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Frequency of clonal hematopoiesis of indeterminate potential in patients over 75 years with a first cardiovascular event. Consequences on inflammation and atherosclerosis

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Fréquence de l'hématopoïèse clonale chez les patients de plus de 75 ans présentant un premier événement cardiovasculaire
Conséquences sur l'inflammation et le volume athéromateux

Thèse pour l'obtention du diplôme d'État de Docteur en Médecine
Cardiovasculaire

Présentée et soutenue publiquement par

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Table des matières

CLONAL HAEMATOPOIESIS AND CARDIOVASCULAR DISEASES: A GROWING RELATIONSHIP 5

RESUME 5
SUMMARY 6
LIST OF ABBREVIATIONS 7
BACKGROUND 8
CHIP 9
How to detect CHIP 9
Prevalence 11
CHIP and hematological malignancy 11
CHIP LINKS TO CVD (TABLE 2) 13
Atherothrombosis 13
Heart failure and valvular heart disease 13
HOW DOES CHIP INCREASE CARDIOVASCULAR RISK? 16
Experimental approaches 16
Human data 17
IMPLICATIONS FOR CLINICAL PRACTICE 19
What is the place for CHIP detection in the management of CVD? 19
Anti-inflammatory therapies 20
CONCLUSIONS 21
REFERENCES 22

MISE A JOUR DES DERNIERES PUBLICATIONS 24

REFERENCES 29

FREQUENCY OF CLONAL HEMATOPOIESIS IN PATIENTS OVER 75 YEARS WITH A FIRST CARDIOVASCULAR EVENT. CONSEQUENCES ON INFLAMMATION AND ATHEROSCLEROSIS 31

LIST OF ABBREVIATIONS 32
INTRODUCTION 33
METHODS 34
Patients 34
Study procedures 35
Collected data 35
Data from the index event 37
Data at inclusion 37
Aims of the study 39
Statistical analyses 40
RESULTS 40
Patients & myocardial infarction data 40
CHIP status and acute phase characteristics 44
CHIP status and inflammatory markers, atheroma burden, and complications 47
DISCUSSION 55
Prevalence of CHIP 55
Inflammation 57
Atherosclerosis burden 58
Influence on prognosis 59
Interaction with risk factors 60
Study strengths and limitations 61
Future directions 61
CONCLUSION 62
REFERENCES 63
APPENDIX 66

SERMENT D'HIPPOCRATE 68

Clonal haematopoiesis and cardiovascular diseases: A growing relationship

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Résumé

Les maladies cardiovasculaires, en particulier l'athérosclérose, sont la principale cause de décès dans le monde, mais leur pathogénie n'est pas encore complètement élucidée. En effet, les facteurs de risque cardiovasculaires traditionnels sont bien connus depuis de nombreuses années mais ne suffisent pas à prédire le risque individuel. L'hématopoïèse clonale de potentiel indéterminé (CHIP) a été décrite récemment. Elle correspond à l'expansion clonale d'une population de cellules hématopoïétiques en réponse à l'acquisition d'une mutation somatique, sans aucun signe clinique ou biologique d'hémopathie maligne. La prévalence de cette affection augmente avec l'âge, pour atteindre 10–20 % de la population générale de plus de 70 ans. De récentes études observationnelles ont montré un lien entre la CHIP et les maladies cardiovasculaires chez l'homme, révélant que les porteurs de CHIP avaient plus de risque d'infarctus du myocarde, d'insuffisance cardiaque ou de rétrécissement aortique serré. Le pronostic de ces pathologies semble également être grevé par la présence d'une CHIP. Des études expérimentales ont par la suite mis en lumière le rôle crucial du système immunitaire et de l'inflammation, en particulier des macrophages produisant de l'interleukine-1β, pour expliquer les conséquences cardiovasculaires de la CHIP, à travers leur action sur la plaque d'athérome et les tissus myocardiques. Notre objectif était d'écrire une revue approfondie de ce qui est connu actuellement sur la CHIP et ses conséquences cardiovasculaires, les mécanismes physiopathologiques conduisant au sur-risque cardiovasculaire, et enfin l'influence attendue sur notre pratique quotidienne et la façon dont nous prenons soin des patients porteurs de CHIP.

Summary

Cardiovascular diseases, particularly atherothrombosis, are the leading cause of death worldwide, but their mechanisms are not yet fully understood. Traditional cardiovascular risk factors have been known for many years, but are not enough to predict individual risk. Clonal hematopoiesis of indeterminate potential (CHIP) has been described recently; it corresponds to the clonal expansion of a population of hematopoietic cells in response to the acquisition of a somatic mutation, without any clinical or biological sign of hematological malignancy. The prevalence of this condition increases with age, reaching 10–20% of the general population aged > 70 years. Recent observational studies have shown a link between CHIP and cardiovascular diseases in humans, revealing that CHIP carriers have a higher risk of myocardial infarction, heart failure and severe aortic valve stenosis. The prognosis of these conditions also seems to be altered by the presence of CHIP. Experimental studies have identified that the immune system and inflammation – particularly interleukin-1β-secreting macrophages – play a critical role in enhancing the cardiovascular consequences of CHIP, through their action on the atherosclerotic plaque and myocardial tissues. We aimed to write an extensive review of what is currently known about CHIP and its cardiovascular consequences, the pathophysiological mechanisms leading to the increased cardiovascular risk and, finally, the expected influence on our daily practice and how we care for patients with CHIP.

List of abbreviations

CHIP: clonal hematopoiesis of indeterminate potential

CVD: cardiovascular disease

DNA: deoxyribonucleic acid

HSC: hematopoietic stem cell

hsCRP: high-sensitivity C-reactive protein

IL: interleukin

NLRP3: nucleotide-binding oligomerization domain-like receptor family pyrin domain containing 3

TAVI: transcatheter aortic valve replacement

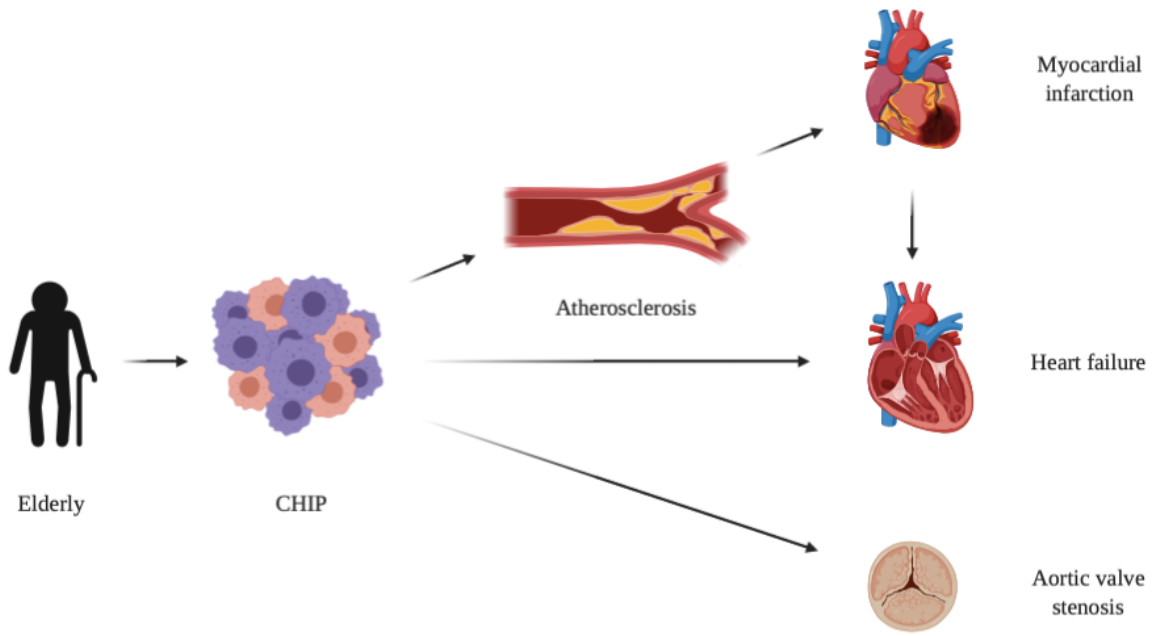
VAF: variant allelic fraction

Background

Atherothrombosis is the main cause of death worldwide. Although our knowledge and ability to treat patients has improved in recent years, some aspects of the disease remain unclear. We know from our daily practice that people without any traditional cardiovascular risk factors can experience cardiovascular events. Furthermore, within a group of people of the same age bearing the same cardiovascular risk factors, some will experience myocardial infarction and others will not. Thus, traditional risk factors account for only 75–80% of events associated with cardiovascular disease (CVD).[1] This underscores the need to identify other contributing factors to enhance our understanding of the disease and improve strategies for both treatment and prevention.

A new concept has emerged recently, which places the role of the hematopoietic system at the center of this investigation. Here we provide a review of the current state of knowledge about clonal hematopoiesis of indeterminate potential (CHIP), its link with CVD, and the potential implications for our daily practice, both current and future.

Central illustration.



Clonal hematopoiesis of indeterminate potential (CHIP) occurs particularly in the elderly; it is the result of an acquired mutation occurring in a population of hematopoietic stem cells, conferring a growth advantage to their progeny. Because of the proinflammatory phenotype, it favors atherothrombosis, leading to coronary artery diseases such as myocardial infarction, and is also associated with heart failure (ischemic and non-ischemic) as well as aortic stenosis, altering the prognosis of CHIP carriers.

CHIP

A somatic mutation is a genetic alteration acquired by a cell that can be passed to the progeny of the mutated cell during the course of cell division. Somatic mutations differ from germ-line mutations, which are inherited genetic alterations that occur in the germ cell.

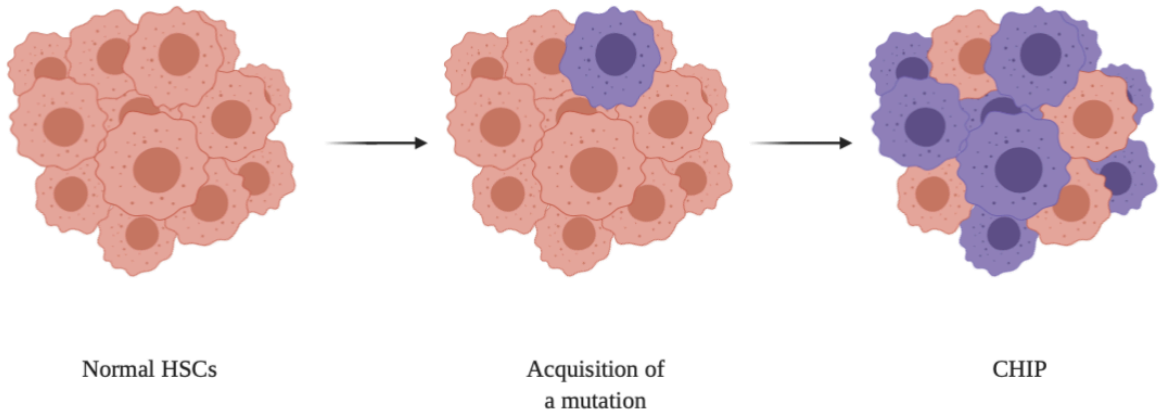
Somatic mutations accumulate in normal tissues as a function of time. The majority of these mutations have no effect on cell behavior, so selection does not act upon them. Rarely, a mutation will arise that confers a growth advantage to the cell in which it occurs. Such a mutation would allow that cell and its progeny, referred to as a "clone," to expand progressively over time. This is now known to occur in a number of tissues, particularly in elderly individuals. When this happens in a hematopoietic stem cell (HSC), a condition known as "clonal hematopoiesis" appears as the result of the contribution of the mutated clone to the production of a substantial proportion of mature blood cells (Figure 1). Clonal hematopoiesis is well known in the scope of hematological malignancies as being a step towards overt neoplasms, such as acute myeloid leukemias, when the somatic mutation involves a leukemia driver gene.

CHIP was described several years ago, and is defined by the clonal expansion of a hematopoietic cell population in response to the acquisition of a leukemia driver gene mutation, at a variant allelic fraction (VAF) ≥ 2%, without any biological or clinical sign of hematological malignancy.[2] Mutations observed in CHIP frequently involve epigenetic regulators, the main ones being DNMT3A, TET2 and ASXL1 (Table 1).[3] Because these somatic mutations do not significantly disturb cell differentiation, mutated mature cells are produced as normal cells, and circulate in the peripheral blood. Thus, during this premalignant state, patients have totally normal blood cell counts. The involvement in CHIP of the $JAK2^{V617F}$ mutation, which is known for being involved in myeloproliferative neoplasms, is a matter of debate.[4-6]

How to detect CHIP

Because the acquired mutation is present in mature blood cells, detecting CHIP is feasible in deoxyribonucleic acid (DNA) from circulating leukocytes from a blood sample. A bone marrow test is not necessary. Next-generation sequencing has been used as the reference technique for genomic DNA sequencing for almost a decade; its cost has decreased rapidly over the last few years, currently allowing the entire genome to be sequenced in a few days for approximately €1000. However, whole-genome sequencing is a suboptimal technique to search for CHIP because of its lack of sensitivity. As CHIP is defined as the presence of a mutation with a VAF ≥ 2%, more than 500 DNA molecules would need to be analyzed to obtain reliable

Figure 1. Generation of a clone



In the bone marrow, a hematopoietic stem cell (HSC) acquires a somatic mutation in a leukemia driver gene, conferring to its lineage a growth advantage leading to clonal expansion. CHIP: clonal hematopoiesis of indeterminate potential.

Table 1. The most commonly mutated genes[a] observed in large cohort of clonal hematopoiesis (7-12)

Mutated gene		Frequency, %
DNMT3A	DNA methyltransferase 3A	49.1
TET2	Ten-eleven translocation 2	18.9
ASXL1	Additional sex combs-like 1	7.1
PPM1D	Protein phosphatase, Mg^{2+}/Mn^{2+}-dependent 1D	2.7
SF3B1	Splicing factor 3B subunit 1	2.7
TP53	Tumour protein 53	2.5
SRSF2	Serine/arginine-rich splicing factor 2	2
CBL	Casitas B-lineage lymphoma	0.6
GNB1	Guanine nucleotide-binding protein subunit beta-1	0.5
Other		14

DNA: deoxyribonucleic acid; Mg: magnesium; Mn: manganese.

[a] The $JAK2^{V617F}$ mutation was excluded.

results for a given gene. To date, such a sequencing depth cannot be achieved easily using whole-genome or even whole-exome sequencing.

Many genes have been identified as being involved in CHIP. However, most of the time, a patient with CHIP has only one mutation. Moreover, only some of these mutations have been identified as mutated recurrently in CHIP, and are detected in > 90% of CHIP carriers (Table 1). The three most frequently mutated genes (TET2, DNMT3A and ASXL1) represent > 70% of CHIP cases. Thus, restricting sequencing analysis to a panel of genes is a worthwhile strategy for detecting CHIP. Indeed, by limiting the genomic regions analyzed, targeted sequencing allows the depth of sequencing to be increased, thus improving the ability to detect CHIP with low-VAF mutations. Such a strategy also reduces the cost and time of analysis. In this way, CHIP could be detected in routine practice in a week for less than €100.

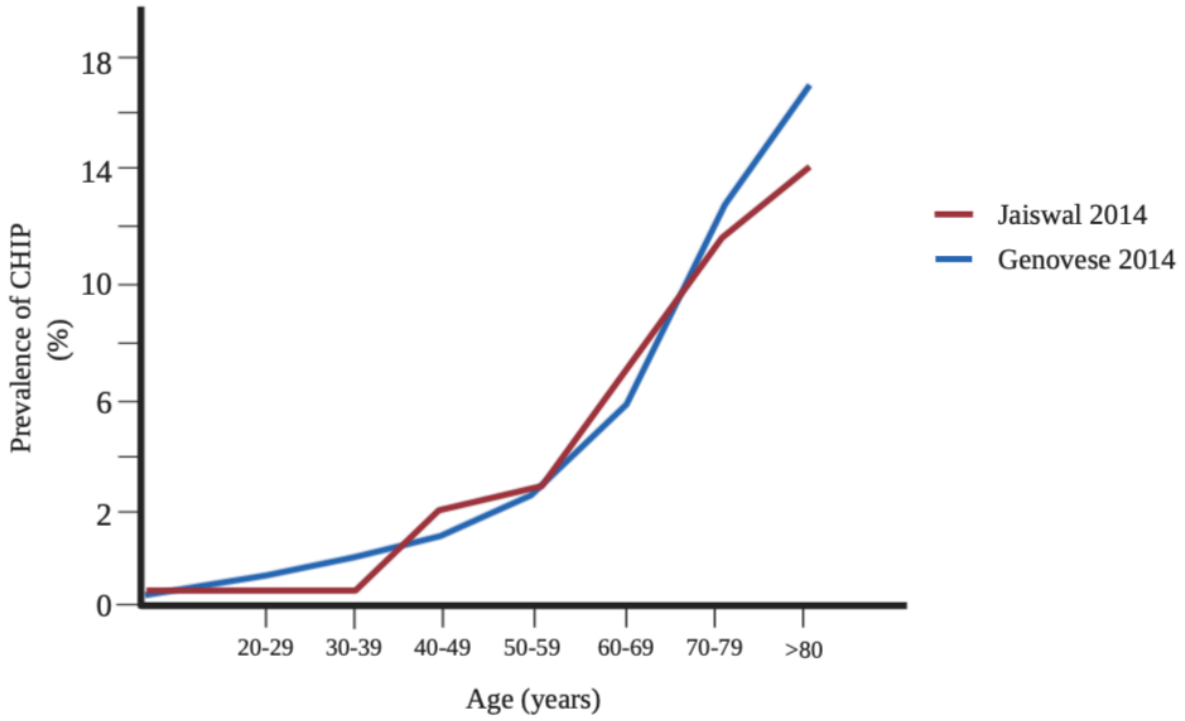
Prevalence

In 2014, two studies evaluated the prevalence of individuals presenting with CHIP (Figure 2).[7, 8] When analyzing blood samples, CHIP seems to be rare before the age of 40 years; its prevalence then increases with age, with an estimated frequency of 5% at 60 years, and reaching 10–20% of those aged > 70 years. Recently, Busque et al. studied a population with a mean age of 75 years, and found that 23% of the cohort had CHIP.[9] Some authors reported a higher prevalence of CHIP in healthy individuals (40% after 50 years of age) when detecting mutations in the bone marrow with a sensitivity threshold of 1%.[10]

CHIP and hematological malignancy

Although Jaiswal et al. demonstrated that CHIP carriers had an 11-fold increased risk of developing hematological malignancies (myelodysplastic syndromes, acute leukemias or even lymphomas), the absolute risk of evolving to a hematological cancer remains low (about 1% per year).

Figure 2. Prevalence of clonal hematopoiesis of indeterminate potential (CHIP) according to studies



CHIP: clonal hematopoiesis of indeterminate potential

CHIP links to CVD (Table 2)

Atherothrombosis

Surprisingly, when Jaiswal et al. looked at the prognosis of CHIP carriers in 2014, they found that in addition to an increased risk of hematological malignancies, they had an increased risk of overall mortality.[8] This increased mortality was driven primarily by increased cardiovascular-related mortality, and – interestingly – not deaths resulting from cancer. In this study, the risk of cardiovascular ischemic events was increased by a factor of 2, and the risk of ischemic stroke by a factor of 2.6. Because of these surprising results, the same team tested the hypothesis that CHIP is correlated to CVD. They chose two cohorts of people with mean ages of 60 and 70 years, bearing cardiovascular risk factors, and followed them for several years; they found a twofold increase in the risk of coronary artery disease in CHIP carriers compared with non-CHIP carriers.[11] Interestingly, in two additional cohorts of people aged < 50 years, they found a fourfold increase in early onset myocardial infarction in those who presented with CHIP.[11] Additionally, there was a significant increase in the coronary artery calcium score evaluated by computed tomography in CHIP carriers: compared with non-carriers, CHIP carriers had a threefold increased risk of having a coronary artery calcium score > 615 Agatston units, which identifies those at highest risk of coronary artery disease.[7, 11]

These observations raise the question of how much influence CHIP has in the development of CVD, when compared with traditional risk factors, such as smoking, hypertension, dyslipidemia, diabetes or family history of CVD. Jaiswal and Libby thus studied the weight of each factor in the development of CVD, based on previous cohorts, and found that the presence of CHIP could be one of the strongest factors associated with CVD, conferring a higher relative risk than smoking, hypertension and dyslipidemia.[12] However, some questions remain. It is still not known whether CHIP is an independent risk factor or if its presence potentiates existing ones, increasing the risk of CVD developing in people harboring traditional risk factors.

Heart failure and valvular heart disease

CHIP made an entrance into the world of CVD through the study of atherosclerotic CVD, but the association is likely to be much deeper. Dorsheimer et al. studied the prevalence and prognosis of CHIP in a population of 200 patients with chronic heart failure, with a mean

Table 2. Overview of main studies regarding clonal hematopoiesis of indeterminate potential (prevalence, risk of cardiovascular disease and prognosis)

Study	Number and origin of patients	Main outcome assessed	Main result	Adjustment for potential confounders	Potential bias/limitations	Main genes concerned	Weight of CHIP on the risk of CVD
Jaiswal et al. 2014 [8]	15,801; international cohort	Detection of acquired somatic mutations in elderly without known hematological disorders	**Increase in frequency of somatic mutations with age**, from 5.6% between 60 and 69 years to 18.4% after 90 years. **Increased risk of death**, mainly from cardiovascular causes	Age, sex, T2D	-	DNMT3A; TET2; ASXL1	**HR for CHD of 2.0** (95% CI 1.2–3.5; P < 0.02), higher than hypertension (1.4), lower than age (2.4 after 60 years) and T2D (3.1); HR for ischemic stroke of 2.6 (95% CI 1.3–4.8; P < 0.003)
Genovese et al. 2014 [7]	12,380; Sweden	Detection of clonal expansion and somatic mutations in healthy people; analyses of subsequent medical outcomes	**Increase in frequency of somatic mutations with age**, with 10.4% of people aged > 65 years being affected. Increased risk of cancer and reduced overall survival	Age, sex	-	DNMT3A; TET2 ASXL1; PPM1D	-

(continued next page)

Jaiswal et al. 2017 [11]	8255; international cohort	Testing the hypothesis that CHIP increases the risk of atherosclerotic CVD	Increased risk of CHD (HR 1.9, 95% CI 1.4–2.7; P < 0.001) and early-onset MI (HR 4.0, 95% CI 2.4–6.7; P < 0.001)	Age, sex, T2D, smoking status, hyperlipidemia, hypertension	Case-control study	DNMT3A; TET2; ASXL1	**Higher HR for CHD than for age (1.0), smoking status (1.04) or diabetes (0.9)**; slightly lower HR for CHD than for hypertension (2.01)
Mas-Peiro et al. 2019 [15]	279; Germany	Assessment of prevalence and prognostic value of DNMT3A and TET2 mutation in patients undergoing TAVI	**33.3% of patients undergoing TAVI had CHIP.** **Increased deaths following TAVI**	Age, sex, NT-proBNP	Monocentric	DNMT3A; TET2	HR for death after TAVI of 4.81 (95% CI 1.49–15.57; P = 0.009)
Dorsheimer et al. 2019 [13]	200; Germany	Assessment of prevalence and prognostic value of CHIP in patients with chronic ischemic heart failure	**18.5% of patients with chronic heart failure had CHIP.** **Increased risk of death** (HR 2.11, 95% CI 1.05–4.24; P = 0.04) **and hospitalization for heart failure** (HR 2.09, 95% CI 1.10–3.95; P = 0.02)	Age, hypertension	Monocentric	DNMT3A; TET2	Higher HR for death and hospitalization than for age (1.04) or hypertension (0.95)

BMI: body-mass index, CHD: coronary heart disease, CHF: chronic heart failure, CHIP: clonal hematopoiesis of indeterminate potential, CVD: cardiovascular diseases, HF: heart failure, HR: hazard-ratio, hsCRP: high-sensitive C-reactive protein, MI: myocardial infarction, TAVI: transcatheter aortic valve implantation, T2D: type 2 diabetes, VAF: variant allele frequency

age of 65 years.[13] The team found an increased prevalence of CHIP in this population compared with what is described in the general population, with more than 18% of the total cohort harboring CHIP.[7, 8, 14] This increased prevalence was constant in each age category, with a more than twofold increase in all groups. Consistent with previous data in the general population, DNMT3A and TET2 were the two genes most affected by somatic mutations. CHIP carriers appeared to have a significantly worse prognosis than non-carriers regarding both death and hospitalization for heart failure. Indeed, 37% of patients harboring CHIP died during a 4-year follow-up period, versus 24% of those who did not have CHIP. The risk of death combined with rehospitalization for heart failure was multiplied by a factor of 3. Interestingly, there seemed to be a dose-response effect between a poor prognosis and the clone size estimated by the VAF.

More recently, Mas-Peiro et al. studied the prevalence and prognosis of patients harboring CHIP among a population with a mean age of 83 years undergoing transcatheter aortic valve replacement (TAVI) for the treatment of degenerative aortic valve stenoses.[15] They found a higher prevalence of CHIP in this population than in healthy controls, with approximately 33% of the TAVI group harboring a mutation in DNMT3A or TET2 (the only two genes studied) at a VAF ≥ 2%. Similar to the general population, the prevalence seemed to increase with age, from 25% in patients between the ages of 55 and 69 years, to more than 52% in those aged > 90 years. Again, as in atherosclerotic CVD and heart failure, the prognosis seemed to be worse in the presence of a candidate driver gene mutation. Over an 8-month follow-up period, it was found that CHIP carriers had a fourfold increase in their risk of death.

Taken together, these observations strongly suggest that clonal hematopoiesis may play a role in the development of different CVD.

How does CHIP increase cardiovascular risk?

Experimental approaches

In 2017, Fuster et al. evaluated the role of TET2 mutations in the development of atherosclerosis in mice.[16] Using a mouse model in which 10% of HSCs were inactivated for TET2, they observed an amplification of blood cells bearing the TET2 mutation, as well as a marked increase in atherosclerotic burden, with a 60% increased thickness of the atherosclerotic plaques in the aortic root compared with control mice. This finding is consistent with the results of a study conducted by Jaiswal et al. in the same year, which reported that mice bearing a

homozygous mutation of TET2 showed an increase in atherosclerotic lesions of the aorta, from 1.4-fold in the aortic root to 2.7-fold in the descending aorta compared with control mice.[11]

Both studies showed that macrophages, which populated the plaques and harbored the mutation, had enhanced inflammatory activity, producing more cytokines, particularly interleukin (IL)-1β. Of note, Fuster et al. showed that this overproduction could be explained by an increase in activity of the nucleotide-binding oligomerization domain-like receptor family pyrin domain containing 3 (NLRP3) inflammasome, an intracellular oligomeric protein complex that cleaves the pro-IL-1β into IL-1β (Figure 3).[16]

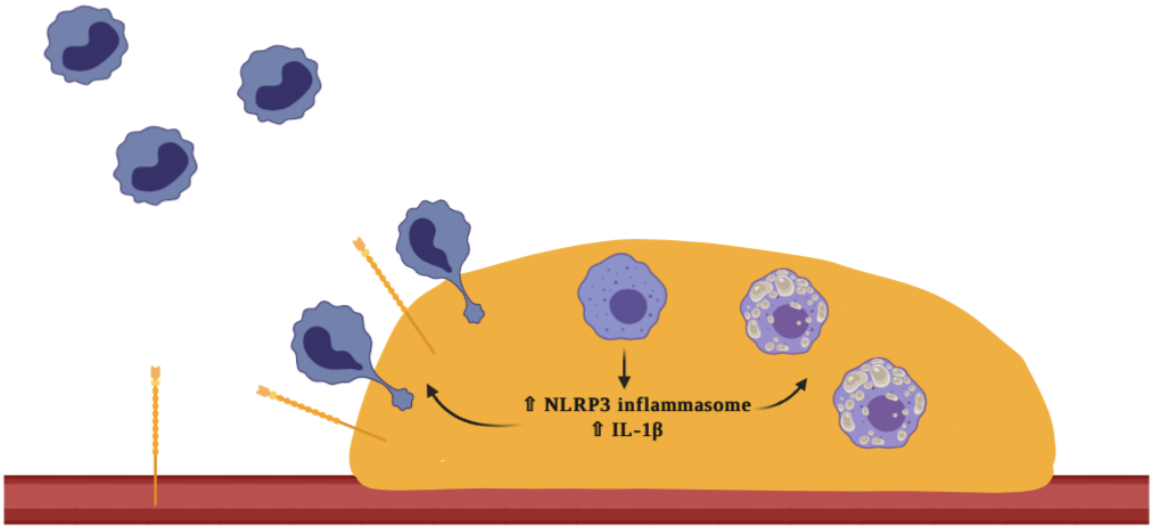
Other studies have focused on mechanisms underlying the relationship between CHIP and heart failure. Sano et al. demonstrated in both ischemic and non-ischemic heart failure models that TET2 mutations were associated with adverse remodeling, a decrease in left ventricular ejection fraction and an increase in left ventricular systolic and diastolic volumes, as well as larger areas of fibrosis in the marginal zone.[17] As for atherosclerosis, this phenomenon was linked with greater macrophage infiltration in the marginal zone, and the expression of various proinflammatory cytokines and chemokines, including IL-1β. In the same year, Sano et al. also demonstrated in another heart failure model that *DNMT3A* mutations were responsible for a similar proinflammatory phenotype of mutated myeloid cells.[18]

In summary, these studies support the hypothesis that *TET2* and *DNMT3A* mutations confer an advantage to mutated cells, as well as a proinflammatory phenotype, responsible for enhanced recruitment of leukocytes to cardiovascular tissues, and leading to the development of atherosclerosis or heart failure (Central illustration). This deleterious phenotype involves the NLRP3 inflammasome, which could represent a therapeutic target in various CVDs linked to CHIP. By comparison, little is known about the pathophysiology linking other CHIP-associated mutations to cardiovascular complications.

Human data

Although studies conducted in a mouse model have suggested that CHIP triggers an inflammatory state, few studies have been conducted in humans to verify this hypothesis. High-sensitivity CRP (hsCRP) has been used as a predictor of cardiovascular events for many years, as it reflects the patient's inflammatory state and their susceptibility to plaque rupture.[19] In a recent study, Busque et al. suggested that CHIP carriers had slightly higher concentrations of hsCRP than non-carriers.[9] Mas-Peiro et al. also speculated that CHIP was associated with a proinflammatory state in patients.[15] Thus, TET2 mutations would be linked to an increase in

Figure 3. Clonal hematopoiesis of indeterminate potential increases atherothrombosis



Circulating mutated monocytes migrate to the atherosclerotic plaque, switching to macrophages. Mutated cells overproduce interleukin (IL)-1β because of excessive activation of the nucleotide-binding oligomerization domain-like receptor family pyrin domain containing 3 (NLRP3) inflammasome, which increases phagocytosis and production of foam cells and recruitment of monocytes by overexpression of adhesion molecules (P-selectin), enhancing atherothrombosis.

the frequency of inflammatory monocytes known to be involved in atherosclerosis development, whereas DNMT3A mutations would favor a proinflammatory polarization of T lymphocytes.

Implications for clinical practice

What is the place for CHIP detection in the management of CVD?

Despite accumulating data, clonal hematopoiesis still needs to be validated as a full-fledged cardiovascular risk factor. To do this, observational studies are needed to reassess the prevalence of CHIP in the general population, as well as in patient populations seen in cardiology clinics worldwide. These studies should seek to confirm the link between the presence of CHIP, a higher inflammation level and a more pronounced atheroma volume.

The medical community will need to assess the best way to include CHIP in daily practice. The first question is how to select patients for CHIP screening. We could focus on patients who have a cardiovascular event without a high level of risk, based on traditional risk factors. This would allow us to better understand why the event happened, and give us tools to improve prevention of a recurrence alongside standard care. Another interesting population to screen could be patients with high concentrations of hsCRP, because they may represent a population with a high prevalence of CHIP. In order to let inflammation decrease and avoid an overestimation as a result of the acute cardiovascular event, hsCRP can be measured a few months after the event, when inflammation comes back to its basal level.[20] Finally, we could propose screening of patients presenting with a severe atheroma load, such as cases of multitroncular coronary artery disease or extensive peripheral artery disease. Screening for CHIP in the scope of primary prevention will be central in the future, if CHIP is verified as a risk factor with as much weight as smoking or dyslipidemia in the development of CVD. Given that CHIP could have a synergistic action with other traditional cardiovascular risk factors, it will be important to adjust current guidelines to account for this risk.

We have seen that screening for CHIP can be quite easy to perform in practice. It can fit to standard care requirements, in that the material is simple to get via a standard blood sample, the results are available within a few days and the costs are decreasing to the point where they will likely become affordable for routine practice.

The identification of CHIP in our daily practice will then raise the question of how to treat these patients.

Anti-inflammatory therapies

Recent studies have tested the hypothesis that anti-inflammatory molecules could improve the prognosis of people with CVD. In 2017, Ridker et al. studied the effect of canakinumab, a human monoclonal antibody targeting IL-1β, in a population of patients with a history of myocardial infarction who had an hsCRP blood concentration of ≥ 2 mg/L.[21] The team showed a decrease in the risk of cardiovascular deaths, non-fatal myocardial infarctions and non-fatal strokes in patients treated with the highest doses of canakinumab after nearly 4 years of follow-up. Canakinumab significantly decreased hsCRP blood concentrations. Interestingly, a subgroup analysis showed a trend towards a reduction in the risk of hospitalization for heart failure and heart failure-related mortality in patients who were receiving canakinumab.[22] These data suggest that targeting IL-1β and reducing the degree of inflammation may be an efficient way to reduce the risk of adverse cardiac events in secondary prevention.

Although canakinumab seems interesting in the prevention of CVD in patients at risk, it is not yet an approved treatment or used routinely in cardiology. Another study therefore evaluated the benefit of a widely used anti-inflammatory therapy – colchicine.[23] This drug is known to reduce inflammation through various mechanisms, in particular interference with NLRP3 inflammasome, resulting in reduction of IL-1β release, which in turn reduces the production of various cytokines and proinflammatory molecules, such as IL-6, tumor necrosis factor α and C-reactive protein.[24] In the COLCOT trial, Tardif et al. compared the effect of colchicine or placebo on prognosis in patients who had recently had a myocardial infarction. The authors showed a significant reduction in major adverse cardiovascular events, mainly driven by a decrease in the risk of stroke and the need for urgent coronary revascularization.

These data demonstrate the beneficial effect of targeting the immune system and reducing inflammation to prevent CVD. CHIP would be responsible for a low-grade inflammation that could favor CVD. Therefore, we can speculate that the efficacy of anti-inflammatory drugs could be mainly driven by an effect on CHIP carriers, not yet identified in these studies, who could be more receptive because of their proinflammatory state.[25] While we wait for more specific treatments, anti-inflammatory therapies may be one way to properly treat patients with CHIP.

Conclusions

CHIP is a common condition, particularly in the elderly, and is associated with an increased risk of CVD. Recent studies have suggested its implication in the pathogenicity of CVD, and models have shown that inflammation and dysregulation of the immune system are at the core of this involvement. As our knowledge grows, development of specific treatments will be a goal in order to move towards more personalized care and improve prevention of CVD.

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Mise à jour des dernières publications

Depuis la rédaction du précédent article, de nouvelles publications sont venues enrichir la littérature dédiée à l'hématopoïèse clonale de potentiel indéterminé. Nous avons intégré les principales références à la Table 2 de l'article précédent, que nous reproduisons page suivante. Nous y avons également intégré les principales études expérimentales concernant la CHIP.

Table. Overview of main studies regarding clonal hematopoiesis of indeterminate potential (prevalence, risk of cardiovascular disease and prognosis)

Clinical studies									
Year of publication	**First author**	**Number and origin of patients**	**Main outcome assessed**	**Prevalence**	**Main result**	**Adjustment for potential cofounders**	**Potential bias/limitations**	**Main genes concerned**	**Weight of CHIP on the risk of CVD**
2014	Jaiswal[1]	15,801 International cohorts	Detection of acquired somatic mutations in elderly without known hematologic disorders	*(VAF ≥ 3.5%, WES)* 60 to 69 y: 5.6% 70 to 79 y: 9.5% 80 to 89 y: 11.7% ≥ 90 y: 18.4%	**Increase in the frequency of somatic mutations with age**, from 5.6% between 60 and 69 years to 18.4% after 90 years **Increased risk of death**, mainly from cardiovascular causes	Age, sex, T2D	/	DNMT3A TET2 ASXL1	**HR for coronary heart disease of 2.0**, higher than hypertension (1.4), lower than age (2.4 after 60 years) and T2D (3.1) HR for ischemic stroke of 2.6
2014	Genovese[2]	12,380 Sweden	Detection of clonal expansion and somatic mutations in healthy people, analyses of subsequent medical outcomes	*(VAF ≥ 4%, WES)* 60-70 y: 6.5% 70-75 y: 12.5% ≥ 75 y: 16%	**Increase in the frequency of somatic mutations with age**, with 10.4% of people older than 65 years being concerned Increased risk of cancer and reduced overall survival	Age, sex	/	DNMT3A TET2 ASXL1 PPM1D	/

2017	Jaiswal[3]	8,255 International cohorts	Testing the hypothesis that CHIP increases the risk of atherosclerotic cardiovascular disease	*(VAF ≥ 4%, WES)* 70 y: 17% of CHIP in CHD vs 10% in non-CHD (HR=1.7) 60 y: 7% of CHIP in CHD vs 4% in non-CHD (HR=2) ≤50 y: 2% of CHIP in CHD, < 1% in non-CHD (HR=4)	Increased risk of CHD (HR of 1.9), early-onset MI (HR of 4.0)	Age, sex, T2D, smoking status, hyperlipidemia, hypertension	Case-control study	DNMT3A TET2 ASXL1	**Higher HR for coronary heart disease than age (1.0), smoking status (1.04), diabetes (0.9)** HR slightly lower than hypertension (2.01)
2019	Mas Peiro[4]	279 TAVR patients Germany	Prevalence and prognostic value of DNMT3A and TET2 mutation in patients undergoing TAVR	*(VAF ≥ 2%, NGS)* **33.3% of CHIP** 55–69 y: 25% 70–79 y: 25.4% 80–89 y: 34.7% 90-99 y: 52.9%	**Increased deaths following TAVR**	Age, sex, NT-proBNP	Monocentric	DNMT3A TET2	HR for death after TAVR of 4.81
2019	Dorsheimer[5]	200 CHF patients Germany	Prevalence and prognostic value of CHIP in patients with CHF	*(VAF ≥ 2%, NGS)* **18.5% of CHIP** 50-59 y: 10% 60-69 y: 21% 70-79 y: 27%	**Increased risk of death** (HR of 2.11) **and hospitalization for HF** (HR of 2.09)	Age, hypertension	Monocentric	DNMT3A TET2	Higher HR for death and hospitalization than age (0.70) and hypertension (0.95)
2020	Busque[6]	1887 patients (1359 CAD, 528 non-CAD) Montreal	Link between CHIP and hsCRP	*(VAF ≥ 2%, NGS)* **22.6% of CHIP** 25.3% in non-CAD 21.5% in CAD	**Higher hsCRP in CHIP-carriers than in non-CHIP**	Age, sex, BMI	/	DNMT3A TET2 ASXL1	/

2020	Assmus[7]	419 CHF patients Germany	Prognostic role of clone size DNMT3A and TET2	*(VAF ≥ 0.5%, NGS)* 52.6% of CHIP	**Poorer prognosis with DNMT3A VAF ≥ 1.15% and TET2 VAF ≥ 0.73%**	Age, blood pressure	/	DNMT3A TET2	/
2020	Guermouche[8]	89 healthy patients (18-80 y) Tours	Prevalence of CHIP in healthy individuals BM and PB samples	*(VAF ≥ 2%, NGS)* 17% of CHIP **50-60 y: 30%** **60-70 y: 40%**	/	/	Monocentric, small cohort	DNMT3A TET2	/
2021	Pascual-Figal[9]	62 CHF patients	Impact of CHIP on HF progression	*(VAF ≥ 2%, NGS)* **39% of CHIP** 60-67 y: 20% 68-83 y: 40% ≥ 84 y: 65%	**More events in CHIP-carriers** (HF-related deaths and hospitalizations)	Age, sex, ischemic etiology, LVEF, NT-proBNP levels	Monocentric, small cohort	DNMT3A TET2	HR for death = 2.79 HR for death+HF = 3.8 HR for HF-death or HF-hospitalization = 4.4
2021	Haring[10]	8709 women without CVD or cancer WHI	Association between healthy lifestyle and prevalence of CHIP	*(VAF ≥ 6%, NGS)* **8.6% of CHIP**	Higher healthy lifestyle score not associated with CHIP **Obesity and smoking associated with more CHIP**	Multivariable adjustment	/	DNMT3A TET2	/
2021	van Zeventer[11]	621 patients > 80 years Netherlands	Prevalence of CHIP Association with comorbidities Prognosis	*(VAF ≥ 1%, NGS)* **62% of CHIP** Increasing with age	**No difference according to history of myocardial infarction** Positive association with COPD	Age, sex	Weak definition of MI	DNMT3A TET2	**No higher risk of death in patients with CHIP**, except for patients without DNM3A and/or TET2 mutation

(continued next page)

Experimental studies				
Year of publication	**First author**	**Model**	**Main outcome assessed**	**Main results**
2017	Fuster[12]	Atherogenic mice transplanted either with 10% WT-BMT cells or 10% KO-BMT (TET2 mutation)	Effect of TET2 mutation on clonal expansion, atherosclerosis, inflammation	Clonal expansion of TET2 mutated cells **Greater atherosclerosis plaque** **Higher NLRP3 inflammasome mediated IL-1ß secretion**
2018	Sano[13]	Mice transplanted either with 10% WT-BMT cells or 10% KO-BMT (TET2 mutation)	Contribution of TET2 mutation on HF	Clonal expansion of TET2 mutated cells **Lower LVEF, increased LV size, more myocardial hypertrophy and fibrosis** Higher NLRP3 inflammasome mediated IL-1ß secretion within the heart **All differences corrected by an NLRP3-inflammasome inhibitor**
2018	Sano[14]	Deficient DNMT3A or TET2 mice compared to controls	Effect of DNMT3A and TET2 on HF	Clonal expansion of DNMT3A and TET2 **Lower LVEF, increased LV size, more myocardial hypertrophy and fibrosis** (DNMT3A and TET2) Higher NLRP3 inflammasome mediated IL-1ß secretion within the heart (TET2)
2020	Sano[15]	Deficient JAK2 mice compared to controls	Effect of JAK2 on HF	Clonal expansion of JAK2 **Higher production of IL-1ß, IL-6, TNFα** Lower LVEF, increased LV size, more myocardial hypertrophy and fibrosis
2021	Heyde[16]	Atherogenic TET2-KO mice compared with non-atherogenic TET2-KO mice	Effect of atherosclerosis on clonal hematopoiesis	**Accelerated clonal expansion of TET2 in atherogenic mice**

BM: bone marrow, BMI: body-mass index, BMT: bone-marrow transplant, CHD: coronary heart disease, CHF: chronic heart failure, CHIP: clonal hematopoiesis of indeterminate potential, COPD: chronic obstructive pulmonary disease, CVD: cardiovascular diseases, HF: heart failure, HR: hazard-ratio, hsCRP: high-sensitive C-reactive protein, KO: knock-out, LVEF: left ventricular ejection fraction, MI: myocardial infarction, NGS: next generation sequencing, NLRP3: nucleotide-binding oligomerisation domain-like receptor family pyrin domain containing 3, PB: peripheral blood, TAVR: transcatheter aortic valve replacement, T2D: type 2 diabetes, VAF: variant allele frequency, WES: whole-exome sequencing, y: years in mean, WHI: Women Health Initiative

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Frequency of clonal hematopoiesis in patients over 75 years with a first cardiovascular event.

Consequences on inflammation and atherosclerosis

L'article qui suit présente les résultats préliminaires de l'étude CHAth (Corrélation entre l'Hématopoïèse clonale, les maladies cardiovasculaires, l'inflammation et l'Athérome). Il s'agit d'une étude prospective, promue par le CHU de Bordeaux et ayant bénéficié d'un financement de la fondation Cœur et Recherche en 2018. Elle est le fruit d'un partenariat entre les services de Maladies coronaires et vasculaires et d'Hématologie biologique du CHU de Bordeaux, en collaboration avec le Centre d'Investigations Cliniques. L'idée de ce projet est née des récentes données de la littérature suggérant l'existence d'un lien entre l'hématopoïèse clonale de potentiel indéterminé (CHIP) et les maladies cardiovasculaires, liens probablement sous-tendus par des mécanismes pro-inflammatoires et athéromateux.
Ce travail a pour but d'estimer la prévalence de CHIP dans une population de patients âgés d'au moins 75 ans et présentant un premier infarctus du myocarde, et de corréler la présence de CHIP à l'inflammation systémique et à la charge en athérome. Pour ce faire, un séquençage de l'ADN était réalisé, et certains marqueurs de l'inflammation étaient dosés (CRP ultra-sensible, IL-1ß, IL-6, IL-10, TNFα). La quantification du volume athéromateux était effectuée par mesure 3D au cours d'un Doppler des troncs supra-aortiques, et était associée à la mesure des index de pression systolique de cheville (IPS) et au recensement des sténoses carotidiennes et coronaires significatives.

Notre rôle était de participer au repérage des patients éligibles au moment de l'événement cardiovasculaire initial, à la programmation du rendez-vous d'inclusion, ainsi qu'au recueil et à l'inscription des données dans un cahier de recueil informatisé. Nous avons également participé à la réalisation des analyses statistiques et avons assuré la rédaction du présent article. Enfin, la rédaction d'une revue de la littérature nous a permis d'appréhender de façon globale la problématique de l'hématopoïèse clonale et de poser les bases de réflexion sur ce projet transversal.

Au moment de la rédaction du présent article, portant sur 106 patients, les inclusions se poursuivaient en vue de la constitution d'une cohorte de 140 patients. L'analyse de certains marqueurs inflammatoires (IL-1ß, IL-6, IL-10, TNFα), des IPS et des sténoses carotidiennes significatives n'étaient pas encore terminée. Les résultats définitifs feront l'objet d'une future publication.

List of abbreviations

ACEI: angiotensin-converting-enzyme inhibitor
ARB: angiotensin II receptor blocker
ASXL1: additional sex combs-like 1
AV: atrioventricular
BCOR: BCL6 corepressor
BCORL1: BCL6 corepressor like 1
BMI: body-mass index
c-HDL: cholesterol-high density lipoprotein
c-LDL: cholesterol-low density lipoprotein
CABG: coronary-artery bypass graft
CAC: coronary-artery calcification
CAD: coronary-artery disease
CBL: casitas B-lineage lymphoma
CCA: common carotid artery
CCS: Canadian Cardiology Society
CHIP: clonal hematopoiesis of indeterminate potential
CV: cardiovascular
CVD: cardiovascular disease
DNA: deoxyribonucleic acid
DNMT3A: DNA methyltransferase 3A
ECG: electrocardiogram
ESC: European Society of Cardiology
GNB1: guanine nucleotide-binding protein subunit beta-1
HSC: hematopoietic stem cell
hsCRP: high-sensitivity C-reactive protein
ICU: intensive care unit
IL: interleukin
JAK-2: Janus kinase 2
LVEF: left ventricular ejection fraction
MAF: minor allele frequency
MI: myocardial infarction
NFE2: nuclear factor, erythroid 2
NGS: next-generation sequencing
NYHA: New-York Health Association
PCI: percutaneous coronary intervention
PPM1D: protein phosphatase, Mg^{2+}/Mn^{2+}-dependent 1D
SAMD9: sterile alpha motif domain containing 9
SAMD9L : sterile alpha motif domain containing 9 like
SARS-Cov2: severe acute respiratory syndrome coronavirus 2
SAT 3D US: supra-aortic trunks with 3D measurement ultrasonography
SF3B1: splicing factor 3B subunit 1
SNP: single-nucleotid polymorphism
SRSF2: serine/arginine-rich splicing factor 2
TAVR: transcatheter aortic valve replacement
TET2: ten-eleven translocation 2
TNFα: tumor necrosis factor
TP53: tumor protein 53
TTE: transthoracic echocardiography
VAF: variant allelic fraction
ZRSR2: zinc finger CCCH-Type, RNA binding motif and Serine/Arginine Rich 2
3D: three-dimensional

Introduction

Atherothrombosis is the main cause of death worldwide. Traditional cardiovascular (CV) risk factors, such as age, diabetes, smoking, dyslipidemia, hypertension, allow us to explain 70 to 85% of CV events our patients suffer, but a significant part of these events remains unexplained by traditional risk factors, given that 15 to 30% of people without any of these risk factors can have an atherosclerotic CV event, whereas a high-risk patient do not automatically experience such an event.[1]

In a search for other risk factors or actors of pathogenesis of atherothrombosis, clonal hematopoiesis of indeterminate potential (CHIP) has emerged as potentially correlated with cardiovascular diseases (CVD).[2] This condition results from the acquisition of a somatic mutation in leukemia-driver genes and corresponds to the clonal expansion of a population of hematopoietic stem cells without any clinical or biological sign of hematologic malignancy.[3] Mutations observed in CHIP frequently involve epigenetic regulators, the main ones being DNMT3A, TET2 and ASXL1.

CHIP has been linked to CVD through mechanisms involving the immune system. Indeed, it has been shown in murine models that clonal hematopoiesis exacerbated inflammatory phenotype of macrophages infiltrating the atherosclerotic plaques, with an over-production of various cytokines, especially IL-1ß.[4] This favored recruitment of monocytes within the plaque, production of foam cells and growth of the plaque. Thus, various studies have shown that CHIP was associated with a higher level of inflammation and a higher atherosclerotic burden in mice. [4][5]

Two main studies over the last years have estimated the prevalence of CHIP in the general population using whole-exome sequencing techniques. Rare before 40 years of age, prevalence of CHIP then increases with age to reach 10 to 20% of the general population after 70 years, when taking into account variant allelic frequency (VAF) of at least 2% in the peripheral blood.[2][6] If CHIP has been associated to an increased risk of myocardial infarction, few studies have prospectively evaluated the prevalence of CHIP in cohorts of people suffering from coronary artery disease (CAD), results of which are controversial.[5][7] Indeed, although Jaiswal et al. found an 1.9 fold increased risk of CAD when carrying CHIP, Busque et al. found no difference between CHIP an non-CHIP carriers regarding prevalence of CAD.

To our knowledge, although CHIP has been shown to favor atherosclerosis development, no study has yet evaluated the correlation between CHIP and atherosclerotic burden in human. Last years, several lines of evidence have shown that ultrasonography with

3D measurement of atherosclerotic volume was a reliable and reproducible method to evaluate atherosclerotic volume on carotid and femoral arteries, and that this volume is well correlated with global CV risk.[8][9]

We conducted a prospective study to evaluate the prevalence of CHIP in a population of patients aged at least 75 years suffering their first myocardial infarction. This prospective design also allowed us to evaluate the inflammation level and the atheroma volume at the closest of the event, and to correlate it with the presence of CHIP.

Methods

Patients

Patients were enrolled between March, 2019, and June 2021, 2 to 7 months after being admitted in the intensive care unit (ICU) of the cardiologic department of the Bordeaux University Hospital for a myocardial infarction.

The two main eligible criteria were having a first myocardial infarction from atherosclerosis origin without evidence of hematologic malignancy.

Eligible patients were ≧ 75 years and were admitted for a first acute coronary event, without any previous other CV event in their history. According to guidelines, myocardial infarction was defined as a myocardial injury with clinical evidence of myocardial ischemia, with significant change in cTroponin levels above the 99th percentile and at least one criteria of the following: symptoms of myocardial ischemia, ischemic ECG changes, development of pathological Q waves, imaging evidence of loss of viable myocardium or new regional wall motion abnormality, identification of a coronary thrombus by angiography.[10] Type 1 myocardial infarctions, and type 2 and 3 if there was evidence of an atherosclerotic coronary disease were included. Myocardial infarctions due to vasospasm, microvascular dysfunction, non-atherosclerotic coronary dissection, oxygen supply/demand imbalance alone were excluded.

The other inclusion criterion was the absence of evidence of hematologic malignancy, known or revealed by blood count at admission at the time of the event. Blood count at admission had to be normal. In case of abnormality on this first blood count, we ran complementary analysis to search for a non-neoplastic cause (hemoconcentration, dilution, inflammation due to an active and reversible condition, vitamin or iron deficiency, bleeding not related to an active neoplasia). If no reversible cause was found and/or if blood count did not normalize during the hospital stay, the patient was excluded. Hemoglobin was normal if

between 12 g/dL and 16 g/dL in women, and 13 g/dL and 17 g/dL in men; platelets count had to be between 150 and 400 G/L, and white blood cells were expected to be between 4 and 10 G/L. Finally, the patient to be affiliated to a social security regimen. Written informed consent was required and obtained before inclusion.

Exclusion criteria encompassed the absence of known atherosclerotic coronary disease, an uncontrolled diabetes (HbA1c > 10%), a previous atherosclerotic CV event before 75 years (myocardial infarction, ischemic stroke, peripheral arterial disease, angina pectoris) in order to compare the most reliably possible the patients at their entry in the disease, a hematologic malignancy (known or revealed by blood count at admission), a known chronic inflammatory disease (rheumatoid arthritis, arteritis, gastro-intestinal illness), an active neoplasia, a long-term anti-inflammatory therapies. Patients who were under legal protective measures, deprived of liberty by court order, unable to give their written informed consent, or having participated to an interventional study on a drug within 30 days before enrolment were ineligible to our study.

The study was registered on the clinicaltrial.gov website under the number NCT04581057, and was approved by a national ethics committee.

Study procedures

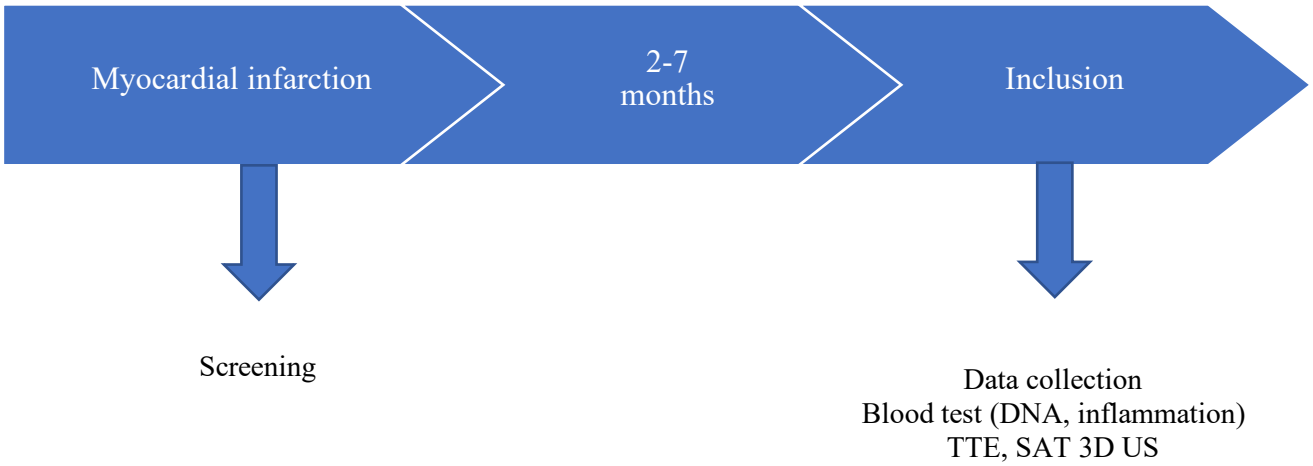
Eligible patients were screened at the time of their admission for myocardial infarction in the ICU (Figure 1). They were informed about the study, and they were included between 2 to 7 months after the acute event, at the time of the appointment for the post-MI clinical assessment.

During this appointment, eligibility was checked again, written informed consent was obtained and eligible patients could be enrolled in the study. All data were collected at this time. Patients were informed that they could withdrew consent and leave the study at any time.

Collected data

All data were collected on a secured e-case report form, and were systematically double-checked by the investigator and a clinical research assistant.

Figure 1. Course of the study



DNA: deoxyribonucleic acid, SAT 3D US: supra-aortic trunks with 3D measurement ultrasonography, TTE: transthoracic echocardiography

Data from the index event

Patient's characteristics (age, gender, body mass index) and traditional alterable CV risk factors (smoking status, hypertension, dyslipidemia, diabetes) were collected. Smoking was defined as having an active smoking activity or having stopped within the last three years. Hypertension was considered either if written in medical records, or defined by a systolic blood pressure of at least 140 mmHg and/or a diastolic blood pressure of at least 90 mmHg from the means of daily measures during the hospital stay, or by the use of anti-hypertensive drugs. Dyslipidemia was defined as a c-LDL > 4.1 mmol/L (> 1.6 g/L) and/or c-HDL < 1.03 mmol/L (< 0.4 g/L) in men and < 1.29 mmol/L (< 0.5 g/L) in women, or by the use of lipid-lowering drugs.[11] A threshold of HbA1c > 6.5% was used to define diabetes, as well as use of anti-diabetic agents.[12] Drugs used to be taken at the time of the event (antiplatelet agents, lipid lowering agents, anti-hypertensive drugs, anti-diabetic drugs) were recorded.

Data concerning myocardial infarction were collected and comprised: existence of a ST-elevation; concerned territory; number of main vessels with a stenosis > 50%, according to the morphological assessment carried out by the cardiologist performing the coronary angiography and written in the report; acute phase treatment (medical treatment, fibrinolysis, angioplasty, coronary artery bypass graft surgery); presence and type of potential complications: heart failure (HF) defined according to the 2016 ESC guidelines[13], ventricular arrythmia defined as sustained ventricular tachycardia or ventricular fibrillation, high-degree atrioventricular block, pericarditis, mechanical complication comprising ischemic mitral regurgitation, ventricular free-wall rupture and ventricular septal defect, intra-cardiac thrombosis diagnosed by echocardiography.

Biological data at the time of the initial admission were recorded (blood count, high-sensitive CRP (hsCRP), creatinine, lipid profile, HbA1c). Finally, the list of drugs that were prescribed at discharge was collected.

Data at inclusion

Clinical and routine biological testing

First, we ensured that patients were still eligible by checking inclusion and exclusion criteria again. We established and checked a list of conditions that, if present, had to make the appointment reported in order not to skew biological and genetic data (Figure S1 in the Appendix). We also ensured that patients had not been vaccinated against SARS-Cov2 within the 8 previous days.

We ran a routine CV evaluation. Patients were asked about presence of dyspnea or angina, evaluated with NYHA and CCS scales. Information was obtained from medical record about potential recurrence of a CV event since the index event (new myocardial infarction, coronary revascularization, stroke, hospitalization for acute HF).

Biological exams were done in the same laboratory, comprising blood count, hsCRP, lipid profile, HbA1c.

Imaging data

Transthoracic echocardiography was performed by the same CV center and left ventricular ejection fraction (LVEF) was recorded.

Supra-aortic trunks ultrasonography with 3D-measurement of atherosclerotic volume was performed by trained physicians, using a Philips iU22 probe equipped with a linear-3D volume convertor VL13-5 (Philips). Images were then analyzed using the Vascular Plaque Quantification software on the QLAB 10.2 system by Philipps, used by echographs equipping the laboratory of echography of the hospital. Ultrasonography with 3D-measurement has been shown to be a reliable, sensitive and reproductive technique, allowing us to calculate the atheroma volume of common and internal carotids.[14] Quantification of atherosclerosis burden in femoral or carotid arteries has been shown to be reliably correlated to global CV risk estimated by traditional risk factors, and carotid plaque burden was shown to be well correlated to the coronary-artery calcification (CAC) score in the prediction of CV events.[8][15]

Functional ischemic testing was performed at the clinicians' discretion.

Mutations and inflammatory markers

A blood sample of 30 mL was drawn and immediately sent within to the Biological Cancer Resources Center of the Bordeaux University Hospital, a certified platform for utilization of biological samples for research. At reception, blood samples were centrifugated and freezed at -20°C (circulating leukocytes for DNA extraction) and -80°C (plasma). Patients were informed that they could oppose to their blood sample storage at any time.

The search for somatic mutations was carried out in the Hematology laboratory of the Bordeaux University Hospital. Blood samples were processed into whole blood. The DNA from the total leukocytes was extracted using Maxwell RSC Instrument (Promega).

The Next Generation Sequencing (NGS) panel used was the panel of genes classically sequenced in the Hematology laboratory of the Bordeaux University Hospital for the diagnosis

and follow-up of myeloid hemopathies. The panel of genes tested is detailed in the Appendix (Table S1). Libraries were prepared using Sure Select XT Low Input kit (Agilent) or using Magnis NGS Prep System (Agilent). Sequencing was performed on a NextSeq 550Dx Instrument (Illumina technology). Bioinformatics analysis was performed according to an in-house pipeline. The sequences were aligned to the reference genome (hg19).

Only variants with a VAF greater than 1% were retained. Single nucleotide polymorphism (SNP) listed in databases (dbSNP, GnomAD, ExaC, 1000 genome, NCI60, Exome Sequencing Project...) were excluded when they presented a minor allele frequency (MAF) > 1% (frequency at which the allele is found in general population). Synonymous mutations and/or mutations impacting a non-coding region were not retained.

CHIP was defined by the presence of at least one mutation in one of the 56 genes tested, with a VAF of at least 2%.[3] For further analyses we took into account different VAF: 1%, 5% and 10%, threshold above which CV risk seems to be significantly increased.[7]

Inflammatory markers (IL-1b, IL-6, IL-10, TNF-a) were analyzed by Bioplex200 technology from the same blood sample. These cytokines have been shown to be involved in the hypothetical pathophysiologic pathway linking CHIP with atherosclerosis.[4][16][17]

At the end of these exams, the patient's participation to the study was over and the patient could go on with his usual CV follow-up.

Aims of the study

The primary goal was to evaluate the prevalence of CHIP in the population of people aged at least 75 years and suffering from their first myocardial infarction.

The secondary goals were the comparison with the known prevalence of CHIP in the general population of the same age, the assessment of inflammation level based on concentration of various markers (hsCRP, IL-1b, IL-6, IL-10, TNF-a) compared between CHIP and non-CHIP carriers, the quantification of atheroma volume with 3D ultrasound analyses compared between CHIP and non-CHIP carriers, and the correlation of inflammation level and atherosclerosis burden with VAF. Influence of CHIP on the risk of recurrence of CV event after hospital discharge was evaluated. Correlation between presence or absence of CHIP and the number of coronary arteries concerned by significant stenosis was also assessed, as well as relationship between CHIP and traditional risk factors. Finally, we studied whether the type of genes involved had an influence on these parameters.

Statistical analyses

The primary goal was to evaluate the prevalence of CHIP in a cohort of patients aged at least 75 years admitted for a first myocardial infarction. One of the secondary goals was the comparison with the known prevalence of CHIP in the general population of the same age. Based upon studies having estimated the prevalence of CHIP in people from the general population aged at least 75 years at 16%, and taking into account a relative-risk of having a CAD when being CHIP-carriers of 1.9, the expected prevalence in our cohort was 30%.[(6)(7)] We calculated that a sample size of 87 patients was needed to achieve 90% power, with a type-I error of 5%.

All comparisons were made with a type I error risk of 5%. Qualitative variables were compared using Chi-square test or Fisher test. Comparison of the observed prevalence with the known prevalence in the general population was made using a z-test for one proportion. Logistic regression models could be used to take into account the adjustment variables if necessary. Quantitative variables were compared using the Student test when the test validity conditions were met (normal distribution, homogeneous variances). When the distribution was not normal, a Mann-Whitney test was used. Linear regression models were used to take into account the adjustment variables. Correlation between quantitative variables were analyzed by the correlation test. Pearson's correlation test was used if the test validity conditions were met (normal distribution, homogeneous variances). If the distribution was not normal, we used a Spearman correlation test. Missing values were managed by excluding the patient from the analysis of the concerned variable.

Analyzes were made using the R software.

Results

Patients & myocardial infarction data

Between April 2019 and June 2021, 106 patients were included. The characteristics of the patients at the time of the event are presented in Table 1.

Table 1. Characteristics of the population at the time of the event depending of the CHIP status (all CHIP mutations, VAF ≥ 2%)

	All patients n=106	Patients without CHIP n=49 (46%)	Patients with CHIP n=57 (54%)
Male, n (%)	69 (65%)	34 (69%)	35 (61%)
Age, years	82.0 [79.0-87.0]	81 [78.0-85.0]	82 [80.0-87.0]
Risk factors			
BMI, kg/m^2	25.1 [22.7-27.5]	25.6 [23.1-27.5]	25.1 [22.5-27.5]
Diabetes, n (%)	33 (31%)	20 (41%)	13 (23%)
HbA1c, %	5.90 [5.60-6.60]	6.15 [5.70-7.07]	**5.70 [5.50-6.10]***
Hypertension, n (%)	80 (76%)	37 (76%)	43 (77%)
Dyslipidemia, n (%)	68 (65%)	16 (33%)	21 (38%)
Smoking, n (%)	3 (2.9%)	2 (4.2%)	1 (1.8%)
≥ 2 CV risk factors, n (%)	68 (65%)	35 (71%)	33 (59%)
Myocardial infarction			
ST-segment elevation, n (%)	58 (55%)	26 (54%)	32 (56%)
Coronary angiography performed, n (%)	104 (98%)	47 (96%)	57 (100%)
Multitroncular lesions, n (%)	50 (48%)	19 (40%)	31 (54%)
Acute phase treatment			
PCI, n (%)	92 (87%)	42 (86%)	50 (88%)
Medical therapy, n (%)	9 (8.5%)	5 (10%)	4 (7%)
PCI and CABG, n (%)	3 (2.8%)	1 (2.9%)	2 (3.5%)
CABG, n (%)	1 (0.9%)	1 (2%)	0 (0%)
Fibrinolysis and PCI, n (%)	1 (0.9%)	0 (0%)	1 (1.8%)

(continued next page)

Acute phase complications before hospital discharge			
All complications°, n (%)	45 (42%)	20 (41%)	25 (44%)
Heart failure, n (%)	39 (37%)	18 (37%)	21 (37%)
Ventricular arrythmia/AV block, n (%)	7 (6.6%)	3 (6.1%)	6 (11%)
Thrombus, n (%)	4 (3.8%)	2 (4.1%)	2 (3.5%)
Pericarditis/Tamponade, n (%)	3 (2.8%)	1 (2%)	2 (3.5%)
Blood test			
Hemoglobin, g/dL	13.60 [12.50-14.30]	13.6 [12.8-14.6]	13.4 [12.1-14.1]
White blood cells, G/L	9.10 [7.30-11.20]	9.40 [7.40-11.20]	8.85 [7.30-11.20]
Neutrophils, G/L	6.75 [5.00-8.43]	6.80 [4.70-8.40]	6.70 [5.60-8.70]
Platelets, G/L	218 [186-265]	218 [176-262]	226 [194-269]
Creatine, µmol/L	77.5 [67.0-101]	81 [67-109]	76 [67-96]
Medical treatment at discharge			
Aspirin, n (%)	94 (95%)	45 (98%)	49 (92%)
P2Y12 receptor inhibitor, n (%)	88 (89%)	39 (85%)	49 (92%)
ACEI, n (%)	75 (76%)	36 (78%)	39 (74%)
ARB, n (%)	14 (14%)	4 (8.7%)	10 (19%)
Statin, n (%)	94 (95%)	42 (91%)	52 (98%)
Beta-blocker, n (%)	92 (93%)	42 (90%)	50 (94%)

(legend next page)

ACEI: angiotensin-converting-enzyme inhibitor, ARB: angiotensin II receptor blocker, AV: atrioventricular, BMI: body-mass index, CABG: coronary-artery bypass graft, CHIP: clonal hematopoiesis of indeterminate potential, CV: cardiovascular, PCI: percutaneous coronary intervention. Results are expressed in median and interquartile ranges for quantitative variables, and numbers and percentages for qualitative variables. For each variable, results do not include patients with missing values.

°One patient could suffer more than one complication, all complications were recorded

* Statistically significant ($p<0.05$)

Forty-nine patients (46%) had an abnormality on blood count at admission, either on white blood cells, hemoglobin or platelets. Twenty-four patients had anemia, twenty-two patients had leukocytosis, and three had low platelets. All of these patients had complementary analysis to search for a reversible cause. Among patients with anemia, 11 had fluid overload, 13 had iron deficiency, three had folates deficiency and two had cyanocobalamin deficiency. Some patients had more than one cause of anemia. Leukocytosis was explained either by inflammation due to the myocardial infarction, pneumonia (3 patients), gout (2 patients) or urinary tract infection (1 patient), and all cases normalized during the hospital stay. The three platelets count abnormalities normalized during the hospital stay, one was due to a platelets aggregate. Considering the entire cohort, the median levels of hemoglobin, platelets and white blood count were normal.

CHIP status and acute phase characteristics

Here, we considered as CHIP carriers patients with at least one mutation on the sequenced 56 genes with a VAF ≥ 2%.

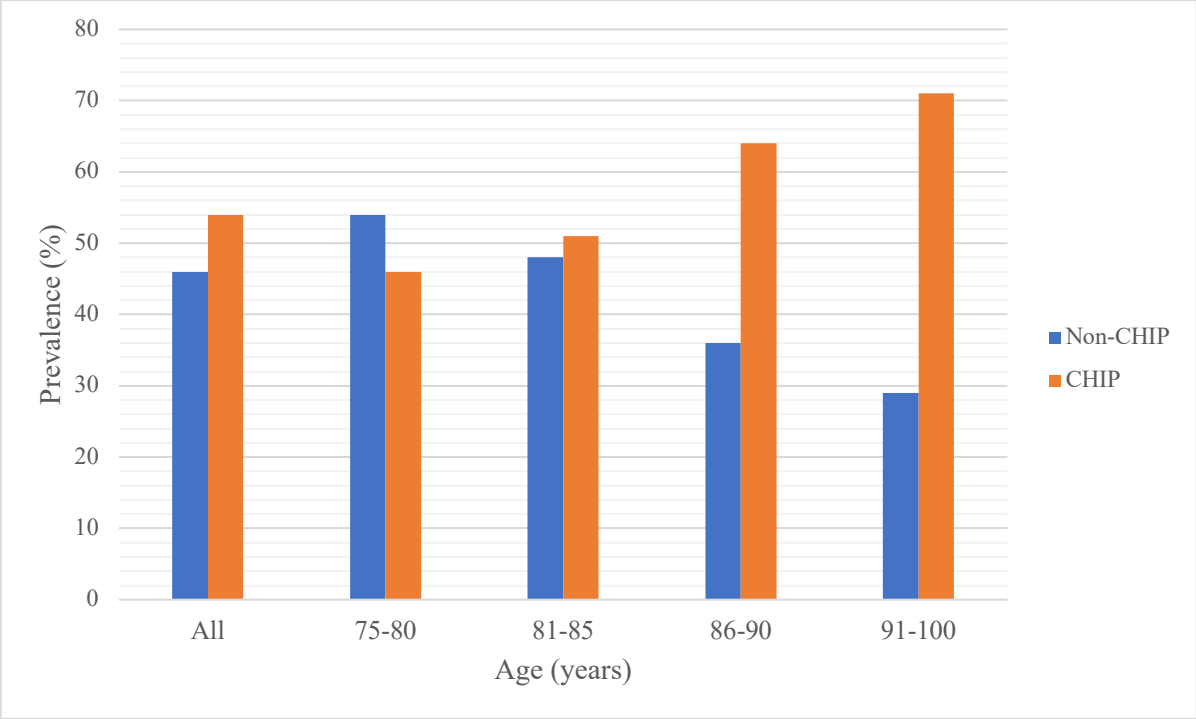
Fifty-seven patients (54%) of the cohort were CHIP carriers. This prevalence was much higher than the one we expected, and significantly higher than the prevalence described in the general population (z =10.7, p<0.0001).

Little differences were observed between CHIP and non-CHIP carriers (Table 1). Patients with CHIP were slightly older (p=0.13). They were less susceptible to have diabetes (p=0.053), and had lower HbA1c (p<0.01). We observed a trend toward a less at-risk profile in CHIP carriers, evaluated by the cumulation of at least two traditional risk factors (p=0.18). There was no major difference between the two groups regarding acute phase complications or biological data.

Figure 2 shows the prevalence of CHIP in the global cohort and according to age. Prevalence tended to increase with age, with 46% of patients aged between 75 and 80 years having a CHIP, 51% of the patients aged 81 to 85 years, 64% of the patients between 86 and 90 years and 71% of the patients after 91 years of age.

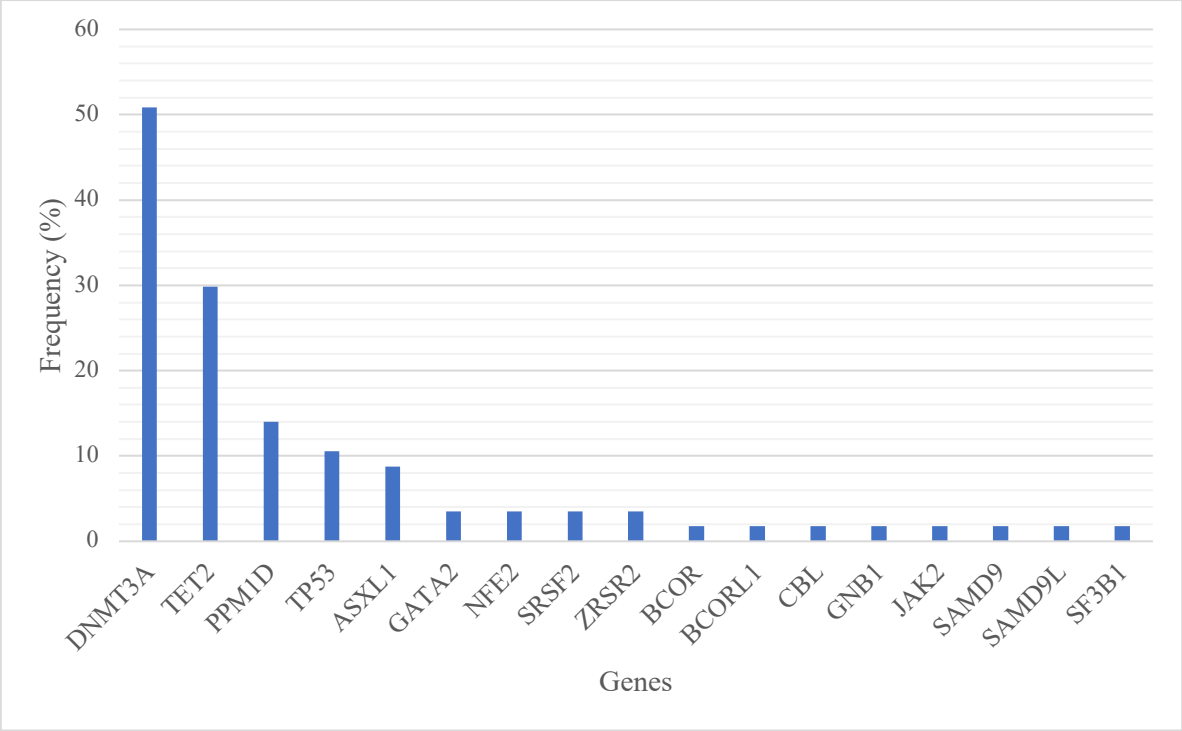
DNMT3A was the gene most likely to be concerned by CHIP, with 51% of the patients having a CHIP presenting at least one mutation of DNMT3A. TET2 was concerned in 29.8% of the cases, before PPM1D (14%), TP53 (10.5%) and ASXL1 (8.8%). All mutated genes are presented in Figure 3. Overall, 41% of the entire cohort were DNMT3A and/or TET2 carriers.

Figure 2. Prevalence of CHIP among patients aged at least 75 years and suffering from myocardial infarction (all CHIP mutations, VAF ≥ 2%)



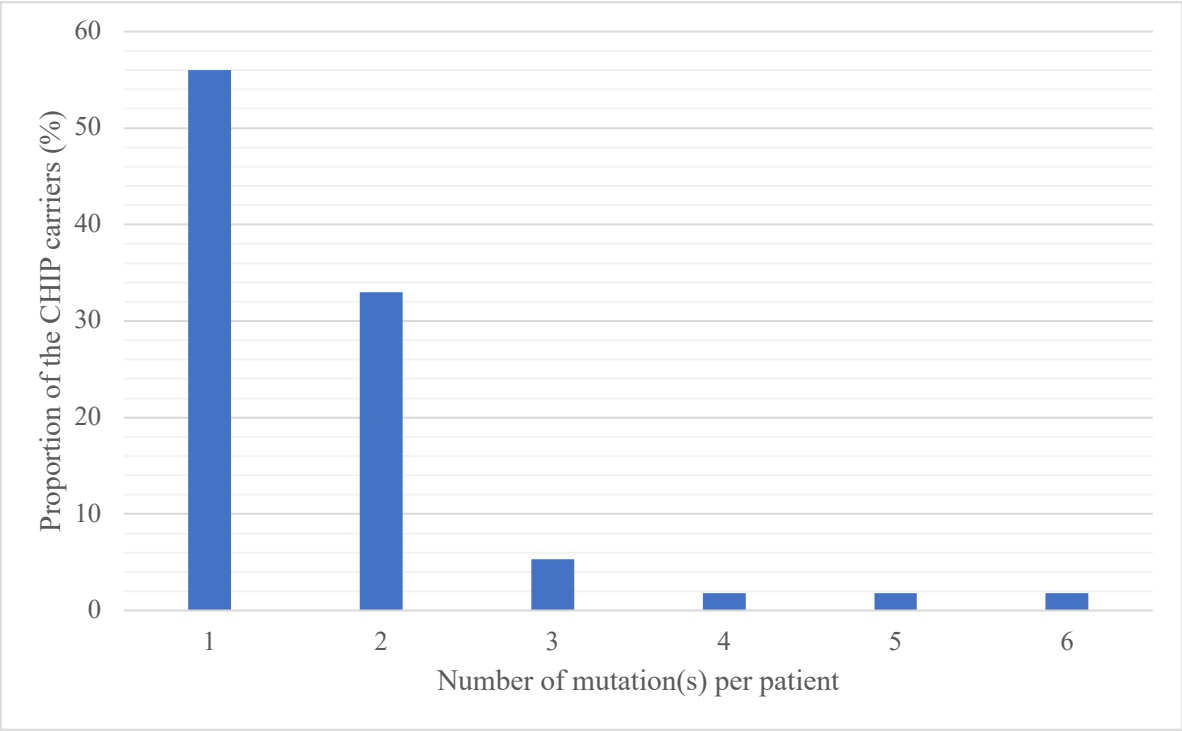
CHIP: clonal hematopoiesis of indeterminate potential, VAF: variant allele frequency

Figure 3. Frequency of mutated genes among patients with CHIP (all CHIP mutations, VAF ≥ 2%)



CHIP: clonal hematopoiesis of indeterminate potential, VAF: variant allele frequency

Figure 4. Number of mutation(s) per patient (all CHIP mutations, VAF ≥ 2%)



CHIP: clonal hematopoiesis of indeterminate potential, VAF: variant allele frequency

Most of the patients only had one mutation (56%). Thirty-three percent had two mutations, and 11% had three or more mutations (Figure 4). Among patients with CHIP, 30 patients (56%) had a VAF ≥ 5% and 21 patients (39%) had a VAF ≥ 10%.

When focusing on elderly patients carrying CHIP (≥ 85 years of age), we found that they were more likely to have bigger clone size than younger patients, although these differences were not significant: they were 41% to have CHIP with VAF of at least 10%, compared with 31% of the younger patients (p=0.47). Fifty percent of them carried two or more CV risk factors compared with 65% of the younger patients (p=0.27) and 71% of the non-CHIP carriers of the same category of age (p=0.08).

CHIP status and inflammatory markers, atheroma burden, and complications

All CHIP (Table 2)

We first considered patients with any CHIP at VAF of at least 2%.

We found no significant difference regarding hsCRP when comparing CHIP and non-CHIP carriers, although we noticed a trend towards an increased value (2.80 [1.25-6.00] *v.* 1.20 [0.70-2.70], p=0.35, Table 2 and Figure 5). The data was missing in 38 patients of the cohort. When evaluating the influence of VAF on hsCRP levels in patients with CHIP, we found that CHIP carriers at VAF ≥ 5% (31 patients) had significantly higher hsCRP levels than non-CHIP carriers (3.60 [1.30; 6.00] *v.* 1.20 [0.70-2.70], p=0.017). After adjustment on age, sex and presence of at least two traditional risk factors, we found no statistically significant difference although the level of correlation was high (Table 4). There was no significant difference regarding hsCRP between CHIP carriers at VAF ≥ 1% and 10% and non-CHIP carriers.

Analysis of supra-aortic trunks ultrasonography with 3D-measurement of atherosclerotic volume was available in 34 patients of the cohort. The main reasons why it was not performed or analyzed in other patients were a too important or calcified atheroma, and technical issues that impede transfer and analyses of the images. We found no statistically significant difference between CHIP carriers at VAF ≥ 2% and non-CHIP carriers regarding atherosclerosis burden, although we noticed a trend towards a higher rate of multitroncular coronary lesions in patients with CHIP (relative risk of 1.4, p=0.16), as well as a higher carotid atheroma volume (p=0.36). We found no statistically significant difference between CHIP carriers and non-CHIP carriers when taking into account other VAF, although the difference between the two groups regarding the rate of multitroncular lesions tended to increase at VAF ≥ 5% (61% *v.* 40%, p=0.07).

Table 2. Characteristics at inclusion and secondary analyzes (CHIP carriers with a VAF ≥ 2%)

	Non-CHIP carriers (n=49)	CHIP-carriers° (n=57)	CHIP-carriers except DNMT3A and/or TET2 carriers° (n=14)
Time from index event, months	5.00 [4.00-6.00]	4.00 [3.00-5.75]	4.00 [4.00-5.75]
Age, years	81.00 [78.00-85.00]	82.00 [80.00-87.00]	83.5 [78.50-87.00]
Male, n (%)	31 (63%)	38 (67%)	10 (71%)
Blood tests			
Hemoglobin, g/dL	13.10 [12.50-14.30]	12.70 [12.10-13.60]	13.00 [12.40-14.50]
White blood cells, G/L	6.40 [5.50-7.50]	6.70 [5.65-7.50]	5.85 [5.50-7.20]
Neutrophils, G/L	4.15 [2.75-4.92]	4.00 [3.20-4.70]	3.75 [3.18-4.28]
Platelets, G/L	205 [155-242]	199 [174-253]	190 [162-206]
Total cholesterol, g/L	1.45 [1.22-1.68]	1.55 [1.23-2.13]	1.46 [1.13-1.70]
c-LDL, g/L	0.76 [0,63-0.83]	**0.85 [0.63-1.09]***	0.86 [0.57-1.04]
c-HDL, g/L	0.45 [0.39-0.54]	0.46 [0.41-0.54]	0.47 [0.39-0.56]
HbA1c, %	6.10 [5.80-6.60]	**5.90 [5.70-6.20]***	5.70 [5.60-6.00]
Recurrence of CV event between hospital discharge and inclusion			
All CV events, n (%)	2 (4.7%)	7 (14%)	1 (7.7%)
Heart failure, n (%)	0 (0%)	4 (8%)	1 (7.7%)

(continued next page)

Myocardial infarction, n (%)	2 (4.7%)	1 (2%)	0 (0%)
Coronary revascularization, n (%)	0 (0%)	2 (4%)	0 (0%)
LVEF (%)	59.50 [49.50-61.00]	56.50 [50.00-65.00]	55.00 [50.00-59.00]
Inflammation			
hsCRP, mg/L	1.20 [0.70-2.70]	2.80 [1.25-6.00]	2.30 [1.00-3.60]
hsCRP > 2 mg/L, n (%)	13 (42%)	23 (66%)	5 (56%)
hsCRP > 5 mg/L, n (%)	6 (19%)	12 (34%)	2 (22%)
Atherosclerosis burden			
Multitroncular lesions, %	40%	54%	57%
Global atheroma volume, mm^3	455 [374-555]	520 [412-612]	515 [492-521]
Right CCA, mm^3	191 [158-248]	252 [164-359]	252 [202-330]
Left CCA, mm^3	241 [164-284]	266 [198-333]	240 [205-347]

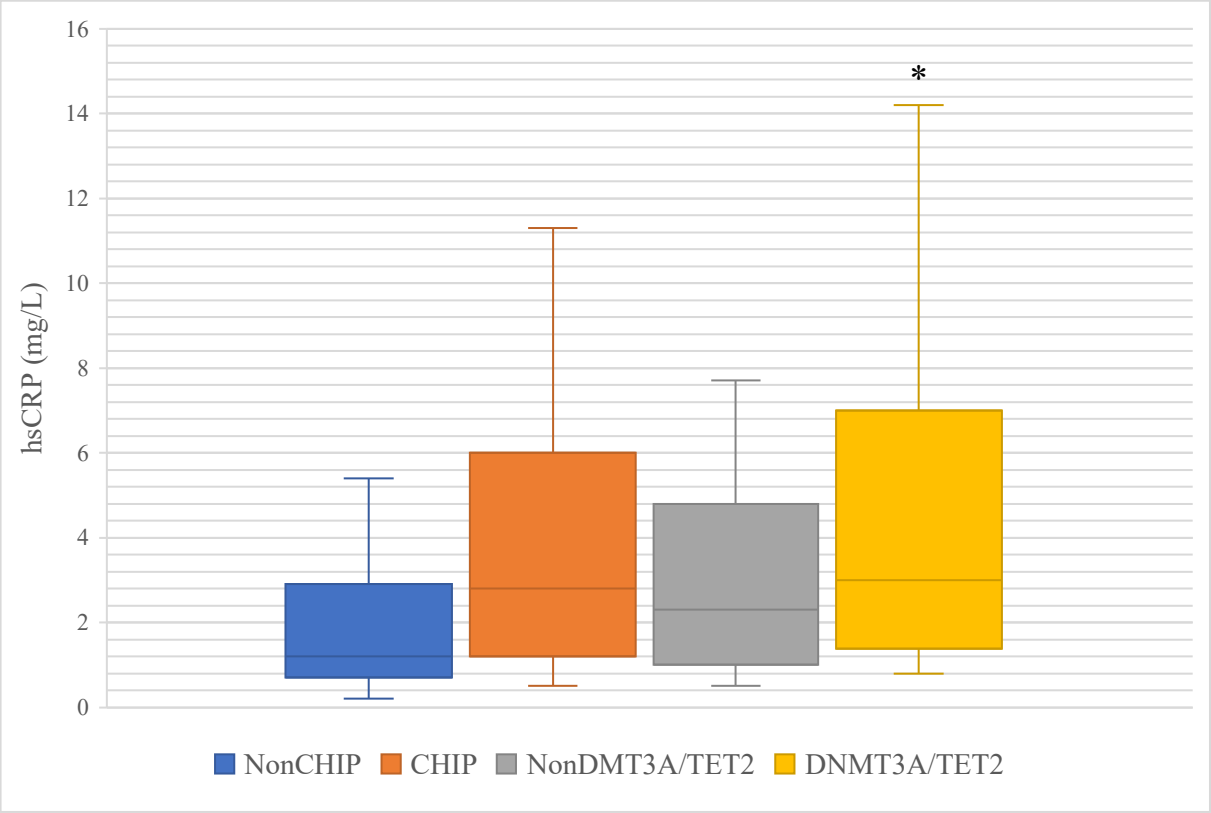
c-HDL: cholesterol-high density lipoprotein, c-LDL: cholesterol-low density lipoprotein, CCA: common carotid artery, CHIP: clonal hematopoiesis of indeterminate potential, CV: cardiovascular, hsCRP: high-sensitivity C-reactive protein, LVEF: left ventricular ejection fraction

Results are expressed in median and interquartile ranges for quantitative variables, and percentages of the concerned group for qualitative variables. For each variable, results do not include patients with missing values.

° All comparisons are made with the non-CHIP group, at a VAF ≥ 2%.

* Statistically significant ($p<0.05$).

Figure 5. hsCRP levels according to CHIP status (VAF 2%, univariate analyses)



hsCRP: high-sensitive C-reactive protein, CHIP: clonal hematopoiesis of indeterminate potential, VAF: variant allelic frequency

*Statistically significant ($p<0.05$). All comparisons are made with the non-CHIP group.

Finally, we noticed a trend towards a higher rate of recurrence of CV event between hospital discharge and inclusion in CHIP carriers compared with non-CHIP carriers, but without any statistical significance. We found no significant difference between CHIP and non-CHIP carriers regarding LVEF.

We then explored whether the type of mutated genes involved had an influence on these various parameters, and specifically studied patients with DNMT3A and/or TET2 mutations, the two genes most likely to be mutated in the cohort. Patients with CHIP excepting those carrying DNMT3A and/or TET2 mutation showed no significant difference in any parameter with non-CHIP carriers.

DNMT3A/TET2 (Table 3)

We focused our analyses on patients carrying DNMT3A and/or TET2 mutation, at VAF of 1%, 2%, 5% and 10%, comparing them to all other patients, namely those without CHIP and those with a CHIP not involving DNMT3A and/or TET2 mutation.

First, we found that patients with large clones had a more favorable profile of risk than non-DNMT3A/TET2 carriers: they were 62% to have at least two traditional risk factors at VAF ≥ 2%, 52% at VAF ≥ 5% and 42% at VAF ≥ 10%, compared with 61% in the non-DNMT3A/TET2 group.

Regarding inflammation, there was a trend toward a raise in the hsCRP level with VAF, from 2.0 mg/L at VAF ≥ 1%, to 3.0 mg/L at VAF ≥ 2% and 3.6 mg/L at VAF ≥ 5% and 10%. Patients with DNMT3A and/or TET2 mutation at VAF ≥ 2% and 5% had a significantly higher hsCRP than control group (3.00 [1.47-6.75] and 3.60 [1.60-8.07] respectively *v.* 1.40 [1.00-3.80], p=0.042 and p=0.043, Table 3 and Figure 5). After adjustment, we found that DNMT3A and/or TET2 mutations were positively correlated with hsCRP but without statistical significance (Table 4). Patients in the DNMT3A/TET2 group were more likely to have hsCRP > 2 mg/L and > 5 mg/L, regardless of the VAF, than patients in the control group, although these differences were not significant. Finally, we found a trend toward an increase in the white blood cells and neutrophils count, anyway within normal range, with VAF, with a statistically significant difference at VAF ≥ 5% and 10% for neutrophils count compared to control group (p=0.034 and 0.046 respectively).

Patients with DNMT3A and/or TET2 mutations were more likely to have multitroncular coronary lesions than controls, with a positive correlation with VAF: 49% of the patients in DNMT3A/TET2 group with a VAF ≥ 1% were concerned, compared with 53% and 62% at VAF ≥ 2% and 5% respectively. They also tended to have higher carotid atheroma volume.

Table 3. Characteristics at inclusion and secondary analyzes according to DNMT3A/TET2 status and VAF

	Non DNMT3A/TET2 (n=49)	DNMT3A/TET2 VAF ≥ 1%° (n=57)	DNMT3A/TET2 VAF ≥ 2%° (n=43)	DNMT3A/TET2 VAF ≥ 5%° (n=21)	DNMT3A/TET2 VAF ≥ 10%° (n=12)
Time from index event, months	5.00 [4.00-6.00]	4.00 [3.00-5.75]	4.00 [3.00-5.75]	4.00 [3.00-5.25]	4.00 [3.00-5.00]
Age, years	81.00 [78.00-87.00]	82 [80.00-86.00]	82.00 [80.00-87.00]	83.00 [81.00-85.00]	82.50 [80.20-85.80]
Male, %	71%	60%	65%	67%	67%
≥ 2 CV risk factors, %	61%	68%	62%	52%	42%
Blood tests					
Hemoglobin, g/dL	13.00 [12.30-14.40]	12.70 [12.1-13.5]	12.40 [12.00-13.40]	13.00 [12.30-13.50]	12.6 [12.30-14.40]
White blood cells, G/L	5.90 [5.40-7.20]	6.85 [5.88-7.95]	6.80 [5.78-7.50]	7.00 [6.52-7.60]	7.15 [6.45-7.72]
Neutrophils, G/L	3.80 [2.90-4.53]	4.50 [3.35-5.00]	4.40 [3.25-4.90]	**4.60 [4.25-4.95]***	**4.70 [4.35-5.35]***
Platelets, G/L	199 [156-230]	218 [175-254]	224 [191-253]	198 [175-248]	212 [172-260]
Total cholesterol, g/L	146 [1.21-1.69]	1.50 [1.28-2.10]	1.55 [1.23-2.13]	1.46 [1.26-2.10]	1.37 [1.14-1.92]
c-LDL, g/L	0.77 [0,62-0.87]	0.78 [0.65-1.04]	0.83 [0.67-1.13	0.83 [0.69-1.22]	0.78 [0.65-1.09]
c-HDL, g/L	0.45 [0.38-0.56]	0.47 [0.40-0.54]	0.46 [0.42-0.54]	0.47 [0.41-0.50]	0.47 [0.39-0.53]
HbA1c, %	6.10 [5.70-6.60]	6.00 [5.70-6.32]	5.90 [5.70-6.25]	5.90 [5.70-6.25]	6.05 [5.53-6.35]
Recurrence of CV event					
All CV events, %	4.4%	15%	16%	**24%***	17%
Heart failure, %	2.2%	6.2%	8.1%	14%	17%
Myocardial infarction, %	2.2%	4.2%	2.7%	4.8%	0%
Coronary revascularization, %	0%	4.2%	5.4%	4.8%	0%
LVEF, %	56.00 [50.00-61.00]	60.00 [46.00-64.50]	60.00 [46.00-64.50]	57.00 [47.00-66.00]	65.5 [44.20-68.00]

(continued next page)

Inflammation					
hsCRP, mg/L	1.40 [1.00-3.80]	2.00 [1.00-5.25]	**3.00 [1.47-6.75]***	**3.60 [1.60-8.07]***	3.60 [1.82-6.25]
hsCRP > 2 mg/L, %	48%	60%	69%	69%	75%
hsCRP > 5 mg/L, %	23%	31%	38%	44%	38%
Atherosclerosis burden					
Multi-troncular lesions, %	47%	49%	53%	62%	58%
Global atheroma volume, mm^3	484 [411-519]	552 [402-608]	552 [395-615]	552 [417-600]	476 [417-562]
Right CCA, mm^3	198 [164-262]	256 [157-393]	256 [152-372]	247 [152-387]	154 [148-282]
Left CCA, mm^3	244 [203-338]	259 [153-317]	270 [175-326]	270 [250-334]	259 [250-272]

c-HDL: cholesterol-high density lipoprotein, c-LDL: cholesterol-low density lipoprotein, CCA: common carotid artery, CHIP: clonal hematopoiesis of indeterminate potential, CV: cardiovascular, hsCRP: high-sensitivity C-reactive protein, LVEF: left ventricular ejection fraction

Results are expressed in median and interquartile ranges for quantitative variables, and percentages of the concerned group for qualitative variables. For each variable, results do not include patients with missing values.

° All comparisons are made with the non-DNMT3A/TET2 group, which encompasses patients without CHIP, and patients with CHIP but no DNMT3A or TET2 mutation.

* Statistically significant ($p<0.05$).

Table 4. Correlation between hsCRP and CHIP status (adjusted model)*

	Coefficient of correlation	p-value
CHIP at VAF ≥ 5%°	8.32 [0.464; 30.4]	0.0509
Age	0.0897 [-0.817; 0.867]	0.85
Sex (female *v.* male)	-6.82 [-19.7; -1.67]	0.12
≥ 2 risk factors	8.36 [1.29; 29.4]	0.061
DNMT3A/TET2 (VAF ≥ 2%)¶	5.09 [-0.635; 22.2]	0.2
Age	0.124 [-0.645; 0.898]	0.76
Sex (female *v.* male)	-5.91 [-19.2; -1.02]	0.15
≥ 2 risk factors	4.56 [-0.0827; 17.8]	0.29
DNMT3A/TET2 (VAF ≥ 5%)¶	8.62 [0.328; 29.9]	0.062
Age	0.128 [-0.717; 1.08]	0.78
Sex (female *v.* male)	-5.99 [-17.4; -0.674]	0.23
≥ 2 risk factors	7.22 [0.493; 21.3]	0.11

CHIP: clonal hematopoiesis of indeterminate potential, VAF: variant allelic frequency

* Linear regression using age, sex and presence of at least two traditional risk factors as adjusting variables

° Comparison is made with non-CHIP carriers

¶ Comparison is made with non-CHIP carriers and CHIP carriers without DNMT3A and/or TET2 mutation

Finally, patients carrying DNMT3A and/or TET2 mutation were more likely to present a recurrence of CV event, with 24% of patients at a VAF ≥ 5% being concerned *v.* 4.4% of the control group (p=0.029), mainly driven by hospitalization for HF (14% *v.* 2.2%, p=0.09). Again, there seemed to exist a correlation between VAF and both recurrence of CV event (15%, 16%, 24% at VAF ≥ 1%, 2% and 5%, respectively) and hospitalization for HF (6.2%, 8.1%, 14% at VAF ≥ 1%, 2% and 5%, respectively).

Discussion

Prevalence of CHIP

In our study, we showed that prevalence of CHIP was high among patients aged at least 75 years and suffering a first myocardial infarction, with 54% of the cohort being concerned by clonal hematopoiesis at VAF ≥ 2%. This is significantly higher than the expected prevalence of 30% we calculated based on what was described in original studies. To our knowledge, this is the first study to have estimated the prevalence of CHIP at the time of the event among patients presenting with an incidental CAD. Indeed, previous studies often estimated the prevalence of CHIP in patients with CAD in whom DNA had been obtained sometimes several years before the event. (5)(7) We can assume that this method could lead to an underestimation of the prevalence of CHIP in cohorts of patients who suffered a CV event, in whom clonal hematopoiesis could develop between the collection of DNA and the event. Thus, Busque et al. found that only 21.5% of the patients with CAD in their cohort carried a CHIP (11 genes studied, VAF ≥ 2%). Although patients in this study were younger than in our cohort (nearly 75 years *v.* 82 years), which can slightly minor the frequency of CHIP, we can presume that the difference can at least partially be explained by the time at which DNA is collected. Nevertheless, some studies described the prevalence of CHIP in patients having another CV condition, with DNA collected as they were already ill. In 2021, Pascual-Figal et al. found a prevalence of CHIP of 39% in 62 patients with chronic HF, irrespective of the etiology, with a mean age of 75 years.[18] Forty-six percent of the patients older than 75 years were concerned by CHIP in this study, close to our findings (54%). Previously in 2019 Mas-Peiro et al. had shown that 33% of a cohort of 279 patient undergoing transcatheter aortic valve replacement (TAVR) for severe aortic valve stenosis and aged of 83 years in median were carriers of a mutation in DNMT3A and/or TET2, slightly lower than our results (41% of DNMT3A and/or TET2 carriers upon the entire cohort in our study).[19] Thus, our study reinforces existing data and shows that prevalence of CHIP indeed is high in populations of patients with CVD.

The prevalence of CHIP we found in our cohort is much higher than the one described in original studies from general population.[2][6] Jaiswal and Genovese in 2014 both estimated the prevalence to be between 15 and 20% in patients aged over 70 years. This significant difference could be in favor of a real effect of CHIP in increasing the CV risk, but could also be explained by the different methods used to sequence DNA. Indeed, princeps studies generally used whole-exome sequencing to search for CHIP, with a lower sensitivity than NGS, allowing to detect CHIP at VAF of 4% or more. We can presume that a lot of patients carrying a CHIP at VAF between 2 and 4% were not detected, leading to a profound underestimation in the prevalence of CHIP in the general population. In 2020, Guermouche et al. evaluated the prevalence of clonal hematopoiesis in a cohort of 89 healthy volunteers aged between 18 and 80 years in Tours (France) using NGS.[20] They found that 17% of the cohort carried a CHIP detectable at a VAF ≥ 2% in the peripheral blood, with 30% of the patients between 50 and 60 years of age and 40% of the patients between 60 and 70 years of age being concerned by CHIP. These results, although needing to be confirmed in larger cohort, could be closer to the real prevalence in the general population and thus incite to relativize the high prevalence we found in our cohort. Moreover, Busque et al. in their study found no statistically significant difference between patients with and without CAD regarding to CHIP status, even with a trend toward a higher prevalence of CHIP in patients without CAD (25.3% *v.* 21.5%).[5] Van Zeventer et al. also found no significant difference in prevalence of CHIP between patients aged at least 80 years according to history of myocardial infarction.[21] In recent yet to be published-data from the Three-City Study, a collaborative program aiming to estimate the risk of dementia attributable to vascular risk factors, the prevalence of CHIP in two populations of patients aged 74 years in mean did not differ between patients having or not suffered from a myocardial infarction. Together, these recent studies have the merit of questioning the reality of the enrichment in CHIP in patients suffering CV events. Recently, Heyde et al. raised the hypothesis that atherosclerosis, rather than only being the consequence of CHIP through inflammation, would in fact be one of the main reasons why CHIP occurs.[22] Indeed, they built an experimental model in which mice with an atherogenic profile developed accelerated clonal hematopoiesis compared with non-atherogenic mice. In human, they showed that patients with atherosclerosis had a higher hematopoietic proliferation rate than healthy patients without atherosclerosis. Thus, we could hypothesize that the high prevalence of CHIP found in patients with CVD would rather be linked to the presence of atherosclerosis than to the occurrence of a CV event. Given that silent atherosclerosis may be common in the general population, this hypothesis could explain why no difference was found between groups in recent studies. In the

quest of the most accurate evaluation of the prevalence and the role of CHIP both in healthy populations and in patients with CVD, studies should therefore focus on the comparison between matched patients with and without atherosclerosis.

In our study, we found that prevalence of CHIP increased with age, with a prevalence up to 70% after 90 years of age. This is in accordance with many previous studies, supporting the hypothesis of an increasing risk of having a CHIP over the lifetime.[2][6][18][19][21][23] In line with previous data, the majority of the patients carried a single mutation. Clone size tended to increase with age, as elderly patients were more susceptible to carry clones with VAF ≥ 10% than younger patients. Finally, we found that DNMT3A and TET2 were the two most frequently mutated genes, in line with what is described since many years.[2][18][23] Both genes normally act as tumor suppressors, by regulating the proliferation of hematopoietic stem cells. A loss of function of these genes drives the hematopoietic system toward either an increased proliferation rate or a propensity to self-renewal rather than differentiation, thus leading to expansion of hematopoietic stem cells (HSC).[24]

Inflammation

Our study highlighted a marked trend toward higher levels of hsCRP in CHIP carriers compared with patients without CHIP, supporting the hypothesis of a pro-inflammatory effect of CHIP. Among patients with CHIP, those with a mutation in DNMT3A and/or TET2 had significantly higher hsCRP than the others in the univariate model. This is consistent with previous studies, indeed Busque et al. showed that patients with a mutation in DNMT3A had higher hsCRP levels than patients without CHIP.[5] Pascual-Figal et al. found that patients carrying a mutation in DNMT3A and/or TET2 had higher IL-6 levels, whereas the rest of the patients with CHIP did not.[18] All together, these data as well as ours are in favor of a pro-inflammatory effect of DNMT3A/TET2-associated CHIP.

We found a statistically significant difference between patients with CHIP at a VAF ≥ 5% and non-CHIP carriers in the univariate model, and a raise of hsCRP levels with VAF in DNMT3A/TET2 carriers. The proportion of patients carrying a CHIP and having hsCRP levels > 2 mg/L, threshold that has previously been identified as having a prognostic value regarding to occurrence of major CV events, also tended to increase with clone size.[25] These results suggest a positive correlation between clone size and inflammation. To our knowledge, only Busque et al. studied the relationship between clone size and inflammation, and found no major difference between CHIP at VAF ≥ 2% and VAF ≥ 10%.[5]

The adjusted model did not show any statistically significant correlation between clone size or DNMT3A/TET2 status and hsCRP, but we still could notice strong trends. One possible explanation of the absence of statistically significant difference in the adjusted model, already discussed by Busque et al., could be that hsCRP may not be the best marker of inflammation in patient with CHIP.[5] Indeed, Bick et al. in 2020 showed that patients with CHIP had significantly higher levels of IL-6 and IL-1ß than patients without CHIP, but that there was no difference regarding hsCRP in 22,092 patients.[26] The same year, studying 35,416 patients with and without CHIP, they found a significant association between CHIP status and hsCRP in a univariate analyses, that failed to be confirmed in the adjusted model.[17] Finally, as seen above, Pacual-Figal et al. showed a significant difference regarding IL-6 levels in patients with DNMT3A/TET2 mutation, but no significant difference in hsCRP levels.[18] Although hsCRP is produced in response to the secretion of IL-6 and IL-1ß, these markers could be more closely correlated to CHIP status. The results of the analyzes of IL-6, IL-10, IL-1ß and TNFα dosing will help us to have a more precise evaluation of inflammatory status of CHIP carriers.

However, given the high level of correlation between CHIP and hsCRP in the adjusted model, approaching statistical significance, we can presume that the ongoing recruitment of a few more patients will give our study the adequate power to reach statistical significance. If so, it could confirm the link between CHIP and inflammation and have clinical implications. Indeed, last years have provided some evidences of a beneficial effect of anti-inflammatory therapies on the prognosis of patients in secondary prevention after a myocardial infarction. Tardif et al. in the COLCOT trial showed that colchicine improved prognosis after a myocardial infarction, reducing the risk of stroke and urgent revascularization.[27] The CANTOS trial showed that high doses of canakinumab administered in patients with hsCRP > 2 mg/L also reduced the risk of further CV events.[28] Thus, showing that CHIP is associated with inflammation in patients suffering a myocardial infarction could help us identify the patients who could benefit the most from anti-inflammatory therapies.

Atherosclerosis burden

To our knowledge, this is the first study to assess both coronary and carotid atherosclerosis disease and to quantify atheroma volume in carotid arteries in patients with CHIP. We found no statistically significant difference between CHIP and non-CHIP carriers regarding atherosclerosis burden. Patients with CHIP had a non-significant relative risk of 1.4 to have multitroncular coronary artery lesions. We also found that patients with CHIP, especially those carrying DNMT3A and/or TET2 mutation, had a slightly but not significantly

higher carotid atheroma volume than non-CHIP carriers. Finally, as for inflammation, we noticed a trend toward a dose-response effect, with a higher proportion of multitroncular lesions in patients with DNMT3A/TET2 mutation with high VAF. Although numerous studies have shown that CHIP was associated with an increased atherosclerotic burden in mutated mice, few studies have evaluated atherosclerosis burden in human carrying CHIP.[7][4] Only Jaiswal et al. in 2017 measured the coronary-artery calcification (CAC) score in a cohort of patients aged 70 years in mean, and compared the results between CHIP and non-CHIP carriers. They found that CHIP carriers had a 3.3 times higher CAC score than non-CHIP carriers in patients without CAD, and 1.8 times higher in patients with CAD. Mas-Peiro et al. in 2019 showed that, in patients undergoing TAVR, DNMT3A/TET2 carriers had the same proportion of carotid and peripheral artery diseases as patients without CHIP.[19] These data, as well as ours, are insufficient to support both hypotheses of CHIP increasing atherosclerosis burden and being favored by atherosclerosis, as suggested by Heyde et al.[22] The upcoming analysis of the ankle-brachial index and the significant carotid stenoses will help to precise the evaluation. However, more data are needed to specifically assess the relationship between CHIP and atherosclerosis burden in human.

Influence on prognosis

In our study, patients with CHIP seemed to have poorer short-term prognosis than non-CHIP carriers. They were more likely to present with a recurrent CV event, mainly HF or coronary revascularization, although the difference was not significant. Patients with DNMT3A and/or TET2 mutation also showed a higher rate of recurrent myocardial infarction. Overall, only the patients carrying DNMT3A and/or TET2 mutation at VAF ≥ 5% had a statistically significantly higher rate of adverse outcomes than the others. However, we noticed a marked trend toward increased rates of events with VAF, suggesting a correlation between clone size and adverse prognosis. These observations are consistent with previous studies having evaluated the prognosis of patients carrying CHIP. Dorsheimer et al. showed that patient with chronic ischemic HF carrying CHIP had a higher risk of adverse outcome than non-CHIP carriers, with a higher incidence of death and hospitalization for HF during follow-up.[23] Pascual-Figal et al. found that patients with chronic HF, irrespective of etiology, had a higher risk of hospitalization for HF when they carried CHIP.[18] In both study, clone size was positively correlated to the risk of poor prognosis, and mutations in DNMT3A and/or TET2 were the most associated with adverse outcomes, all consistent with our findings. Moreover, Mas-Peiro et al. when studying the prognosis of patients undergoing TAVR also showed that

DNMT3A and/or TET2 carriers were more at risk to die during follow-up than patients without those mutations.[19] Together, these data are in favor of a deleterious role played by CHIP, particularly DNMT3A and TET2, upon prognosis of patients with CVD.

Interaction with risk factors

Evaluating the presence of two or more concomitant traditional risk factors in each patient gave us the possibility to evaluate more accurately the profile of CV risk of our cohort, and the interaction between CHIP and traditional risk factors. To our knowledge, no previous study has made such a global evaluation of the profile of risk in CHIP patients. In our study, patients with CHIP showed a trend toward a less at-risk profile than non-CHIP carriers, although results were not statistically significant. This trend was bigger in elderly patients. As for inflammation, atheroma and prognosis, we highlighted a link between clone size and the profile of risk, as patients with DNMT3A/TET2 mutations with large clones were markedly less susceptible to cumulate two or more risk factors. Surprisingly, CHIP carriers were also less susceptible to have diabetes, and had lower HbA1c. This is in contradiction with recent data, which tended to show that CHIP favored diabetes and insulin resistance. In 2020, Fuster et al. showed that inactivation of TET2 in mice was associated with more insulin resistance through overproduction of IL-1ß in adipose tissue.[29] Previous studies mainly showed no significant difference between CHIP and non-CHIP carriers regarding diabetes status.[5][19][23] Jaiswal et al. in 2017 showed a higher prevalence of diabetes in patients carrying CHIP.[2] Only Pascual-Figal et al. (54% v. 66%, p=0.52) and Chavkin et al. (27% v. 31%, p=0.64) showed a little trend toward a lower frequency of diabetes in groups of patients with CHIP, but not in the same proportion as we did.

The trend toward a negative correlation between cumulation of risk factors and CHIP we observed in our study raises some questions about their mutual influence on each other. Could CHIP protect against the development of traditional risk factors, diabetes in particular ? Could a high at-risk profile act as a protector against CHIP ? This hypothesis would be in contradiction with data indicating that inflammation enhances clonal hematopoiesis, as we know that risk factors are associated with a higher level of inflammation.[24][30][31][32] Recently, smoking has been linked to CHIP in a study in postmenopausal women.[33] In the same study, they showed that obesity was also associated with a higher prevalence of CHIP. Moreover, recent data have suggested that diabetes, through chronic hyperglycemia, favored TET2 loss-of-function, thus increasing the risk of cancer.[34] Data on hyperlipidemia are controversial.[35]

Heyde et al. recently showed that hyperlipidemia without atherosclerosis was not sufficient to drive clonal hematopoiesis.[22]

Overall, our results raise the question of the nature of the links between CHIP and traditional CV risk. More work is needed to specifically study the bilateral relationship between CHIP and risk factors.

Study strengths and limitations

The main limitation of our study is the limited sample size, which gave us not enough power to highlight clear statistically significant differences between CHIP and non-CHIP carriers in secondary analyses. Missing values and unavailability of the 3D measurement of atheroma volume for each patient also participated in this lack of power. The ongoing recruitment will help to increase power and fix these issues. Another limitation is the relatively short delay between the event and the inclusion, which could limit the ability to record recurrence of CV event. We chose this timing in order not lose too many patients between the screening at index event and the inclusion, given that we studied a quite old and fragile population. We are currently extending the recording of CV events to six months after the inclusion in order to get more power to address this important question.

Our study also has some strengths. The prospective model allowed us to search for CHIP at the closest of the event, thus to give the most accurate estimation of the prevalence of CHIP in a population of patients suffering from a myocardial infarction. It also gave us the possibility to evaluate inflammation and atheroma at the closest of the event, in a timing that let the inflammation reach its chronic level, to avoid interferences with these parameters and conditions that could have happened later in the course of life (cancer, inflammatory diseases…). Moreover, for the first time an evaluation of carotid atheroma volume has been done in human with CHIP, and associated to the characterization of coronary lesions to evaluate the global atherosclerosis burden of this patients.

Future directions

Future studies should aim to assess the comparative prevalence of CHIP in populations of people with and without atherosclerosis, to identify the populations the most concerned by CHIP, and to test the recent hypothesis suggesting that the relationship between CHIP and CVD may not be unilateral. Three-dimensional measurement of atheroma volume could be used in order to properly select individuals of each group. Powerful studies are needed to confirm the link between CHIP and atheroma volume in human. Evaluation of the efficacy of anti-

inflammatory therapies in patients with CHIP compared with non-CHIP carriers could be considered. Finally, links between CHIP and traditional CV risk factors will also have to be assessed, in order to understand the mutual influence they have on each other.

Conclusion

Our study showed that CHIP is highly prevalent in a population of patients suffering a first myocardial infarction after 75 years. This study also raised questions about reciprocal links between CHIP and inflammation, atherosclerosis burden and traditional risk factors, that will need to be addressed by further studies.

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Appendix

Figure S1. Checklist at inclusion

ETUDE CHAth		
Nom : _____ **Prénom :** _____ **Code participant :** ⌊_⌊_⌊_⌋ ⌊_⌋ . ⌊_⌋	**PRELEVEMENT INFLAMMATION**	***Version n°1.0 du 08/06/20 MM*** ***Protocole v1.1_20200220***

	Oui	**Non**
Infection ou fièvre **> 38,5°C** dans les **15j** avant hospi Température : ..... Date début : ..... Durée :	☐	☐
Opération chirurgicale dans le **mois** avant hospi Type : ..... Date :	☐	☐
Anti-inflammatoires dans les **7j** avant hospi (non au long cours) Traitement : ..... Poso (/24h) : ..... Date début : ..... Durée :	☐	☐

Table S1. Gene panel used in the Hematology laboratory of the Bordeaux University Hospital for the diagnosis and follow-up of myeloid hemopathies

Gene	Transcript		Gene	Transcript		Gene	Transcript	
ANKRD26	NM_014915.2	5'UTR	**GNAS**	NM_000516.5	exon 8	**RIT1**	NM_006912	exon 5
ASXL1	NM_015338.5	whole exon	**GNB1**	NM_002074.4	exon 5-6	**RUNX1**	NM_001754.4	whole exon
ASXL2	NM_018263	exon 11-12	**IDH1**	NM_005896.2	whole exon	**SAMD9**	NM_017654	whole exon
BCOR	NM_001123385	whole exon	**IDH2**	NM_002168.2	whole exon	**SAMD9L**	NM_001303497.2	whole exon
BCORL1	NM_021946	whole exon	**IKZF1**	NM_006060.6	whole exon	**SETBP1**	NM_015559.2	exon 4
CALR	NM_004343.3	exon 9	**JAK2**	NM_004972	whole exon	**SF3B1**	NM_012433.2	exon 13-16
CBL	NM_005188.2	whole exon	**KDM6A**	NM_021140.2	whole exon	**SH2B3**	NM_005475.2	whole exon
CCND2	NM_001759.3	exon 5	**KIT**	NM_000222.2	whole exon	**SMC1A**	NM_006306	whole exon
CEBPA	NM_4364.3	whole exon	**KRAS**	NM_033360.2	whole exon	**SMC3**	NM_005445.3	whole exon
CSF3R	NM_156039.3	whole exon	**MPL**	NM_005373.2	whole exon	**SRSF2**	NM_003016.4	whole exon
CUX1	NM_181552.3	whole exon	**MYC**	NM_001354870	whole exon	**STAG2**	NM_001042749.2	whole exon
DDX41	NM_016222	whole exon	**NFE2**	NM_006163.2	whole exon	**TERC**	NM_001566.1	whole exon
DNMT3A	NM_175629.1	whole exon	**NPM1**	NM_002520	exon 11	**TERT**	NM_198253	whole exon
ETNK1	NM_018638	exon 3	**NRAS**	NM_002524.3	whole exon	**TET2**	NM_001127208.1	whole exon
ETV6	NM_001987.4	whole exon	**PHF6**	NM_001015877	whole exon	**TP53**	NM_000546.4	whole exon
EZH2	NM_004456.4	whole exon	**PPM1D**	NM_003620	exon 6	**U2AF1**	NM_001025203.1	whole exon
FLT3	NM_004119	whole exon	**PTEN**	NM_000314.4	exon 5-7	**WT1**	NM_001198551	exon 7 + 9
GATA1	NM_002049.3	whole exon	**PTPN11**	NM_002834.3	exon 3 + 13	**ZRSR2**	NM_005089	whole exon
GATA2	NM_032638	whole exon + intron 4	**RAD21**	NM_006265.2	whole exon			

Serment d'Hippocrate

A l'heure d'être admis à exercer la médecine, je promets et je jure de rester fidèle aux lois de l'honneur et de la probité.

Mon premier souci sera de rétablir, de préserver ou de promouvoir la santé dans tous ses éléments, physique et mentaux, individuels et sociaux.

Je respecterai toutes les personnes, leur autonomie et leur volonté, sans aucune discrimination selon leur état ou leurs convictions. J'interviendrai pour les protéger si elles sont affaiblies, vulnérables ou menacées dans leur intégrité ou leur dignité. Même sous la contrainte, je ne ferai pas usage de mes connaissances contre les lois de l'humanité.

J'informerai les patients des décisions envisagées, de leurs raisons et de leurs conséquences.

Je ne tromperai jamais leur confiance et n'exploiterai pas de pouvoir hérité des circonstances pour forcer les consciences.

Je donnerai mes soins à l'indigent et à quiconque me les demandera. Je ne me laisserai pas influencer par la soif du gain ou la recherche de la gloire.

Admis dans l'intimité des personnes, je tairai les secrets qui me seront confiés. Reçu à l'intérieur des maisons, je respecterai les secrets des foyers et ma conduite ne servira pas à corrompre les mœurs.

Je ferai tout pour soulager les souffrances. Je ne prolongerai pas abusivement les agonies. Je ne provoquerai jamais la mort délibérément.

Je préserverai l'indépendance nécessaire à l'accomplissement de ma mission. Je n'entreprendrai rien qui dépasse mes compétences. Je les entretiendrai et les perfectionnerai pour assurer au mieux les services qui me seront demandés.

J'apporterai mon aide à mes confrères ainsi qu'à leurs familles dans l'adversité.

Que les hommes et mes confrères m'accordent leur estime si je suis fidèle à mes promesses ; que je sois déshonoré et méprisé si j'y manque.

Fréquence de l'hématopoïèse clonale chez les patients de plus de 75 ans présentant un premier événement cardiovasculaire. Conséquences sur l'inflammation et le volume athéromateux.
Contexte. L'hématopoïèse clonale de potentiel indéterminé (CHIP) est décrite depuis quelques années comme étant fréquente au sein de la population générale, et semble associée à un risque plus élevé de maladies cardiovasculaires. Des données expérimentales tendent à montrer que le lien entre la CHIP et l'athérosclérose ou l'insuffisance cardiaque serait principalement médié par l'inflammation. A ce jour, peu de données sont disponibles chez l'Homme.
Objectifs. Notre étude avait pour but d'évaluer la prévalence de la CHIP chez des patients âgés de plus de 75 ans et présentant un premier infarctus du myocarde, ainsi que les liens entre CHIP, inflammation et athérosclérose. L'interaction avec les facteurs de risque et l'influence sur le pronostic des patients étaient également étudiées.
Méthodes. Nous avons mené une étude prospective, monocentrique, incluant des patients âgés d'au moins 75 ans présentant un premier infarctus du myocarde. Les patients étaient inclus 2 à 7 mois après l'événement, et bénéficiaient d'un prélèvement sanguin permettant le séquençage de l'ADN à la recherche de CHIP et le dosage de la CRP ultra-sensible, d'un Doppler des troncs supra-aortiques avec quantification 3D du volume athéromateux, et d'une ETT. La récidive d'un événement cardiovasculaire était également recherchée.
Résultats. Sur les 106 patients inclus, 54% étaient porteurs d'une CHIP. Les porteurs de CHIP présentaient un niveau d'inflammation plus élevé et une charge en athérome modestement plus élevée que les patients indemnes de CHIP. Ces différences semblaient particulièrement corrélées à la taille du clone et à la présence d'une mutation des gènes DNMT3A et/ou TET2. Les patients porteurs de CHIP cumulaient en moyenne moins de facteurs de risque cardiovasculaire, mais présentaient un sur-risque d'événement cardiovasculaire à distance de l'infarctus, notamment d'hospitalisation pour insuffisance cardiaque.
Conclusion. Cette étude montre que la prévalence de CHIP est élevée chez les patients âgés présentant un infarctus du myocarde, et est en faveur d'un rôle pro-inflammatoire de la CHIP et d'une altération du pronostic. Les liens avec l'athérome et les facteurs de risque devront être réévalués dans de futurs travaux.

Frequency of clonal hematopoiesis of indeterminate potential in patients over 75 years with a first cardiovascular event. Consequences on inflammation and atherosclerosis.
Context. Clonal hematopoiesis of indeterminate potential (CHIP) has been described for several years as being common in the general population, and to be associated with an increased risk of cardiovascular disease. Experimental data tend to show that the link between CHIP and atherosclerosis or heart failure is mainly mediated by inflammation. To date, little data is available in human.
Goals. Our study aimed to assess the prevalence of CHIP in patients over 75 years of age with a first myocardial infarction, as well as the links between CHIP and inflammation and atherosclerosis. The interaction with risk factors and the influence on patient prognosis were also studied.
Methods. We conducted a prospective, single-center study, including patients aged at least 75 years with a first myocardial infarction. The patients were included 2 to 7 months after the event, and had a blood test allowing DNA sequencing in search for CHIP and the dosing of high-sensitive CRP, a supra-aortic trunks ultrasonography with 3D quantification of atherosclerotic volume, and TTE. Recurrence of a cardiovascular event was also sought.
Results. Of the 106 included patients, 54% were carriers of CHIP. CHIP carriers had a higher level of inflammation and a modestly higher atheroma load than patients without CHIP. These differences seemed to be particularly correlated with the size of the clone and the presence of a mutation in the DNMT3A and/or TET2 genes. Patients with CHIP had on average fewer cardiovascular risk factors, but had an increased risk of cardiovascular event after the myocardial infarction, in particular hospitalization for heart failure.
Conclusion. This study shows that the prevalence of CHIP in elderly patients with myocardial infarction is high, and support a pro-inflammatory role of CHIP and altered prognosis. Links with atheroma and risk factors will need to be reassessed in future works.

Mots clés : CHIP, inflammation, athérosclérose, facteurs de risque
Keywords: CHIP, inflammation, atherosclerosis, risk factors
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