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Peripheral blood cytopenias in the aging general population and risk of incident hematological disease and mortality

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Key Points

- Hematological malignancies develop in only a small fraction of community-dwelling individuals with a peripheral cytopenia.
- Neutropenia is not associated with increased mortality or development of hematological malignancies in older community-dwelling subjects.

Peripheral blood cytopenias may precede the development of hematological malignancies and frequently pose clinical challenges in the older population. The natural course of (mild) cytopenias during aging and their association with hematological disorders in community-dwelling individuals are not well studied. Within the population-based Lifelines cohort (n = 167 729), we studied changes in peripheral blood counts, occurrence of cytopenias, and associated hematological outcomes in the context of aging. Development of hematological malignancies and (cause-specific) mortality were evaluated by linkage to nationwide registries. Anemia and thrombocytopenia emerged with older age, in line with a general age-related decline in these blood counts. For neutropenia, no increase in prevalence with older age was observed. Using standard reference limits to define cytopenias, anemia (hazard ratio [HR], 1.84; 95% confidence interval [CI], 1.59-2.12), thrombocytopenia (HR, 1.58; 95% CI, 1.32-1.89), and, especially the concomitant presence of anemia and thrombocytopenia (HR, 4.75; 95% CI, 2.98-7.55) were associated with inferior overall survival. Only a minor proportion of deaths was explained by diagnosed hematological malignancies, with the majority attributable to other causes. Neutropenia, either isolated (HR, 0.88; 95% CI, 0.73-1.06) or combined with another cytopenia, did not affect overall survival. For individuals aged ≥60 years, 5-year cumulative incidence of hematological malignancies was 0.60% (95% CI, 0.50-0.70), with higher incidences among those with anemia ($P < .001$) or thrombocytopenia ($P < .001$) but not neutropenia ($P = .201$). Highest cumulative incidences of diagnoses and mortality from hematological malignancies were observed in individuals with >1 cytopenia. We conclude that anemia and thrombocytopenia, but not neutropenia, are associated with inferior overall survival of community-dwelling individuals. Hematological malignancies develop in a small fraction of these cases.

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The Lifelines cohort study adheres to standards for data availability. The data catalogue of Lifelines is publicly accessible at www.lifelines.nl. All international researchers can obtain data at the Lifelines research office (research@lifelines.nl), for which a fee is required. Data on incident hematological malignancies were obtained from PALGA, the nationwide network and registry of histo- and cytopathology in The Netherlands (for more information: https://www.palga.nl/gegevensaanvragen/gegevensaanvragen.html). Results from cause of death analyses are based on calculations by the authors using nonpublic microdata from Statistics Netherlands. Under certain conditions, these microdata are accessible for statistical and scientific research. For further information contact microdata@cbs.nl.

The full-text version of this article contains a data supplement.

Introduction

Older individuals presenting with idiopathic cytopenia may pose diagnostic and therapeutic challenges, because the etiology and natural course remain unknown in the majority of cases.[1,2] With the number of people aged ≥60 years increasing to 22% of the total population in 2050,[3] an increasing number of (asymptomatic) older individuals will be found to have abnormal peripheral blood counts, including cytopenias.

Subtle alterations in peripheral blood counts may be present several years prior to the diagnosis of myelodysplastic syndromes (MDSs).[4] MDSs are malignant myeloid disorders that are characterized by a disturbed differentiation of blood cell lineages, formation of dysplastic cells, and peripheral blood cytopenias. The prevalence of MDS increases steadily with age.[5,6] It has been suggested that (lower-risk) MDS may account for a substantial proportion of (unexplained) cytopenias in older patients.[7] The term "idiopathic cytopenia of undetermined significance" (ICUS) has recently been introduced to describe a condition with unexplained cytopenia(s) that does not (yet) fit the diagnostic criteria of MDS or another hematopoietic disorder.[2,8-10] Although a peripheral cytopenia may be the first sign of a malignant hematological disorder, knowledge about the long-term hematological outcomes for individuals with (mild) cytopenia is limited, and the natural disease course of ICUS is not well established. For example, the thresholds for defining a cytopenia relevant for the diagnosis of MDS or ICUS are still a matter of controversy.[10-14] Further, it remains unknown whether these individuals develop overt disease or have blood levels lower than normal without apparent clinical significance.

We studied changes in peripheral blood counts, occurrence of cytopenias, and associated hematological outcomes in the context of aging, making use of the prospective and population-based Lifelines cohort comprising 167 729 community-dwelling individuals. By linkage to nationwide pathology and mortality registers, we were able to estimate the probabilities for the development of and death from hematological malignancies.

Methods

Cohort selection

For this study, we included 152 180 individuals aged ≥18 years from the prospective Lifelines Cohort Study. Lifelines is a multidisciplinary prospective population-based cohort study of 167 729 persons living in the northern part of The Netherlands. It uses a broad range of investigative procedures to assess the biomedical, sociodemographic, behavioral, physical, and psychological factors that contribute to the health and disease of the general population, with a special focus on multimorbidity and complex genetics.[15,16] Lifelines participants are broadly representative of the general background population in the northern part of The Netherlands with respect to chronic diseases and overall health.[16] A follow-up visit was scheduled after ~5 years. The Lifelines study is conducted according to the principles of the Declaration of Helsinki. An informed consent form was signed by every participant at study inclusion. The local medical ethical committee approved the study protocol. Detailed descriptions of Lifelines recruitment procedures and representativeness are given in the supplemental Appendix.

Definition of cytopenias

At the baseline visit, peripheral blood samples were drawn after an overnight fast and directly processed for routine clinical chemistry assays at the University Medical Center Groningen. Complete and differential blood counts were determined using an XE-2100 Automated Hematology System (Sysmex, Kobe, Japan).

First, we used standard reference values to define cytopenias. Anemia, in accordance with World Health Organization (WHO) criteria,[17] was defined as hemoglobin concentration <13.0 g/dL in men and <12.0 g/dL in women. Thrombocytopenia was defined as platelet counts $<150 \times 10^9$/L, and neutropenia was defined as absolute neutrophil counts $<1.8 \times 10^9$/L. When possible, we evaluated alternative proposed cutoffs for increasing severity of the cytopenia, based on the cutoff criteria that have been proposed to define ICUS and cytopenias in MDS (Table 1; supplemental Table 1). Moderate and severe anemia were defined by hemoglobin levels <11.0 g/dL and <10.0 g/dL, respectively. Platelet counts $<100 \times 10^9$/L were used to define moderate thrombocytopenia. Absolute neutrophil counts $<1.5 \times 10^9$/L and $< 0.8 \times 10^9$/L were used to define moderate and severe neutropenia, respectively. Additionally, the prevalence of severe cytopenias was evaluated according to cutoff criteria corresponding to the toxicity criteria of the National Cancer Institute.[18]

Outcomes

Overall survival (OS) was defined as the time from study inclusion until death. Survival status of participants was ascertained by consulting the Municipal Persons Records Database (last consultation: June 2020).

The Lifelines cohort was linked to the Dutch Nationwide Network and Registry of Histo- and Cytopathology (PALGA) to retrieve positive histology reports for diagnosis of hematological malignancies.[19] The PALGA registry is a fully automated archive of pathology reports and covers all pathology laboratories and pathology reports in The Netherlands. All retrieved histopathology reports were manually screened to identify diagnoses of malignant hematological disease. For deceased participants, the primary cause of death, as reported on the death certificate by the physician, was obtained by linkage to the national registry (Statistics Netherlands). *Tenth Revision of the International Statistical Classification of Diseases and Related Health Problems* codes C81x-C96x and D45x-D47x were used to classify death from hematological neoplasms or other causes. Extensive information about the linkage and coding procedures is outlined in supplemental Methods.

Statistical analyses

Student *t* tests or Mann-Whitney *U* tests were used to evaluate between-group differences for parametric and nonparametric data, respectively. We used the Kaplan-Meier estimator for visualization of OS. Cox proportional hazard regression models were used to evaluate risks for all-cause mortality. For development of hematological malignancies and cause-specific survival, cumulative incidence curves were constructed, and Fine-Gray competing risk-regression was performed, with death or death from other causes, respectively, considered a competing risk. Cumulative incidences were compared between groups using Gray's test. When the number of events allowed for reliable regression estimation,[20,21] we stratified analyses by age (<60 or ≥60 years) or evaluated different

Table 1. Baseline characteristics of the Lifelines cohort and prevalence of cytopenias according to different proposed cutoff criteria.

	n	Total cohort	<60 y old	≥60 y old
No. of individuals	152 180	152 180	129 728	22 452
Males	152 180	63 130 (41.5)	52 886 (40.8)	10 244 (45.6)
Age, median (range), y	152 180	44 (18-93)	42 (18-59)	65 (60-93)
Hemoglobin level, mean (SD), g/dL	147 170	14.14 (1.27)	14.12 (1.29)	14.24 (1.16)
Platelet count, mean (SD), $\times 10^9$/L	147 074	249.9 (56.6)	251.7 (56.4)	239.7 (56.8)
White blood cell count, mean (SD), $\times 10^9$/L	147 171	6.10 (1.79)	6.13 (1.73)	5.92 (2.11)
Neutrophil count, mean (SD), $\times 10^9$/L	144 676	3.33 (1.23)	3.36 (1.25)	3.15 (1.10)
Anemia (<12.0 g/dL [women] or <13.0 g/dL [men])*	147 170	6 159 (4.2)	5 403 (4.2)	756 (3.4)
Moderate (<11.0 g/dL)†	147 170	1 272 (0.9)	1 186 (0.9)	86 (0.4)
Severe (<10.0 g/dL)‡,§	147 170	442 (0.3)	419 (0.3)	23 (0.1)
Thrombocytopenia ($<150 \times 10^9$/L)*	147 074	2 408 (1.6)	1 777 (1.4)	631 (2.8)
Moderate ($<100 \times 10^9$/L)†,§	147 074	153 (0.1)	117 (0.09)	36 (0.2)
Neutropenia ($<1.8 \times 10^9$/L)*,‡	144 676	6 925 (4.8)	5 643 (4.3)	1 282 (5.7)
Moderate ($<1.5 \times 10^9$/L)†	144 676	2 050 (1.4)	1 644 (1.3)	406 (1.8)
Severe ($<0.8 \times 10^9$/L)§	144 676	30 (0.02)	27 (0.02)	3 (0.01)

Unless otherwise noted, data are n (%).
SD, standard deviation.
*Generally accepted reference values, argued for by Greenberg et al to define cytopenias in MDSs[11] and used to define ICUS in 2017.[10]
†Used to define ICUS in 2007.[13]
‡WHO criteria to define cytopenias in MDS.[14]
§Alternative criteria to define cytopenias relevant for prognosis in MDS (IPSS-R).[12]

cytopenia severities. Age- and sex-corrected hazard ratios (HRs) are presented with the 95% confidence interval (CI). For OS, additional multivariable models were constructed, including the number of medications as a covariable, as a proxy for comorbidity. Statistical analyses were performed using R statistical computing software.

Results

Prevalence of cytopenias in conjunction with age-related changes in peripheral blood counts

Characteristics of the evaluable Lifelines cohort are presented in Table 1. Hemoglobin levels and platelet counts were available for 147 170 and 147 074 individuals, respectively. The complete differential blood cell count, including neutrophil levels, was available for 144 676 individuals. We first assessed the natural course of blood counts upon aging. For the hemoglobin concentration, a decrease upon aging was observed for males, whereas concentrations in women only declined at ≥80 years of age. For older individuals, a gradual decline in platelet counts was observed, whereas neutrophil counts increased at older age (Figure 1A-C).

To investigate the prevalence of cytopenias in our cohort, we first adopted the general reference values for cytopenias (Table 1; Figure 1D-F). Anemia, defined in accordance with WHO reference limits, was observed in 6159 of 147 170 evaluable individuals (4.2%). A total of 2408 of 147 074 individuals (1.6%) had thrombocytopenia. The prevalence of neutropenia was 4.8% (6925/144 676). The prevalence of anemia and thrombocytopenia increased in older individuals, whereas the prevalence of neutropenia decreased. Among individuals aged ≥80 years, 9.4% and 5.2% were found to be anemic and thrombocytopenic, respectively. Next, alternative cutoffs were evaluated. We identified a smaller subset of individuals having moderate to severe anemia: 1272 individuals (0.9%) with hemoglobin levels <11.0 g/dL and 442 (0.3%) with hemoglobin levels <10.0 g/dL. Moderate thrombocytopenia (platelet counts $<100 \times 10^9$/L) was detected in only 153 (0.1%) individuals. Moderate neutropenia (neutrophil count $<1.5 \times 10^9$/L) was detected in 2050 individuals (1.4%). Only 30 individuals (0.02%) had severe neutropenia (neutrophil count $<0.8 \times 10^9$/L).

The prevalence of severe cytopenias in accordance with National Cancer Institute toxicity criteria is shown in supplemental Table 2. Hemoglobin levels <8.0 g/L were detected in 0.03% of individuals, and platelet counts $<75 \times 10^9$/L were found in 0.04%. The prevalence of neutropenia with neutrophil counts $<1.0 \times 10^9$/L was 0.07%.

Multilineage cytopenia was detected in 861 individuals (supplemental Figure 1). Most individuals had anemia and neutropenia (n = 525; 70 were aged ≥60 years); the prevalence decreased with age. Concurrent thrombocytopenia and neutropenia were detected in 277 individuals (69 of whom were aged ≥60 years). A small proportion of individuals had anemia and thrombocytopenia (n = 111; 31 were aged ≥60 years). The prevalence of concurrent anemia and thrombocytopenia, as well as concurrent thrombocytopenia and neutropenia, increased with age. Pancytopenia was rare in this population-based cohort (n = 26).

Sex differences in peripheral blood counts translate into differences in the prevalence of cytopenias

The differences between the sexes that were observed with regard to peripheral blood counts also translated into differences in the prevalence of cytopenias. Consistent with higher platelet levels in females (mean, 263 ± 57 vs 231 ± 50 in men; $P < .001$), thrombocytopenia was predominantly detected in males. Anemia in

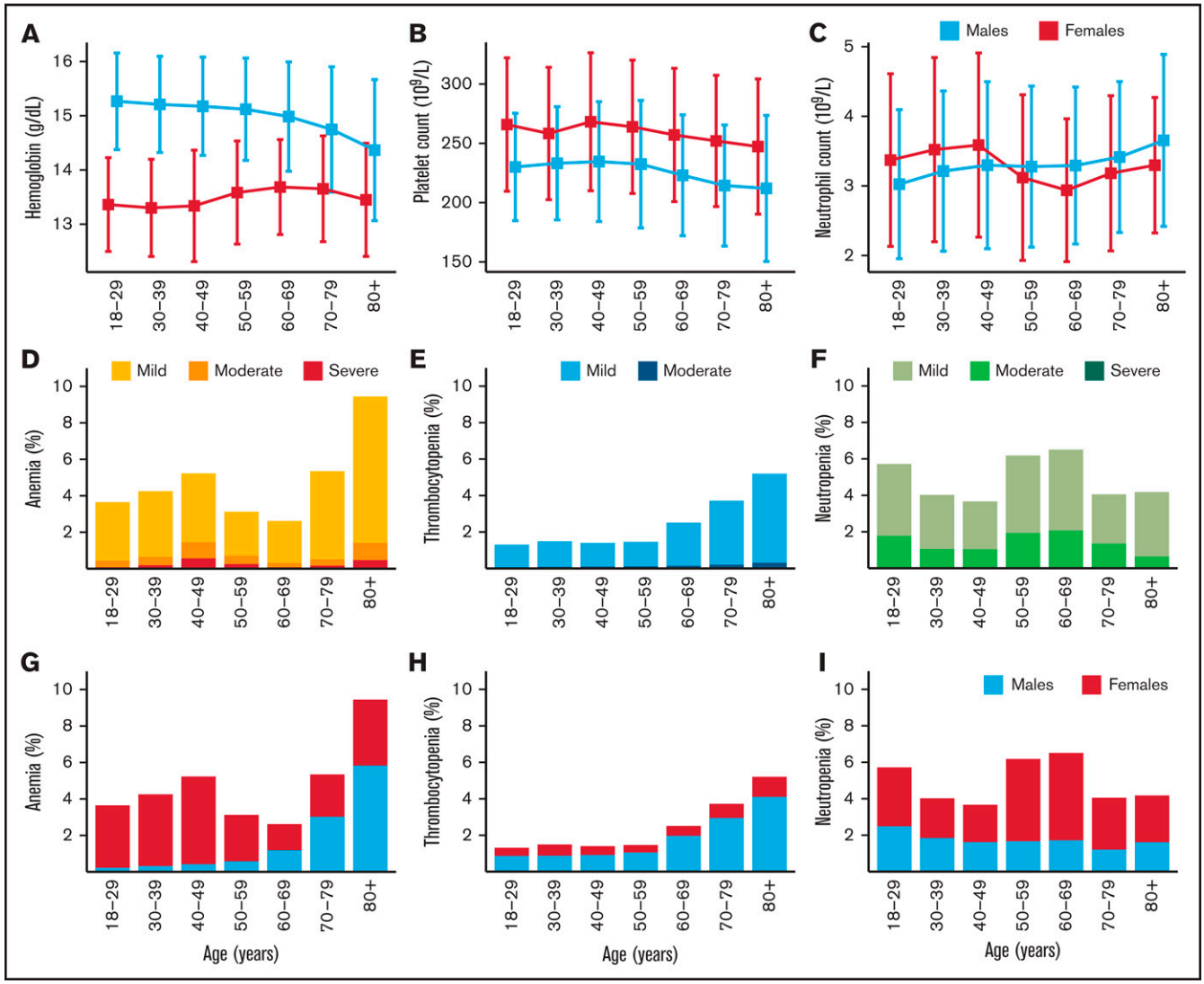

Figure 1. Age-related changes in blood counts and emergence of cytopenias. Mean levels for hemoglobin concentration (A) and platelet (B) and neutrophil (C) counts according to age, shown separately for male and female Lifelines participants. Error bars represent standard deviations. Prevalence of anemia (D), thrombocytopenia (E), and neutropenia (F), according to age. Darker shades represent the prevalence of cytopenias of increasing severity. Contribution of male and female participants to the total prevalence of anemia (G), thrombocytopenia (H), and neutropenia (I) across different age categories. Anemia: hemoglobin concentration <13.0 g/dL in men and <12.0 g/dL in women (moderate: hemoglobin concentration <11.0 g/dL; severe: hemoglobin concentration <10.0 g/dL); thrombocytopenia: platelet count $<150 \times 10^9$/L (moderate: platelet count $<100 \times 10^9$/L); neutropenia: neutrophil count $<1.8 \times 10^9$/L (moderate: neutrophil count $<1.5 \times 10^9$/L; severe: neutrophil count $<.8 \times 10^9$/L).

patients younger than 60 years of age was primarily detected in females (4910/5403; 91% of anemic cases). In contrast, there was an increasing prevalence of anemia in males aged ≥60 years, which was in line with the decreasing hemoglobin concentration for male participants in these age categories. The relative female predominance for neutropenia at age ≥ 60 years (934/1282; 73% of cases) was also consistent with relatively lower neutrophil levels for women in these age categories (mean, 3.0 ± 1.0 vs 3.3 ± 1.1 in males; $P < .001$) (Figure 1G-I).

Unilineage, but not multilineage, cytopenias resolve in the majority of cases

A total of 110 675 Lifelines participants returned for the follow-up visit that was scheduled at a median of 46 months (range, 13-133) later. Among evaluable individuals with any peripheral cytopenia at baseline visit (10 480/14 605), 59% had no detectable cytopenia at follow-up. The evolution of peripheral blood counts for individuals with a cytopenia is displayed in Figure 2. The respective cytopenia was persistent for 1437 of 4356 (33%) individuals with anemia, 769 of 1715 (45%) individuals with thrombocytopenia, and 1897 of 5069 (37%) individuals with neutropenia. Individuals with multilineage cytopenia were more likely to have ≥1 detectable cytopenia at follow-up compared with individuals with unilineage cytopenia (58% vs 40%, respectively; $P < .001$).

Anemia and thrombocytopenia, but not neutropenia, are associated with inferior OS

During a median follow-up period of 8.3 years, 2986 deaths occurred, corresponding to an overall mortality rate of 2.29 per 1000 person-years. Age- and sex-adjusted HRs for all-cause death,

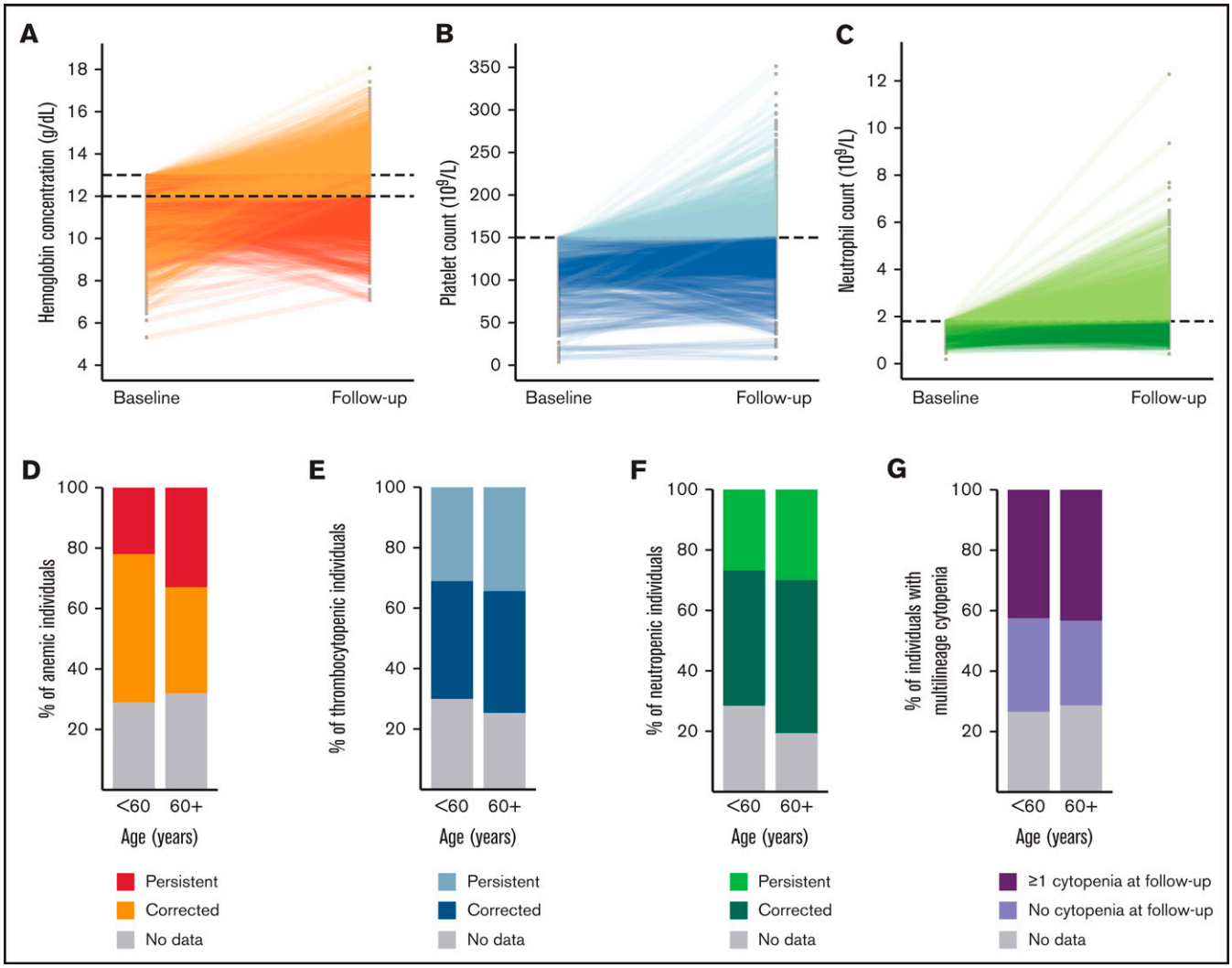

Figure 2. Evolution of peripheral blood counts over time and persistence of detected cytopenias. Evolution of peripheral blood counts over a median period of 36 months for evaluable individuals with anemia (A; n = 4356), thrombocytopenia (B; n = 1715), and neutropenia (C; n = 5069). The proportion of individuals with persistence vs correction of the respective cytopenia for individuals with anemia (D), thrombocytopenia (E), and neutropenia (F), stratified for individuals aged <60 years and ≥60 years. (G) For individuals with >1 cytopenia, the proportion of individuals with ≥1 cytopenia vs absence of cytopenias at follow-up is shown, stratified for individuals aged <60 years and ≥60 years. The proportion of individuals lost to follow-up is shown in gray. Anemia: hemoglobin concentration <13.0 g/dL in men and <12.0 g/dL in women; thrombocytopenia: platelet count $<150 \times 10^9$/L; neutropenia: neutrophil count $<1.8 \times 10^9$/L.

according to the presence of a cytopenia at the baseline visit, are given in Table 2. Anemia, defined using general WHO reference limits, was associated with an increased risk for all-cause mortality (age- and sex-adjusted HR, 1.84; 95% CI, 1.59-2.12; $P < .001$). When using more stringent criteria for moderate anemia (hemoglobin concentrations <11.0 g/dL), HRs for death increased (2.76; 95% CI, 2.04-3.73; $P < .001$). Likewise, individuals with thrombocytopenia (HR, 1.58; 95% CI, 1.32-1.89; $P < .001$) and, especially, moderate thrombocytopenia ($<100 \times 10^9$/L; HR, 4.02; 95% CI, 2.46-6.57; $P < .001$) were at an increased risk for death. In contrast, neutropenia did not affect OS (HR, 0.88; 95% CI, 0.73-1.06; $P = .168$), even when lower cutoffs of 1.5×10^9/L for moderate neutropenia (HR, 0.92; 95% CI, 0.66-1.27; $P = .604$) or 0.8×10^9/L for severe neutropenia (HR, 2.61; 95% CI, 0.37-18.54; $P = .337$) were used (supplemental Figures 2 and 3). When corrected for the number of medications used, as a proxy for comorbidity, the significance of these findings did not change (Table 2). Therefore, in the entire cohort, anemia and thrombocytopenia, but not neutropenia, were associated with an increased risk for all-cause mortality.

We next evaluated whether the observed associations with OS are different for individuals of older age (supplemental Figures 2 and 3). For anemia, inferior OS was associated with older (≥60 years) individuals compared with individuals aged <60 years (P value for interaction <.001). In contrast, although the absolute mortality rates were higher for individuals aged ≥60 years, risk estimates for all-cause mortality were not significantly different for participants aged ≥60 years with thrombocytopenia (P value for interaction = .298) or neutropenia (P value for interaction = .935) compared with younger individuals (supplemental Table 3). Taken together, the risk of death was significantly higher in older individuals (≥60 years) with anemia compared with younger (<60 years) anemic individuals,

Table 2. Risk of all-cause mortality according to the presence of peripheral cytopenias

	Unadjusted		Adjusted*		Adjusted†	
	HR (95% CI)	P	HR (95% CI)	P	HR (95% CI)	P
Anemia (<12.0 g/dL [women] or <13.0 g/dL [men])	1.74 (1.51-2.00)	<.001	1.84 (1.59-2.12)	<.001	1.60 (1.38-1.84)	<.001
Moderate (<11.0 g/dL)	1.71 (1.26-2.30)	<.001	2.76 (2.04-3.73)	<.001	2.41 (1.78-3.27)	<.001
Severe (<10.0 g/dL)	1.60 (0.95-2.71)	.078	2.61 (1.54-4.41)	<.001	2.41 (1.42-4.07)	.001
Thrombocytopenia (<150 × 10^9/L)	2.86 (2.39-3.41)	<.001	1.58 (1.32-1.89)	<.001	1.52 (1.27-1.83)	<.001
Moderate (<100 × 10^9/L)	5.98 (3.66-9.78)	<.001	4.02 (2.46-6.57)	<.001	3.74 (2.29-6.12)	<.001
Neutropenia (<1.8 × 10^9/L)	0.91 (0.76-1.09)	.301	0.88 (0.73-1.06)	.168	0.95 (0.79-1.14)	.548
Moderate (<1.5 × 10^9/L)	0.95 (0.69-1.32)	.763	0.92 (0.66-1.27)	.604	0.98 (0.70-1.35)	.878
Severe (<0.8 × 10^9/L)	1.83 (0.26-12.96)	.547	2.61 (0.37-18.54)	.337	2.81 (0.40-19.93)	.302

Different cutoffs were used to evaluate the increasing severity of cytopenias, as indicated. Individuals with absence of cytopenia for the respective lineage were used as a reference.
*The multivariable model included age and sex as covariates.
†The multivariable model included age, sex, and the number of medications used as covariates.

whereas the risk of death for thrombocytopenia or neutropenia was comparable in both age groups.

When evaluating individuals with multilineage cytopenia, OS was severely affected by the concomitant presence of anemia and thrombocytopenia compared with anemia ($P < .001$) or thrombocytopenia ($P < .001$) alone. This was observed for individuals aged <60 years and ≥60 years (Figure 3A-B). In contrast, the concomitant presence of neutropenia did not affect mortality risks associated with anemia ($P = .140$) or thrombocytopenia ($P = .646$) (Figure 3C-F). Risk estimates for mortality in the presence of multilineage cytopenia were subsequently evaluated in multivariable models, with age and sex included as covariates (supplemental Table 4). Substantially increased mortality risks were found for individuals with anemia and thrombocytopenia (HR, 4.75; 95% CI, 2.98-7.55; $P < .001$). An increased risk for mortality was also observed for individuals with thrombocytopenia and neutropenia (HR, 2.16; 95% CI 1.32-3.54; $P = .002$). In contrast, no increased risk for death was observed for individuals having anemia and neutropenia (HR, 1.29; 95% CI, 0.71-2.33; $P = .401$). The highest risks of death were observed for individuals with pancytopenia (HR, 11.92; 95% CI; 4.96-28.67; $P < .001$).

Higher cumulative incidence of hematological malignancies for individuals with a peripheral cytopenia

Linkage to PALGA yielded 3039 pathology excerpts from 1417 individuals, of whom the majority (2763; 91%) complied with high certainty linkage criteria and were included for further analysis (supplemental Methods).

Over a median follow-up of 7.7 years, 483 individuals were diagnosed with a first incident myeloid (n = 132) and/or lymphoid (n = 356) hematological malignancy after inclusion in the Lifelines study. The overall probability of developing any hematological malignancy at 5 years was 0.19% (95% CI, 0.17-0.21), with a higher cumulative incidence for individuals aged ≥60 years (0.60%; 95% CI, 0.50-0.70). Among individuals with any cytopenia (n = 14 605), a hematological malignancy was diagnosed in 98, with a 5-year probability of 0.50% (95% CI, 0.40-0.60; $P < .001$; supplemental Table 5). Due to the small numbers of events, we were only able to evaluate cumulative incidences for peripheral cytopenias as defined using general reference values.

A higher cumulative incidence of hematological malignancies was observed, especially for older individuals with anemia (5-year probability, 2.5%; 95% CI, 1.4-3.6) and thrombocytopenia (5-year probability, 2.5%; 95% CI, 1.3-3.8; Figure 4A-D). In contrast, neutropenia was not associated with a higher probability of developing hematological malignancies in older individuals ($P = .201$; Figure 4F).

Because the occurrence of cytopenias and the development of hematological malignancies may be affected by age and sex, we performed age- and sex-adjusted competing risk regression (supplemental Tables 5 and 6). In multivariable models, anemia (HR, 2.47; 95% CI, 1.80-3.39) remained significantly associated with a higher cumulative burden of hematological malignancies for individuals aged <60 years and ≥60 years. Likewise, thrombocytopenia in patients aged <60 years (HR, 3.86; 95% CI, 2.24-6.66) and aged ≥60 years (HR, 3.13; 95% CI, 1.91-5.15) remained associated with a higher probability of developing incidental hematological malignancies, which were predominantly of lymphoid origin. For individuals with neutropenia, significant associations were retained for individuals aged <60 years (HR, 2.31; 95% CI, 1.52-3.52), whereas it was confirmed that neutropenia was not associated with a significant higher cumulative incidence of hematological malignancies in individuals aged ≥60 years (HR, 1.52; 95% CI, 0.89-2.61; $P = .130$).

The highest cumulative incidences of hematological malignancies were observed for individuals with multilineage cytopenias (Figure 4G-H). A total of 14 individuals developed a hematological malignancy during follow-up, of whom 4 had pancytopenia and 4 had concurrent anemia and thrombocytopenia. Among individuals of all ages with multilineage cytopenia, the estimated 5-year cumulative incidence of hematological malignancies was 1.6% (95% CI, 0.8-2.5), with an age- and sex-corrected subdistribution HR of 5.13 (95% CI, 3.00-8.79; supplemental Tables 5 and 6).

Hematological malignancies explain a minor fraction of all deaths

Linkage to the national registry succeeded for 152 111 Lifelines participants. Data on cause of death were subsequently available for 2723 of 2739 (99%) deceased participants during the period

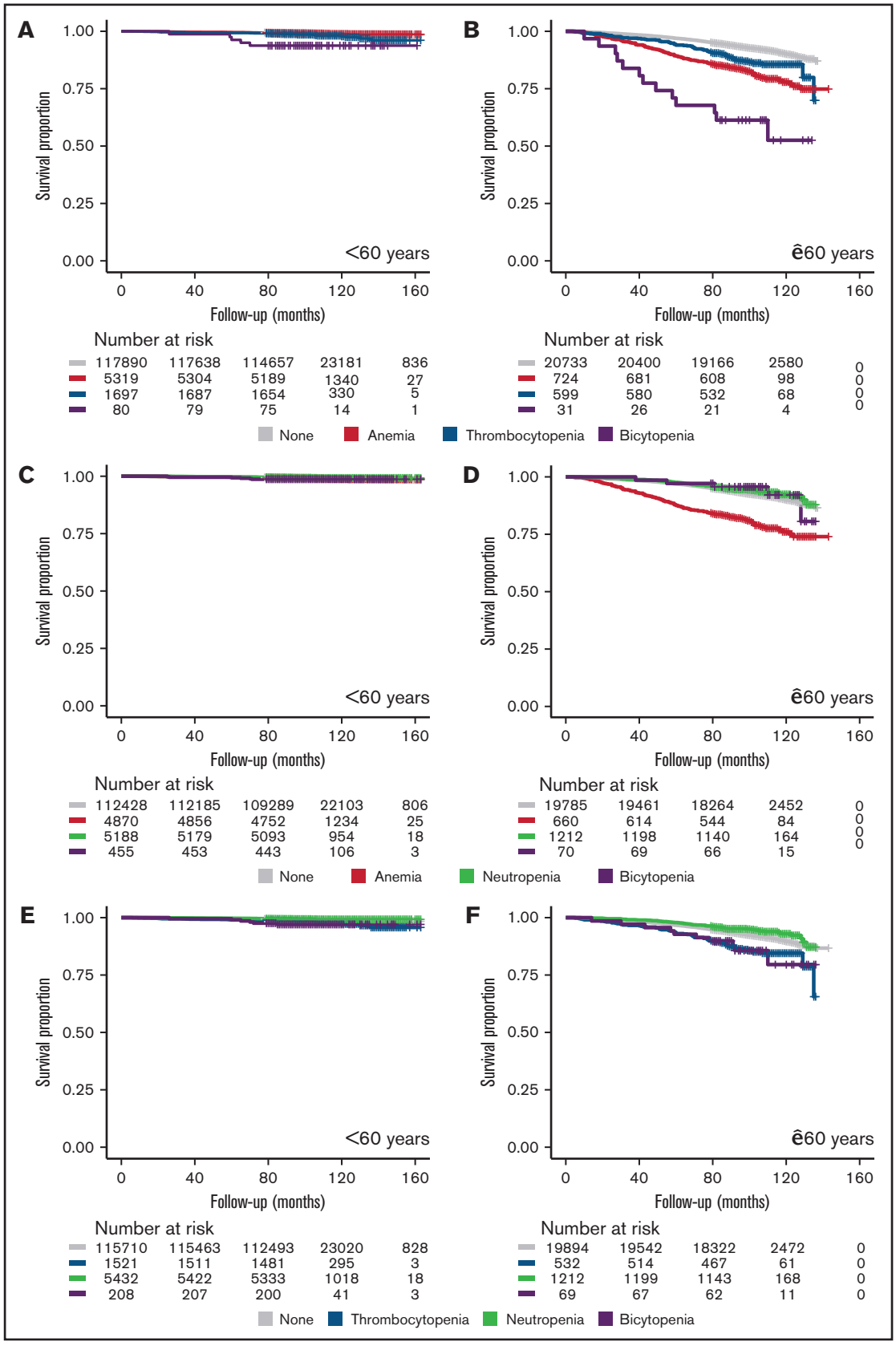

Figure 3. Survival impact of multilineage cytopenias. Kaplan-Meier curves showing OS according to the presence of multilineage cytopenia in individuals aged <60 years (left panels) and ≥60 years (right panels). Individuals were classified as having multilineage (purple) or unilineage cytopenias in the respective lineages or in the absence of the respective cytopenias (gray). (A-B) Anemia and thrombocytopenia. (C-D) Anemia and neutropenia. (E-F) Thrombocytopenia and neutropenia. Anemia: hemoglobin concentration <13.0 g/dL in men and <12.0 g/dL in women; thrombocytopenia: platelet count $<150 \times 10^9$/L; neutropenia: neutrophil count $<1.8 \times 10^9$/L.

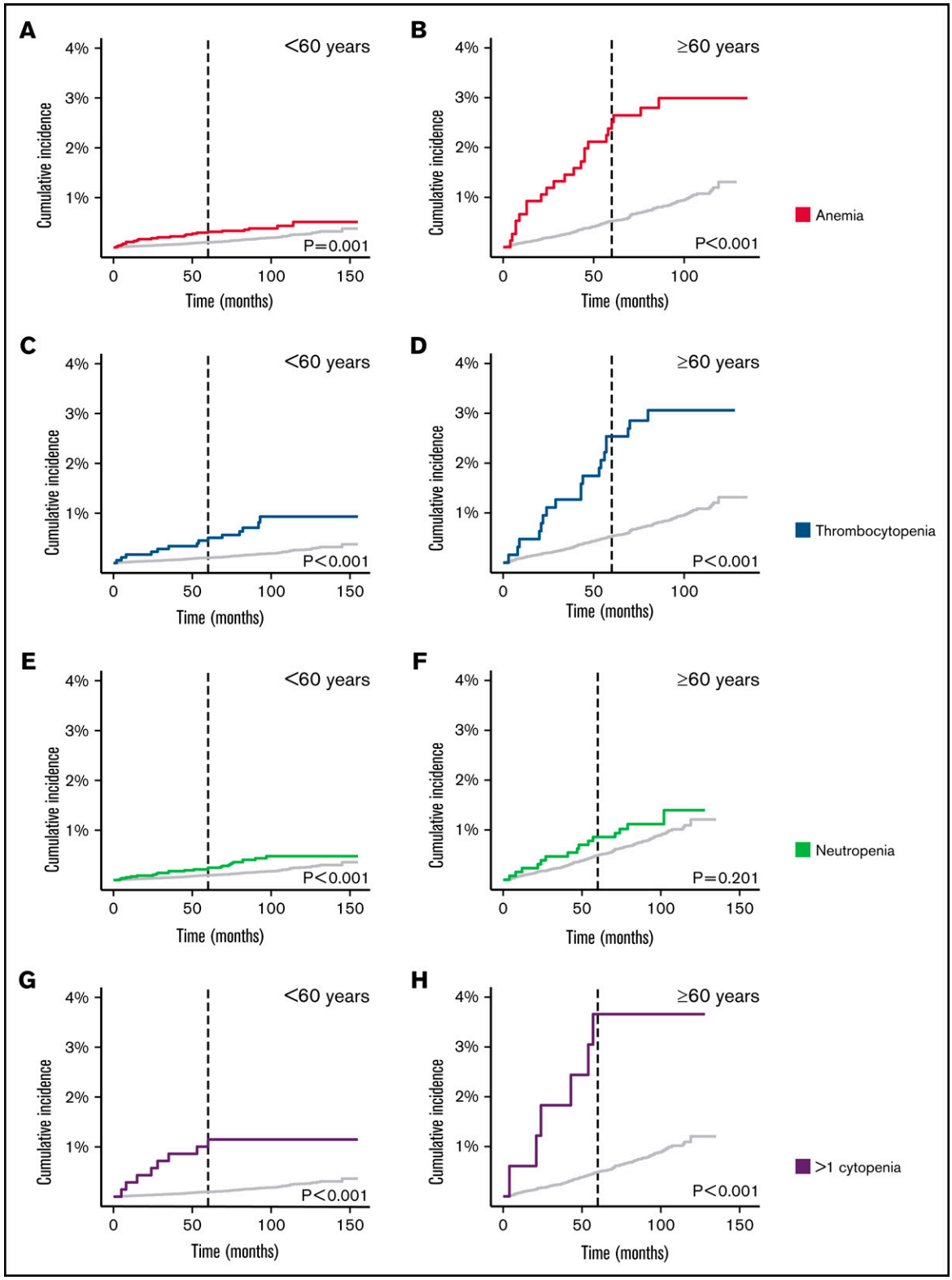

Figure 4. Cumulative incidence of hematological malignancies for community-dwelling individuals with peripheral cytopenias. Cumulative incidence of anemia (A-B), thrombocytopenia (C-D), neutropenia (E-F), or >1 cytopenia (G-H) in individuals aged <60 years and aged ≥60 years. Anemia: hemoglobin concentration <13.0 g/dL in men and <12.0 g/dL in women; thrombocytopenia: platelet count $<150 \times 10^9$/L; neutropenia, neutrophil count $<1.8 \times 10^9$/L. Aalen-Johansen estimates were used for the cumulative incidence function; *P* values were calculated using Gray's test.

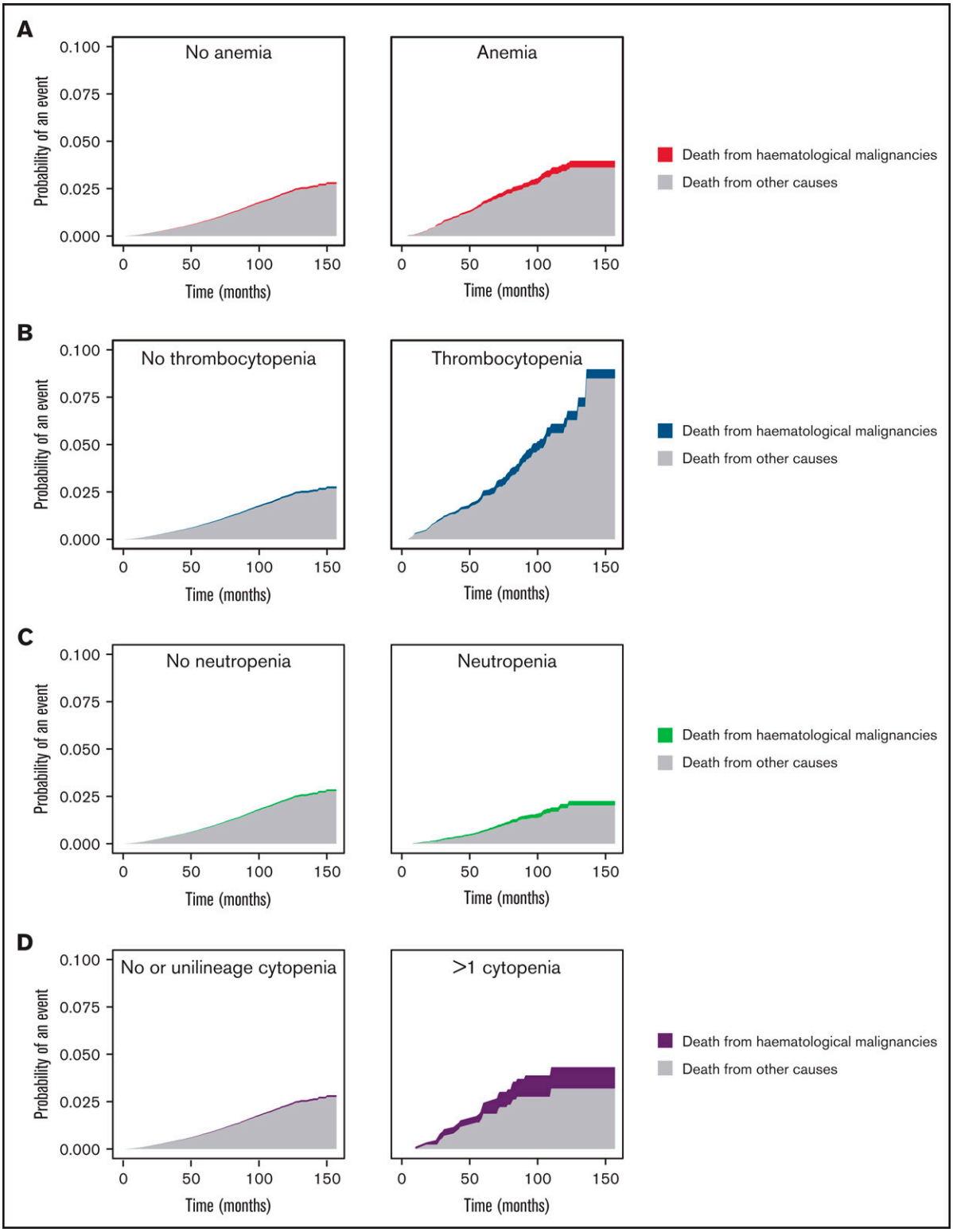

Figure 5. Cumulative incidence of death from hematological malignancies for community-dwelling individuals with and without peripheral cytopenias. Stacked cumulative incidence graphs show the probabilities of mortality from hematological malignancies vs other causes, stratified according to the presence of anemia (A), thrombocytopenia (B), neutropenia (C), or >1 peripheral cytopenia (D). Data on primary cause of death were obtained by linkage to the national registry of death statistics. Results for this analysis are based on calculations by the authors using nonpublic microdata from Statistics Netherlands. Anemia: hemoglobin concentration <13.0 g/dL in men and <12.0 g/dL in women; thrombocytopenia: platelet count <150 × 10^9/L; neutropenia: neutrophil count <1.8 × 10^9/L.

Table 3. Subdistribution hazard ratios for death from hematological malignancies according to the presence of unilineage or multilineage cytopenias in the entire evaluable Lifelines cohort (n = 152 096).

	n*	Unadjusted HR (95% CI)	P	Adjusted HR (95% CI)†	P
Anemia	6156	4.71 (2.83-7.86)	<.001	4.55 (2.67-7.73)	<.001
Thrombocytopenia	2408	7.32 (3.91-13.70)	<.001	3.71 (1.90-7.27)	<.001
Neutropenia	6925	3.41 (1.90-6.14)	<.001	3.41 (1.87-6.20)	<.001
>1 cytopenia	861	18.20 (9.13-36.40)	<.001	14.37 (7.00-29.47)	<.001

Estimates were obtained by competing risk regression, according to the method of Fine and Gray, taking into account death from other causes as a competing risk. Individuals with an absence of the respective cytopenia or a multilineage cytopenia were used as the reference. Anemia: hemoglobin concentration <13.0 g/dL in men and <12.0 g/dL in women; thrombocytopenia: platelet count $<150 \times 10^9$/L; neutropenia: neutrophil count $<1.8 \times 10^9$/L.

*Number of individuals with the respective cytopenia evaluable for cause of death.

†The multivariable model included age and sex as covariates.

from January of 2007 to December of 2019 (supplemental Methods). In total, 110 (4.0%) deaths were attributable to hematological neoplasms. Supplemental Table 8 shows the distribution of deaths from hematological malignancies across the entire Lifelines cohort.

A primary cause of death from hematological malignancies was observed for only a minor fraction of cytopenic individuals: 18 with anemia, 11 with thrombocytopenia, and 13 with neutropenia. The probabilities of death from hematological malignancies were higher for individuals with anemia ($P < .001$), thrombocytopenia ($P < .001$), or neutropenia ($P < .001$) (Figure 5A-C). However, the majority of individuals suffered death from causes other than hematological malignancies (supplemental Figure 4). The probability of death from hematological malignancies was highest among individuals with >1 cytopenia ($P < .001$), with 9 deaths attributable to hematological malignancies (Figure 5D). We further substantiated the association between peripheral cytopenias and mortality from hematological malignancies in a multivariable model with adjustment for age and sex. Anemia (HR, 4.55; 95% CI, 2.67-7.73), thrombocytopenia (HR, 3.71; 95% CI, 1.90-7.27), neutropenia (HR, 3.41; 95% CI, 1.87-6.20) and the presence of >1 cytopenia (HR, 14.37; 95% CI, 7.00-29.47) remained associated with a higher probability of death from hematological malignancies (Table 3). So, although neutropenia was not associated with inferior OS, it was associated with a higher probability of death from hematological malignancies, similar to anemia and thrombocytopenia, although the major proportion of deaths was explained by other causes.

Discussion

Within the well-characterized population-based Lifelines cohort (n = 167 729), we aimed to investigate the occurrence of peripheral cytopenias with aging and its association with the development of hematological disorders in community-dwelling individuals.

The prevalence of anemia in our cohort, using WHO criteria, was comparable with population-based studies in Sweden and Germany,[22,23] although other investigators have reported a higher burden of anemia in older individuals (eg, NHANES cohort).[24-26] For thrombocytopenia, prevalence rates in our study were higher compared with population-based European and outpatient US cohorts.[27-29] These discrepancies might be the result of cohort-specific differences in age, ethnicity, and recruiting procedures. The prevalence of moderate neutropenia (ie, neutrophils $<1.5 \times 10^9$/L) was comparable to a general practice registry cohort of comparable ethnicity.[30,31] However, the prevalence of mild neutropenia (neutrophils $<1.8 \times 10^9$/L) in our population-based cohort was relatively high compared with this general practice–based cohort and might also reflect differences in sample population.[30,31] Severe neutropenia, in accordance with previous studies,[30-32] was very uncommon.

The differential diagnosis of peripheral cytopenias encompasses a wide range of etiologies. When other underlying conditions have been excluded, the presence of an underlying hematological disorder may be suspected, especially in older individuals. Although we were unable to retrieve the medical cause of peripheral cytopenias, we were able to evaluate their natural history in a real-life cohort of individuals representative of the background population. Follow-up data revealed that cytopenias resolved in the majority of individuals. Although individuals with a peripheral cytopenia were at a higher risk for the development of hematological malignancies, the 5-year cumulative incidence of 0.5% is considerably different from risk estimates described in other settings. For example, in a recent study evaluating all patients visiting a hematology outpatient clinic and having a bone marrow biopsy with inconclusive results, a substantially higher proportion of individuals developed a malignant hematological disorder, with a 5-year cumulative incidence of 8.9%.[33] Significantly higher probabilities were observed in another study of patients with unexplained cytopenia after diagnostic work-up for malignancy.[7] We conclude that the risk of developing malignant disease may very well be proportional to the setting in which the cytopenia is observed.

The large sample size of our study allowed for a comprehensive investigation of peripheral blood counts in relation to the emergence of cytopenias with aging. Most cytopenias were mild, with peripheral blood counts just below general reference values. Advancing age was associated with higher prevalences of anemia and thrombocytopenia, consistent with an age-related decline in platelet count and hemoglobin concentrations. In contrast, we observed a slight increase in neutrophil levels with aging and no age-related increase in neutropenia, which is consistent with a myeloid bias upon advancing age.[34] Further, the absence of a decrease in neutrophil levels with age is also in accordance with findings from the population-based NHANES cohort,[32] which indicates that aging hematopoiesis may have differential effects on the various mature hematopoietic lineages. In addition, differences in peripheral blood counts between the sexes translated into differences in cytopenia prevalence. In accordance with previous reports, a major sex dependence was observed for platelet counts.[29,35-37] The impact of this biological variation in peripheral blood counts should be taken into account when defining the boundaries between (pre)malignant hematological disease and the (inevitable) consequences of aging.

Anemia and thrombocytopenia, especially when more stringent cutoffs were used, negatively affected OS. This is in accordance with previous reports.[28,38-44] Although the probability of death from hematological neoplasms was higher for cases with a peripheral cytopenia, the majority of deaths were attributable to other causes. Thus, other underlying conditions primarily affect the prognosis of community-dwelling individuals with anemia or thrombocytopenia and deserve medical attention. Neutropenia, either isolated or in combination with another cytopenia and irrespective of severity, did not affect OS. Few population-based studies have assessed mortality rates for neutropenic individuals. In line with our observations, these studies also did not observe a survival disadvantage for mild neutropenia.[28,30] Cumulative incidences of hematological malignancies were highest for older individuals with anemia and thrombocytopenia, but they were not different for older individuals with neutropenia. A recent study reported a higher risk for neutropenic individuals to develop hematological malignancies, when detected in the primary care setting of the CopDiff cohort.[31] Notably, we did find a higher risk for developing malignancies for younger individuals. Because the analyses in the CopDiff cohort were not stratified by age, the results may not be compared directly. In addition, the consequences of cytopenias in the primary care setting could be different from our community-dwelling population as a result of the selection of more severe cases because those individuals were recruited in the primary care setting. The highest incidences for development and death from hematological malignancies were observed for individuals with multiple cytopenias. In addition, a higher proportion of these individuals had persistent cytopenia, and OS was severely impacted in the presence of concomitant anemia and thrombocytopenia. This is in line with recent results from the CopDiff cohort, in which myeloid malignancies developed almost exclusively in individuals with neutropenia and the concomitant presence of anemia or thrombocytopenia.[31] Taken together, the (hematological) implications of mild neutropenia at an older age may be limited compared with anemia or thrombocytopenia. The presence of multiple cytopenias warrants clinical attention.

With the advent of next-generation sequencing, an additional diagnostic complexity in the hematological evaluation of peripheral cytopenias has emerged.[45] Clonal molecular and cytogenetic aberrations are well-known hallmarks of MDS[6,46,47] and may also be detected in (otherwise healthy) aging individuals, which is now referred to as clonal hematopoiesis (CH).[48,49] CH is detected at a high prevalence in patients with (unexplained) cytopenias who have been referred to a hematology clinic for evaluation[7,50-52] and in community-dwelling individuals with anemia.[53] The condition is now frequently referred to as "clonal cytopenia of unknown significance" (CCUS), and patients presenting with CCUS were shown to have a natural history similar to that of low-risk MDS.[7] However, CCUS may be characterized by a heterogeneous nature and include individuals along the spectrum from a low to a high risk for clonal malignant disorders. The current study adds knowledge to cytopenia levels that might be relevant to identify community-dwelling individuals at risk for these MDS-precursor conditions. Anemia and thrombocytopenia were associated with an increased risk for developing hematological malignancies and adverse outcomes, even when standard hematologic reference values were used. Although neutropenia will be detected in a major proportion of (older) individuals, its relevance for the general prognosis and risk of hematological malignancies in the older general population was very limited. Future studies are needed to evaluate the role of next-generation sequencing in asymptomatic patients or community-dwelling individuals with incidentally discovered cytopenias, for hematological and nonhematological outcomes.

Several limitations of this study need to be acknowledged. First, we were only able to analyze the primary cause of death, which may not include a diagnosed hematological neoplasm when individuals are dying with comorbid conditions. Second, the diagnosis of hematological malignancies was inferred from positive histology reports. For this reason, we cannot exclude the possibility of some incident diagnoses remaining unnoticed (eg, when a bone marrow biopsy was not performed or when a diagnosis was not confirmed by histopathology). Lastly, although the dataset is unique in its large scale, data to discern the etiology of cytopenias were not available, limiting the in-depth analysis of explained and unexplained cases. Strengths of the present study include the large number of subjects, the use of "real-world" data representing the general population, and its prospective design. The implications of anemia, thrombocytopenia, and neutropenia were investigated simultaneously. The cohort was not biased by referral or decision-making to have blood drawn, and there is a high chance that the cytopenia was discovered incidentally at study inclusion. Although caution should be exercised when translating these findings to patients presenting in the hematology clinic, the results are representative and informative for the general population.

In summary, our study provides important insight into the emergence of peripheral cytopenias and associated outcomes at the population level. Biological variation by age and sex should be taken into account when formulating criteria to define clinically relevant cytopenias. In this population-based cohort, hematological malignancies developed in only a very small fraction of cytopenic individuals. Neutropenia was not associated with inferior OS or development of hematological malignancies in older community-dwelling individuals. In contrast, patients with anemia, thrombocytopenia, and, especially with combined cytopenias may benefit from intensified surveillance.

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Authorship

Contribution: I.A.v.Z. and G.H. designed the study and wrote the first draft of the manuscript; I.A.v.Z. performed statistical analyses; A.D. reviewed and coded the pathology reports; and all authors analyzed and interpreted data and approved the final version of the manuscript.

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