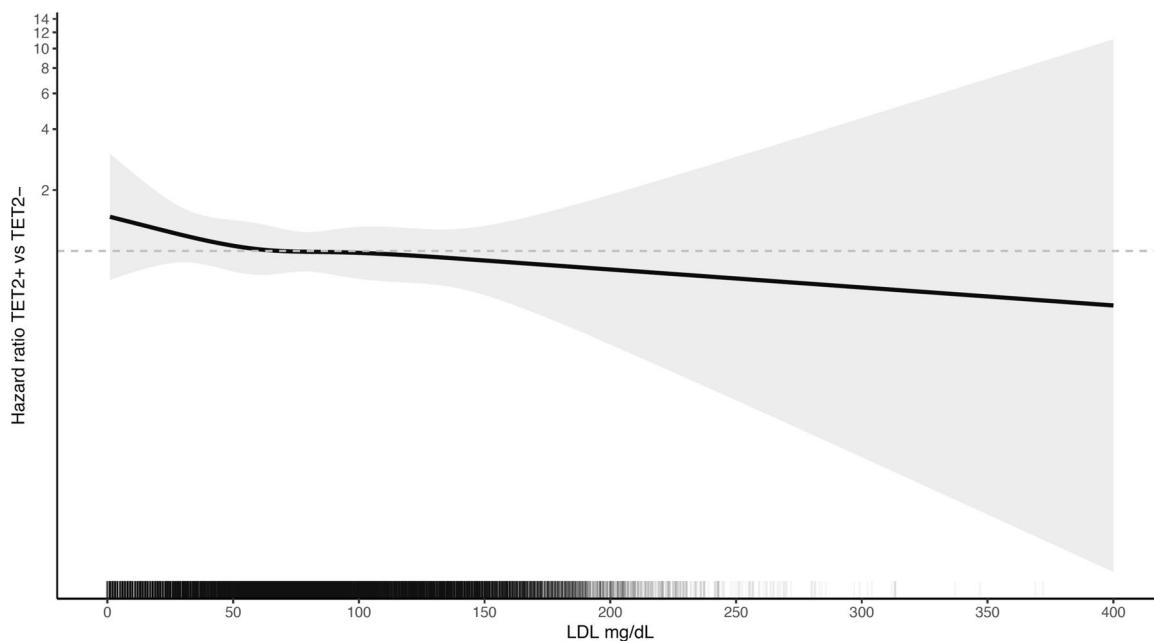
Extended Data Fig. 1 | The association between specific CHIP driver mutations and major cardiovascular events in patients with and without history of MI (n = 63,700). DNA damage genes: TP53 and PPM1D, spliceosome genes: SF3B1, SRSF2, U2AF1. HRs and 95% CI are reported adjusted for age, age2,

sex, BMI, current smoking, history of hypercholesterolemia, systolic blood pressure, history of diabetes, statin use, ancestry (PC 1–10), prior MI, stroke, AF, malignancy, eGFR, PAD, CHF, and trial. Two-sided p-values were obtained via Wald test.



Extended Data Fig. 2 | Association of CHIP subgroups with major cardiovascular events. The association between CHIP (a) and TET2 (b) and major cardiovascular events stratified by IL6R Asp358Ala genotypes (n = 53,298). HRs and 95% CIs are reported adjusted for age, age2, sex, BMI, current smoking,

history of hypercholesterolemia, systolic blood pressure, history of diabetes, statin use, ancestry (PC1–10), prior MI, stroke, AF, malignancy, eGFR, PAD, CHF, and trial. Two-sided p-values were obtained via Wald test.



Extended Data Fig. 3 | Association between TET2+ and major adverse cardiovascular events as a function of LDL-C level. Hazard ratio estimates are derived from an interaction model between TET2 status and a 3-knot restricted cubic spline of LDL-C. 95% CI bands (in gray) were obtained via bootstrap. The model is adjusted for age, age², sex, BMI, current smoking, history of

hypercholesterolemia, systolic blood pressure, history of diabetes, statin use, ancestry (PC 1–10), prior MI, stroke, AF, malignancy, eGFR, PAD, CHF, and trial. P-interaction = 0.26. PEGASUS LDL-C values were approximated by transformed ApoB values. LDL-C values were not available in ENGAGE.

Extended Data Table 1 | Baseline characteristics by trial

Characteristic	DECLARE-TIMI 58 N = 12,685	ENGAGE AF-TIMI 48 N = 13,628	FOURIER (TIMI 59) N = 16,211	PEGASUS-TIMI 54 N = 12,791	SAVOR-TIMI 53 N = 8,385
Age ± SD	64 ± 6.9	70.6 ± 9.3	62.5 ± 9	65.4 ± 8.4	65.3 ± 8.5
Male Sex (%)	8125 (64.1)	8336 (61.2)	12321 (76)	9734 (76.1)	5599 (66.8)
BMI (kg/m ²) ± SD	32.5 ± 6	30 ± 5.9	29.5 ± 5.1	28.7 ± 5	31.5 ± 5.6
ASCVD (%)	5315 (41.9)	6591 (48.4)	16204 (100)	12791 (100)	6411 (76.5)
Prior MI (%)	2788 (22)	1689 (12.4)	13208 (81.5)	12791 (100)	3179 (37.9)
Prior Stroke (%)	791 (6.2)	2377 (17.4)	2945 (18.2)	54 (0.4)	906 (10.8)
History of PAD (%)	771 (6.1)	592 (4.3)	2269 (14)	736 (5.8)	1036 (12.4)
History of Hypertension (%)	11395 (89.8)	12895 (94.6)	13008 (80.2)	9924 (77.6)	6768 (80.7)
History of CHF (%)	1257 (9.9)	7985 (58.6)	3772 (23.3)	2473 (19.3)	892 (10.6)
History of Diabetes (%)	12454 (98.2)	5038 (37)	5696 (35.1)	4116 (32.2)	8382 (100)
History of Hypercholesterolemia (%)	10392 (81.9)	7598 (55.8)	5987 (36.9)	10023 (78.4)	6073 (72.4)
History of CKD (%)	955 (7.5)	1661 (12.2)	2917 (18)	2993 (23.4)	2464 (29.4)
History of AFIB (%)	881 (6.9)	13628 (100)	1463 (9)	599 (4.7)	619 (7.4)
History of CAD (%)	4433 (34.9)	4825 (35.4)	14104 (87)	12791 (100)	5211 (62.1)
History of Coronary Revascularization (%)	3751 (29.6)	1772 (13)	10961 (67.6)	10869 (85)	3683 (43.9)
History of Cancer (%)	393 (3.1)	726 (5.3)	596 (3.7)	866 (6.8)	555 (6.6)
Current Smoker (%)	1786 (14.1)	991 (7.3)	4669 (28.8)	2155 (16.8)	1050 (12.5)
Systolic Blood Pressure (mm Hg) ± SD	135.1 ± 15.3	130.7 ± 15.1	132.5 ± 15.7	132.9 ± 17	136.5 ± 16.8
eGFR (mL/min/1.73m ²) ± SD	85.9 ± 21.7	66.6 ± 17.4	76 ± 18.5	72.4 ± 18.2	72.5 ± 22.8
Hemoglobin (g/dL) ± SD	13.9 ± 1.4	14 ± 1.5	14.2 ± 1.3	14.2 ± 1.3	13.8 ± 1.5
White Blood Count (count/µL) ± SD				7.2 ± 1.9	
Platelets (10 ³ /µL) ± SD	231.6 ± 60.4	202.2 ± 55	221.8 ± 58.3	212.8 ± 57	218.7 ± 62.1
Lipid-lowering Therapy (%)	9617 (75.8)	6826 (50.1)	16202 (99.9)	11892 (93)	6611 (78.8)
Antiplatelet Therapy (%)	7959 (62.7)	4023 (29.5)	15021 (92.7)	8514 (66.6)	633 (7.5)
Anticoagulant Therapy (%)	887 (7)	13628 (100)	1418 (8.7)	0 (0)	723 (8.6)
IL6R rs2228145 (%)	A-A 4930 (39.4) C-A 5655 (45.2) C-C 1913 (15.3)	A-A 4452 (40.2) C-A 5118 (46.2) C-C 1500 (13.6)	A-A 6060 (40.9) C-A 6859 (46.2) C-C 1915 (12.9)	A-A 3712 (38.6) C-A 4560 (47.4) C-C 1354 (14.1)	A-A 2015 (38.2) C-A 2495 (47.3) C-C 760 (14.4)
Ethnicity (%)	Asian 1147 (9) Black 397 (3.1) Other 452 (3.6) White 10689 (84.3)	Asian 111 (0.8) Black 950 (7) Other 391 (2.9) White 12176 (89.3)	Asian 914 (5.6) Black 277 (1.7) Other 240 (1.5) White 14780 (91.2)	Asian 957 (7.5) Black 165 (1.3) Other 114 (0.9) White 11555 (90.3)	Asian 404 (4.8) Black 286 (3.4) Other 1096 (13.1) White 6599 (78.7)

Extended Data Table 2 | Unadjusted and adjusted hazard ratios for study endpoints in CHIP+ patients compared to CHIP-

Model Outcome	Base Model: Unadjusted	Model 2: Adjusted by age, age2, sex	Model 3: Model 2 + Risk Factors	Model 4: All covariates
Major CV events	1.18 (1.09 - 1.27) <0.0001	1.10 (1.02 - 1.19) 0.0171	1.09 (1.01 - 1.18) 0.0303	1.07 (0.99 - 1.16) 0.0861
Myocardial infarction	1.16 (1.01 - 1.33) 0.0377	1.12 (0.98 - 1.29) 0.109	1.10 (0.96 - 1.27) 0.161	1.06 (0.93 - 1.22) 0.386
Ischemic stroke	1.22 (1.02 - 1.47) 0.028	1.05 (0.87 - 1.25) 0.632	1.04 (0.87 - 1.25) 0.677	1.04 (0.86 - 1.24) 0.708
Coronary revascularization	1.06 (0.95 - 1.19) 0.309	1.15 (1.02 - 1.29) 0.02	1.14 (1.02 - 1.28) 0.0258	1.10 (0.98 - 1.24) 0.101
CV death	1.36 (1.18 - 1.55) <0.0001	1.06 (0.92 - 1.21) 0.423	1.04 (0.91 - 1.19) 0.563	1.05 (0.91 - 1.20) 0.52
Non-CV Mortality	1.59 (1.34 - 1.88) <0.0001	1.23 (1.04 - 1.46) 0.0156	1.21 (1.02 - 1.43) 0.0281	1.17 (0.99 - 1.39) 0.0736
Cancer Mortality	1.62 (1.28 - 2.05) <0.0001	1.33 (1.05 - 1.68) 0.0196	1.30 (1.03 - 1.65) 0.0293	1.20 (0.95 - 1.53) 0.128
All-cause Mortality	1.45 (1.31 - 1.61) <0.0001	1.14 (1.03 - 1.27) 0.0126	1.12 (1.01 - 1.25) 0.0295	1.11 (1.00 - 1.23) 0.0531

Risk factors = smoking, hypertension, hypercholesterolemia, diabetes, and eGFR.

Additional covariates in model 4 = BMI, principal components 1 to 10, lipid-lowering therapy, prior MI, heart failure, atrial fibrillation, PAD, prior stroke, history of cancer, and trial.

Extended Data Table 3 | Sensitivity analysis of the association between CHIP and major cardiovascular events using leave one trial out (N-1) approach

Trial	aHR(95% CI)	P-value	n/N (%)
All Trials	1.07(0.99,1.16)	0.086	7453/63700 (11.7%)
w/o ENGAGE	1.09(0.99,1.20)	0.065	5719/50072 (11.4%)
w/o FOURIER	1.08(0.99,1.18)	0.079	5776/47489 (12.2%)
w/o SAVOR	1.05(0.97,1.15)	0.228	6535/55315 (11.8%)
w/o PEGASUS	1.08(0.99,1.17)	0.104	5689/51015 (12%)
w/o DECLARE	1.06(0.97,1.16)	0.186	(11.2%)

HRs and 95% CI are reported adjusted for age, age², sex, BMI, genetic principal components 1–10, lipid-lowering therapy, blood pressure, diabetes, hyperlipidemia, eGFR, prior myocardial infarction, heart failure, atrial fibrillation, peripheral artery disease, prior stroke, history of cancer, trial and smoking. Two-sided p-values were obtained via Wald test.

Extended Data Table 4 | Competing risk analysis using Fine–Gray model to evaluate the association between CHIP and (a) major vascular events and (b) MACE stratified by trial

A)

Outcome	aHR Fine-Gray	P-value Fine-Gray	aHR Cox PH	P-value Cox PH	n/N (%)
MACE	1.07(0.99,1.16)	0.100	1.07(0.99,1.16)	0.086	7453/63700 (11.7%)
Coronary Revascularization	1.1(0.98,1.23)	0.120	1.10(0.98,1.24)	0.101	3732/63700 (5.9%)
Ischemic Stroke	1.03(0.86,1.24)	0.730	1.04(0.86,1.24)	0.708	1362/63700 (2.1%)
PAD	0.91(0.67,1.23)	0.540	0.92(0.67,1.24)	0.572	537/63694 (0.8%)
Cancer Death	1.22(0.96,1.55)	0.110	1.20(0.95,1.53)	0.128	641/63700 (1%)
Cardiovascular Death	1.05(0.91,1.2)	0.520	1.05(0.91,1.20)	0.520	2201/63700 (3.5%)
Non-CV Death	1.18(0.99,1.4)	0.063	1.17(0.99,1.39)	0.074	1279/63700 (2%)

B)

Trial	aHR Fine-Gray	P-value Fine-Gray	aHR Cox PH	P-value Cox PH	n/N (%)
All (Trial Level M-A)	1.07 (0.99-1.16)	0.050	1.08 (1.00-1.17)	0.045	7453/63700 (11.7%)
ENGAGE	1.04(0.89,1.22)	0.610	1.04(0.89,1.21)	0.617	1734/13628 (12.7%)
FOURIER	1.03(0.86,1.22)	0.780	1.03(0.86,1.22)	0.766	1677/16211 (10.3%)
SAVOR	1.19(0.96,1.47)	0.120	1.19(0.96,1.48)	0.110	918/8385 (10.9%)
PEGASUS	1.04(0.86,1.26)	0.690	1.05(0.86,1.26)	0.647	1360/12791 (10.6%)
DECLARE	1.12(0.94,1.33)	0.200	1.13(0.95,1.34)	0.160	1764/12685 (13.9%)

Extended Data Table 5 | Association between CHIP+ and TET2+ and major cardiovascular events stratified by statin use

Group	CHIP	HR	CI_L	CI_U	P_value
All	CHIP+	1.07	0.99	1.16	0.0819
No Statin		1.06	0.90	1.26	0.476
Statin Use		1.08	0.99	1.18	0.103
All	TET2+	1.00	0.87	1.15	0.979
No Statin		1.06	0.80	1.40	0.679
Statin Use		0.99	0.84	1.16	0.882

Reporting Summary

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- For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes
- Estimates of effect sizes (e.g. Cohen's d , Pearson's r), indicating how they were calculated

Our web collection on [statistics for biologists](#) contains articles on many of the points above.

Software and code

Policy information about [availability of computer code](#)

Data collection

N/A

Data analysis

Somatic mutations were detected using the GATK Mutect2 (doi:<https://doi.org/10.1101/861054>, BioRxiv) somatic mutation calling algorithm (v4.2.5.0 for ENGAGE AF, SAVOR, PEGASUS, and DECLARE, and v4.1.4.1 for FOURIER). The interactionRCS R package (<https://CRAN.R-project.org/package=interactionRCS>) was used.

For manuscripts utilizing custom algorithms or software that are central to the research but not yet described in published literature, software must be made available to editors and reviewers. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Portfolio [guidelines for submitting code & software](#) for further information.

Data

Policy information about [availability of data](#)

All manuscripts must include a [data availability statement](#). This statement should provide the following information, where applicable:

- Accession codes, unique identifiers, or web links for publicly available datasets
- A description of any restrictions on data availability
- For clinical datasets or third party data, please ensure that the statement adheres to our [policy](#)

Due to contractual agreements with trial sponsors, trial data are not available to share.

Research involving human participants, their data, or biological material

Policy information about studies with [human participants or human data](#). See also policy information about [sex, gender \(identity/presentation\), and sexual orientation](#) and [race, ethnicity and racism](#).

Reporting on sex and gender

We report sex in this study and adjust for it in the analysis. There were no differences in results by sex.

Reporting on race, ethnicity, or other socially relevant groupings

We report race/ethnicity in Table 1 and Extended Table 1.

Population characteristics

Population characteristics have been outlined in detail in Table 1 and Extended Table 1.

Recruitment

Patients were recruited to one of the five trials included in this analysis based on the inclusion/exclusion criteria in each protocol. Potential bias from trial recruitment is discussed in the Discussion/Limitations and addressed with multiple sensitivity analyses.

Ethics oversight

Each trial's ethical oversight is described in the trial publication paper.

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Field-specific reporting

Please select the one below that is the best fit for your research. If you are not sure, read the appropriate sections before making your selection.

Life sciences Behavioural & social sciences Ecological, evolutionary & environmental sciences

For a reference copy of the document with all sections, see nature.com/documents/nr-reporting-summary-flat.pdf

Life sciences study design

All studies must disclose on these points even when the disclosure is negative.

Sample size

Sample size was determined to show treatment effect in each trial. This study included all of our trials with genotyped.

Data exclusions

Trial patients who did not consent for exome sequencing were unable to have CHIP evaluated and therefore had to be excluded from this analysis.

Replication

We ran these analyses across 5 unique clinical trials, and evaluated the results together, individually, and using a leave one out approach (N-1).

Randomization

Within each trial, random allocation of therapy was used. This allowed us to test CHIP x treatment interactions within certain trials.

Blinding

Each trial was a double blind placebo-control trial where both patient and investigator were blinded to the treatment assignment. In addition, clinical endpoints were adjudicated by a centralized, blinded clinical endpoints committee. Finally, exome sequencing and CHIP calling was not performed until after the trial, so nobody knew CHIP status during the trial.

Reporting for specific materials, systems and methods

We require information from authors about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each material, system or method listed is relevant to your study. If you are not sure if a list item applies to your research, read the appropriate section before selecting a response.

Materials & experimental systems

- | | |
|-------------------------------------|--|
| n/a | Involved in the study |
| <input checked="" type="checkbox"/> | <input type="checkbox"/> Antibodies |
| <input checked="" type="checkbox"/> | <input type="checkbox"/> Eukaryotic cell lines |
| <input checked="" type="checkbox"/> | <input type="checkbox"/> Palaeontology and archaeology |
| <input checked="" type="checkbox"/> | <input type="checkbox"/> Animals and other organisms |
| <input type="checkbox"/> | <input checked="" type="checkbox"/> Clinical data |
| <input checked="" type="checkbox"/> | <input type="checkbox"/> Dual use research of concern |
| <input checked="" type="checkbox"/> | <input type="checkbox"/> Plants |

Methods

- | | |
|-------------------------------------|---|
| n/a | Involved in the study |
| <input checked="" type="checkbox"/> | <input type="checkbox"/> ChIP-seq |
| <input checked="" type="checkbox"/> | <input type="checkbox"/> Flow cytometry |
| <input checked="" type="checkbox"/> | <input type="checkbox"/> MRI-based neuroimaging |

Clinical data

Policy information about [clinical studies](#)

All manuscripts should comply with the ICMJE [guidelines for publication of clinical research](#) and a completed [CONSORT checklist](#) must be included with all submissions.

Clinical trial registration PEGASUS (NCT01225562), ENGAGE (NCT00781391), FOURIER (NCT01764633), DECLARE (NCT01730534), SAVOR (NCT01107886)

Study protocol Each of the 5 trials has a study protocol that can be found in association with the primary publication.

Data collection Clinical data was collected prospectively at the site level and entered into the central database for each trial. The years that each trial was actively collecting data were as follows: DECLARE (2013-2018), FOURIER (2013-2017), PEGASUS (2010-2014), SAVOR (2010-2013), ENGAGE (2008-2013).

Outcomes Primary and secondary endpoints for each trial were pre-specified in the trial protocols and statistical analysis plans. They were collected by sites and sent to the CEC for blinded adjudication based on the CEC Charter for each trial.