The Milan Geriatrics 75+ Cohort Study:

unravelling the determinants of healthy ageing and longevity

Giulia Ogliari

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The research described in this thesis was supported with a personal PhD grant by the Università degli Studi di Milano. Financial support by the Leiden University Medical Center for printing of this thesis is acknowledged.

ISBN: 978-90-6464-989-9

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PROEFSCHRIFT

ter verkrijging van

de graad van Doctor aan de Universiteit Leiden,

op gezag van Rector Magnificus prof.mr. C.J.J.M. Stolker,

volgens besluit van het College voor Promoties

te verdedigen op dinsdag 5 april 2016

klokke 11:15 uur

door

Giulia Ogliari

geboren te Treviglio, Italy

in 1980

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To my family,

Professor Carlo Vergani,

Roelof

The Milan Geriatrics 75+ Cohort Study:

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Thesis chapters

Chapter 1

Introduction

Introduction

An ageing Europe, an ageing Italy

Europe's population is rapidly ageing[1,2]. Adults aged 75 years and over now account for 8.3% of the European population, a proportion which is expected to increase up to 10.7% by 2030[2]. In parallel, the birth rate is decreasing[2]. As a result, the population age structure will dramatically change. The proportion of older adults in non-working age will increase compared to that of adults in working age[2]. In the future, older adults may lack social support from younger generations. Therefore, it will be crucial to preserve functional independence in old age.

This demographic transition is already evident in Italy. The population proportion over 75 years is now 10.7% of the total (6.6 million out of 62 million inhabitants)[2]. Italy's life expectancy at birth is now 79.5 years for men and 84.9 years for women, among the highest in the world[3]. Such structural changes in the population represents a big challenge for both society and medicine[4]. With ageing, the burden of age-related diseases, disability and functional dependency increases[5]. Among age-related diseases, cardiovascular diseases and dementia are prominent[5-7]. Their prevalence dramatically increases with advancing age[5-9]. Both cardiovascular diseases and dementia are leading causes of disability and mortality[5].

It can be postulated that age-related diseases lead to a loss of function of different physiological systems and therefore to a state of frailty, an increased vulnerability to stressors, which eventually results in functional decline or death[10-13]. Conversely, mechanisms that preserve the homeostasis of different physiological systems may favor resilience to stressors, and eventually delay functional decline or death.

The homeostasis of the cardiovascular system is crucial for preserving cognitive and functional status[14-17].

However, the mechanisms behind homeostasis may change with ageing. With ageing, changes in different physiological parameters may occur. The optimal values of these parameters as well as the threshold of disease may shift with age. Blood pressure and thyroid status may be among these parameters. Older adults may benefit from different set-point of homeostasis,

compared to younger adults. A deeper insight in the homeostasis of older adults is necessary to tailor interventions aimed at delaying functional decline and mortality in old age.

The aim of this thesis is to explore the homeostasis of older adults, with emphasis on the cardiovascular system. This thesis will examine the associations of cardiovascular parameters (blood pressure and its variability, heart rate, heart rate variability) and thyroid status with clinically relevant outcomes (functional and cognitive status, mortality).

Blood pressure: shifting the cut-off values

With ageing, systolic blood pressure increases, while diastolic blood pressure increases until the age of 60 years and then gradually decreases[18]. Optimal blood pressure targets in old age are still controversial, as reflected by divergent recommendations in different international guidelines[19-23]. In middle-age, higher blood pressure is strongly and consistently associated with adverse health outcomes, including increased risk for dementia and mortality[24]. However, these associations attenuate or even reverse with ageing[24]. Findings from population-based studies have suggested that these associations may be modified by chronological and biological age[25-30]. Indeed, lower blood pressure may be associated with increased mortality risk in the oldest and in the frailest adults[25-30]. In contrast, findings from trials indicate that antihypertensive treatment effect may not vary according to frailty[31]. For instance, the Hypertension in the Very Elderly Trial (HYVET) showed no difference in antihypertensive treatment benefit between the frailer and the fitter participants[31]. However, HYVET excluded older adults with dementia, thus potentially limiting the generalizability of its findings[31].

Thyroid status: shifting the cut-off values

Thyrotropin (TSH), free thyroxine (fT4) and free triiodotironine (fT3) have profound effects on the ageing process, which may vary according to sex and age[32-34]. Optimal thyroid status in old age is an area of controversy[33,34]. The distribution of TSH progressively shifts towards higher values with ageing[35,36]. This shift may arise from a higher prevalence of occult thyroid disease or from selective survival of individuals with a constitutively lower thyroid status[37,38].

Therefore, it is debated whether the upper reference limit for TSH should be age- and sex-specific[33,34].

Heart rate, heart rate variability

Heart rate variability is the physiological variation in the beat-to-beat time interval[39]. By modulating heart rate and heart rate variability, the autonomic nervous system keeps blood pressure constant within a certain range, so to maintain adequate perfusion to vital organs. In particular, higher heart rate variability is a homeostatic mechanism to buffer detrimental variations in blood pressure in response to stressors[40,41].

Blood pressure variability

Visit-to-visit blood pressure variability is the intra-individual variation in blood pressure measures over different clinic visits[42]. Higher blood pressure variability, independent of mean blood pressure, has been associated with clinical and subclinical vascular organ damage[42]. Higher blood pressure variability may reflect impaired homeostasis, in particular impaired baroreflex function, in the context of central autonomic dysregulation[43]. Furthermore, it may cause oscillations in perfusion of vital organs, including the brain, the heart and the kidney, thus leading to damage of these organs.

The Milan Geriatrics 75+ Cohort Study

Current evidence for the treatment of older adults comes from population-based cohort studies and randomized clinical trials. However, this evidence may not be easily extrapolated to patient populations, whom clinicians encounter in everyday clinical practice (Figure 1). Clinical trials tend to selectively recruit fit older adults with few comorbidities. The HYVET trial aimed at solving the controversies on antihypertensive treatment in the very old individuals, by specifically enrolling adults aged 80 years or over[44]. However, a recent population based study showed that only one out of ten older adults would have been eligible for inclusion in HYVET[45]. Despite HYVET's focussed aim and large sample, the benefits and harms of

antihypertensive treatment in frailer old adults remain a controversial topic. Population-based studies may enrol older adults with a broader spectrum of impairment and co-morbidities. However, also population-based studies may be affected by a response bias, as particularly frail older adults tend to refuse participation in these studies[46]. The generalizability of data from trials and population-based studies to patients' populations is debatable. Clinicians are confronted with lack of data in patients' populations, in which comorbidities, functional and cognitive impairment may be more prevalent and severe, and their interplay within homeostasis more complex.

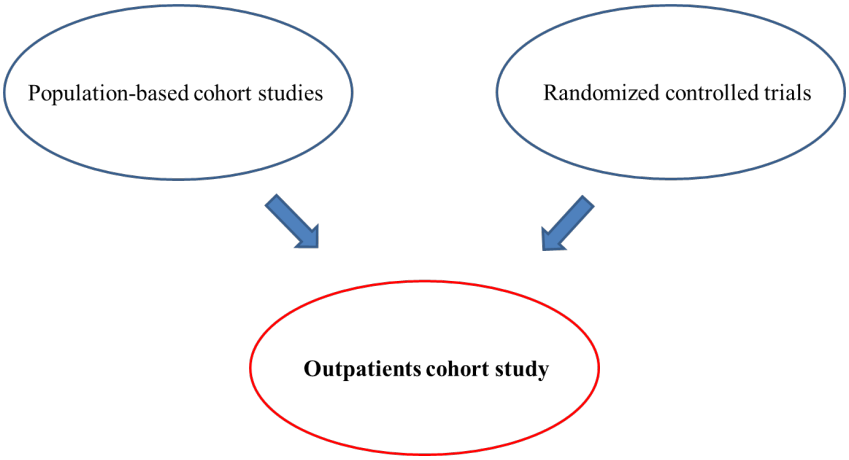


Figure 1. Bridging the gap. Current evidence is derived from population-based cohort studies and randomized controlled trials. Both types of studies may be biased by failing to include frail individuals, due to either lower response rate in the frailer or exclusion criteria. Data are lacking on older outpatients, a potentially diverse population, thus the need for an outpatients cohort study.

To bridge the gap between current evidence and clinical practice needs, we designed the Milan Geriatrics 75+ Cohort Study, a prospective hospital-based outpatient cohort study (Figure 1). This study included 1861 men and women aged 75 years and older who were consecutively referred for a first comprehensive geriatric visit to the Geriatric Unit of the IRCCS Ca' Granda, Milan, Italy, in the period between January 3, 2000 and March 25, 2004. These participants routinely underwent an extensive standardized structured medical examination and comprehensive geriatric assessment. As the Italian health care system guarantees universal

coverage, the Milan Geriatrics 75+ Cohort Study represents the population seeking geriatric care with no restriction based on socio-economic status[47].

The Milan Geriatrics 75+ Cohort Study enrolled mainly women (about two thirds of participants). This may have resulted from higher life expectancy, higher prevalence of comorbidities such as dementia, and higher health care utilization in women compared to men[48,49]. This significant proportion of women allowed us to explore sex-differences in the association between thyroid status and mortality.

The PROSPER Study

The PROspective Study of Pravastatin in the Elderly at Risk (PROSPER) was a randomised, double blind, placebo controlled trial designed to investigate the effect of pravastatin in the prevention of vascular events[50,51]. The PROSPER cohort included older adults aged 70-82 years with pre-existing, or risk factors for, cardiovascular disease, from three collaborating centres in Ireland, Scotland, and the Netherlands. Approximately half of the participants had a diagnosis of cardiovascular disease, defined as myocardial infarction or stable angina, intermittent claudication, stroke or transient ischaemic attack, or previous vascular surgery. The rest of the participants had one or more major cardiovascular risk factors, defined as hypertension, cigarette smoking, or diabetes mellitus. The PROSPER participants had high functional and cognitive status at baseline. Therefore, the PROSPER cohort allowed us to explore the associations between cardiovascular risk factors (heart rate, heart rate variability and blood pressure variability) and functional decline in a cohort with high baseline functional status and at high risk for cardiovascular disease.

Outline of this thesis

Chapter 2, 3 and 4 report findings from the Milan Geriatrics 75+ Cohort Study.

Chapter 2 explores the association between blood pressure and cognition, and whether it varies according to age and functional status.

Chapter 3 examines the relationship between blood pressure and mortality risk, and whether it varies according to functional and cognitive status.

Chapter 4 investigates the association between thyroid status and mortality risk in euthyroid older adults, and whether it differs by sex and age.

Chapter 5 and 6 report findings from the PROSPER cohort.

Chapter 5 presents new evidence on the association of heart rate and heart rate variability with functional decline in older adults at high risk of cardiovascular disease.

Chapter 6 analyses the relationship between blood pressure variability and functional decline in older adults at high risk of cardiovascular disease.

Chapter 7 summarises and discusses the main findings of this thesis.

References

1. United Nations, Department of Economic and Social Affairs, Population Division (2013). World Population Ageing 2013. ST/ESA/SER.A/348.
2. United States Census Bureau. International Programs. International Data Base. Available at: http://www.census.gov/population/international. Accessed October 27, 2015.
3. Vaupel JW. The remarkable improvements in survival at older ages. Philos Trans R Soc Lond B Biol Sci 1997;352:1799–804.
4. Christensen K, Doblhammer G, Rau R, et al. Ageing populations: the challenges ahead. Lancet 2009;374:1196–208.

5. World Health Organization. The global burden of disease: 2004 update. Available at http://www.who.int/healthinfo/global_burden_disease/2004_report_update/en/index.html. Accessed October 30, 2015.
6. Global Atlas on Cardiovascular Disease Prevention and Control. Mendis S, Puska P, Norrving B editors. World Health Organization, Geneva 2011.
7. World Health Organization and Alzheimer's Disease International. Dementia: A Public Health Priority, 2012. Available at http://www.who.int/mental_health/publications/dementia_report_2012/en. Accessed October 27, 2015.
8. Berr C, Wancata J, Ritchie K. Prevalence of dementia in the elderly in Europe. Eur Neuropsychopharmacol 2005;15:463-71.
9. Mozaffarian D, Benjamin EJ, Go AS, et al; American Heart Association Statistics Committee and Stroke Statistics Subcommittee. Heart disease and stroke statistics--2015 update: a report from the American Heart Association. Circulation 2015;131:e29-322.
10. Fried LP, Tangen CM, Walston J, et al; Cardiovascular Health Study Collaborative Research Group. Frailty in older adults: evidence for a phenotype. J Gerontol A Biol Sci Med Sci 2001;56:M146-56.
11. Li Q, Wang S, Milot E, et al. Homeostatic dysregulation proceeds in parallel in multiple physiological systems. Aging Cell 2015 Sep 29. doi: 10.1111/acel.12402. [Epub ahead of print]
12. Mitnitski AB, Mogilner AJ, MacKnight C, et al. The mortality rate as a function of accumulated deficits in a frailty index. Mech Ageing Dev 2002;123:1457-60.
13. McEwen BS, Stellar E. Stress and the individual. Mechanisms leading to disease. Arch Intern Med 1993;153:2093-101.
14. Eggermont LH, de Boer K, Muller M, et al. Cardiac disease and cognitive impairment: a systematic review. Heart 2012;98:1334-40.
15. Qiu C, Winblad B, Marengoni A, et al. Heart failure and risk of dementia and Alzheimer disease: a population-based cohort study. Arch Intern Med 2006;166:1003-8.
16. Jefferson AL, Beiser AS, Himali JJ, et al. Low cardiac index is associated with incident dementia and Alzheimer disease: the Framingham Heart Study. Circulation 2015;131:1333-9.
17. Jefferson AL, Himali JJ, Beiser AS, et al. Cardiac index is associated with brain aging: the Framingham Heart Study. Circulation 2010;122:690-7.

18. Burt VL, Whelton P, Roccella EJ, et al. Prevalence of hypertension in the US adult population. Results from the Third National Health and Nutrition Examination Survey, 1988-1991. Hypertension 1995;25:305-13.
19. James PA, Oparil S, Carter BL, et al. 2014 evidence-based guideline for the management of high blood pressure in adults: report from the panel members appointed to the Eighth Joint National Committee (JNC 8). JAMA 2014;311:507-20.
20. Mancia G, Fagard R, Narkiewicz K, et al. 2013 ESH/ESC guidelines for the management of arterial hypertension: the Task Force for the Management of Arterial Hypertension of the European Society of Hypertension (ESH) and of the European Society of Cardiology (ESC). Eur Heart J 2013;34:2159-2219.
21. National Institute for Health and Clinical Excellence. Hypertension (CG127). http://www.nice.org.uk/guidance/cg127 . Accessed October 08, 2015.
22. Canadian Hypertension Education Program. 2015. https://www.hypertension.ca/images/CHEP_2015/CHEP2015_Full_EN.pdf . Accessed October 08, 2015.
23. American Diabetes Association. Standards of medical care in diabetes—2015. Diabetes Care 2015;38(Suppl. 1):S49–S57
24. Lewington S, Clarke R, Qizilbash N, et al; Prospective Studies Collaboration. Age-specific relevance of usual blood pressure to vascular mortality: a meta-analysis of individual data for one million adults in 61 prospective studies. Lancet 2002;360:1903-13.
25. Satish S, Freeman DH Jr, Ray L, et al. The relationship between blood pressure and mortality in the oldest old. J Am Geriatr Soc 2001;49:367–374.
26. Boshuizen HC, Izaks GJ, van Buuren S, et al. Blood pressure and mortality in elderly people aged 85 and older: community based study. BMJ 1998;316:1780-4.
27. van Bemmel T, Gussekloo J, Westendorp RG, et al. In a population based prospective study, no association between high blood pressure and mortality after age 85 years. J Hypertens 2006;24:287–292.
28. Odden MC, Peralta CA, Haan MN, et al. Rethinking the association of high blood pressure with mortality in elderly adults: the impact of frailty. Arch Intern Med 2012;172:1162-8.
29. Peralta CA, Katz R, Newman AB, et al. Systolic and diastolic blood pressure, incident cardiovascular events, and death in elderly persons: the role of functional limitation in the Cardiovascular Health Study. Hypertension 2014;64:472-80.

30. Post Hospers G, Smulders YM, Maier AB, et al. Relation between blood pressure and mortality risk in an older population: role of chronological and biological age. J Intern Med 2015;277:488-97.
31. Warwick J, Falaschetti E, Rockwood K, et al. No evidence that frailty modifies the positive impact of antihypertensive treatment in very elderly people: an investigation of the impact of frailty upon treatment effect in the HYpertension in the Very Elderly Trial (HYVET) study, a double-blind, placebo-controlled study of antihypertensives in people with hypertension aged 80 and over. BMC Med 2015;13:78.
32. Taylor PN, Razvi S, Pearce SH, et al. Clinical review: A review of the clinical consequences of variation in thyroid function within the reference range. J Clin Endocrinol Metab 2013;98:3562-71.
33. Jonklaas J, Bianco AC, Bauer AJ, et al. Guidelines for the treatment of hypothyroidism: prepared by the american thyroid association task force on thyroid hormone replacement. Thyroid 2014;24:1670-751.
34. Pearce SH, Brabant G, Duntas LH, et al. 2013 ETA Guideline: Management of Subclinical Hypothyroidism. Eur Thyroid J 2013;2:215-28.
35. Hollowell JG, Staehling NW, Flanders WD, et al: Serum TSH, T4, and thyroid antibodies in the United States population (1988 to 1994): National Health and Nutrition Examination Survey (NHANES III). J Clin Endocrinol Metab 2002; 87: 489–499.
36. Surks MI, Hollowell JG. Age-specific distribution of serum thyrotropin and antithyroid antibodies in the US population: implications for the prevalence of subclinical hypothyroidism. J Clin Endocrinol Metab 2007;92:4575-82.
37. Asvold BO, Vatten LJ, Midthjell K, et al. Serum TSH within the reference range as a predictor of future hypothyroidism and hyperthyroidism: 11-year follow-up of the HUNT Study in Norway. J Clin Endocrinol Metab 2012;97:93-99.
38. Atzmon G, Barzilai N, Surks MI, et al. Genetic predisposition to elevated serum thyrotropin is associated with exceptional longevity. J Clin Endocrinol Metab 2009;94:4768-75.
39. Heart rate variability. Standards of measurement, physiological interpretation, and clinical use. Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology. Eur Heart J 1996;17:354-81.
40. Sloan RP, Demeersman RE, Shapiro PA, et al. Cardiac autonomic control is inversely related to blood pressure variability responses to psychological challenge. Am J Physiol 1997; 272:H2227-32.

41. Sloan RP, DeMeersman RE, Shapiro PA, et al. Blood pressure variability responses to tilt are buffered by cardiac autonomic control. Am J Physiol 1997;273:H1427-31.
42. Rothwell PM. Limitations of the usual blood-pressure hypothesis and importance of variability, instability, and episodic hypertension. Lancet 2010; 375: 938-48.
43. Meel-van den Abeelen AS, Lagro J, Gommer ED, et al. Baroreflex function is reduced in Alzheimer's disease: a candidate biomarker? Neurobiol Aging 2013; 34: 1170-6.
44. Beckett NS, Peters R, Fletcher AE, et al; HYVET Study Group. Treatment of hypertension in patients 80 years of age or older. N Engl J Med 2008;358:1887-98.
45. Jacobs JM, Stessman J, Ein-Mor E, et al. Hypertension and 5-year mortality among 85-year-olds: the Jerusalem Longitudinal Study. J Am Med Dir Assoc 2012;13:759.e1-6.
46. der Wiel AB, van Exel E, de Craen AJ, et al. A high response is not essential to prevent selection bias: results from the Leiden 85-plus study. J Clin Epidemiol 2002;55(11):1119-25.
47. Lo Scalzo A, Donatini A, Orzella L, et al. Italy: Health system review. Health Systems in Transition, 2009; 11(6)1-216.
48. Gao S, Hendrie HC, Hall KS, et al. The relationships between age, sex, and the incidence of dementia and Alzheimer disease: a meta-analysis. Arch Gen Psychiatry 1998;55:809-15.
49. Hubbard RE, Rockwood K. Frailty in older women. Maturitas 2011;69:203-7.
50. Shepherd J, Blauw GJ, Murphy MB, et al. The design of a prospective study of Pravastatin in the Elderly at Risk (PROSPER). PROSPER Study Group. PROspective Study of Pravastatin in the Elderly at Risk. Am J Cardiol 1999;84:1192-1197.
51. Shepherd J, Blauw GJ, Murphy MB, et al. Pravastatin in elderly individuals at risk of vascular disease (PROSPER): a randomised controlled trial. Lancet 2002;360:1623-1630.

Chapter 2

Blood pressure and cognition

Manuscript based on this chapter has been published as:

Ogliari G, Sabayan B, Mari D, Rossi PD, Lucchi TA, de Craen AJ, Westendorp RG. Age- and Functional Status-Dependent Association Between Blood Pressure and Cognition: The Milan Geriatrics 75+ Cohort Study. J Am Geriatr Soc. 2015;63(9):1741-8.

ABSTRACT

Objectives: To evaluate whether the relationship between blood pressure (BP) measures and cognitive function is different according to age and functional status in older outpatients.

Design: Cross-sectional.

Setting: Outpatient hospital-based Milan Geriatrics 75+ Cohort Study.

Participants: Individuals aged 75 and older (N = 1,540).

Measurements: Blood pressure, Mini-Mental State Examination (MMSE), basic activities of daily living (ADLs), and instrumental activities of daily living (IADLs) were assessed. Associations between BP measures and MMSE score were first analyzed in the total population using linear regression models and were then further examined according to strata of age, ADLs, and IADLs. All analyses were adjusted for sociodemographic factors and presence of comorbidities.

Results: In the total population, higher systolic BP (SBP), diastolic BP (DBP), pulse pressure (PP), and mean arterial pressure (MAP) were all associated with higher MMSE score (all $P < .05$). Each 10-mmHg higher SBP and DBP was associated with a 0.26- and 0.55-point higher MMSE score, respectively. The associations between MMSE score and SBP, DBP, and MAP differed materially according to strata of age and functioning and were most pronounced in those aged 85 and older, with ADL impairments, and with IADL impairments.

Conclusion: Higher BP is associated with better cognitive function in the oldest old and in those with impaired functional status.

INTRODUCTION

Controversy persists on the relationship between blood pressure (BP) and cognitive function in old age[1]. Midlife hypertension has been consistently associated with an increased risk for cognitive impairment and dementia in later life[2-5]. On the contrary, data regarding the association between BP and cognition in older adults are conflicting. Some population-based observational studies have shown an inverse association between higher BP and cognitive function[6], whereas others have shown a direct association[7–9] or no association[10]. Whether this heterogeneity reflects differences in age[11] and level of frailty[12] of the participants is debated. It has been suggested that higher BP may be needed to maintain brain perfusion in biologically older individuals with widespread atherosclerotic vascular damage[13].

Most of the evidence in the literature is for older adults in population-based studies. Less is known about older adults who require outpatient medical assistance. The generalizability of data from population-based studies to clinical practice is questionable. Older outpatients may be frailer than older adults in the general population. In everyday clinical practice, healthcare professionals are confronted with these outpatients' needs. It is of critical importance to investigate this potentially diverse population. Therefore, the current authors investigated the relationship between BP and cognitive function in the Milan Geriatrics 75+ Cohort Study, an outpatient hospital-based cohort study. The objective was to assess whether higher BP is associated with better cognitive function in geriatric outpatients over a wide range of age and functional dependency.

METHODS

Study Design and Participants

The Milan Geriatrics 75+ Cohort Study is an outpatient hospital-based prospective cohort study of outpatients of the Geriatric Unit of the IRCCS Ca' Granda Foundation Maggiore Policlinico Hospital, Milan, Italy. Between January 3, 2000, and March 25, 2004, 3,608 new consecutive outpatients visited the Geriatric Unit. They routinely underwent an extensive standardized structured medical examination and comprehensive geriatric assessment. With the informed consent of these individuals, data were collected in structured paper records that were consecutively numbered and stored in the Geriatric Archive; 3,499 (97.0%) paper records were

retrieved. Of these, 2,267 were for people aged 75 and older at the time of the first visit. Seventy-four individuals had no comprehensive geriatric assessment, and 332 had neither a Mini-Mental State Examination14 (MMSE) nor an activities of daily living[15] (ADLs) score; these individuals were excluded from the final cohort. Therefore, the Milan Geriatrics 75+ Cohort Study includes 1,861 patients aged 75 and older. The current study included 1,540 individuals for whom BP and cognitive evaluation were available. The medical ethical committee of IRCCS Ca' Granda approved the study.

Comprehensive Geriatric Assessment

Outpatients accessed the Geriatric Unit only through the referral for a geriatric visit by a physician (their general practitioner in the majority of cases). The reason for first visit was recorded. Outpatients were required to bring all current medications and all medical documents including letters of discharge from acute-care hospitals, rehabilitation centers or emergency departments, drug prescriptions, reports of visits with other physicians, and reports of the Italian Commissions for the Ascertainment of Civil Disability to their first visit, which was with a trained physician and lasted 2 hours. Physicians collected demographic data, physiological anamnesis, past and present medical history, and current medication use in paper records through structured multiple-choice lists of demographic variables and comorbidities. Physicians also performed a basic neurological examination, took anthropometric measurements, and evaluated functional and cognitive status. Laboratory tests were ordered if recent ones were not available, and the results were recorded. Physicians registered chronic, cyclical, and as-needed drugs, including prescription and over-the-counter medications. Antihypertensive drugs were defined according to Anatomical Therapeutic Chemical (ATC) classification codes C02, C03, C07, C08, and C09[16]. Close relatives frequently accompanied participants to the first visit and acted as informants for the validation of data on functional status and drug assumption.

Blood Pressure

Arterial BP was measured during the first visit using a mercury sphygmomanometer at heart level, with an adjustable cuff, in the seated position, after at least 5 minutes of rest and no

vigorous exercise in the preceding 30 minutes. A special cuff was available for obese subjects. Systolic BP (SBP) and diastolic BP (DBP) were manually auscultated. Pulse pressure (PP) was calculated as SBP minus DBP and mean arterial pressure (MAP) as 1/3SBP + 2/3DBP.

Cognitive and Functional Status

Cognitive function was assessed using the 30-item Mini-Mental State Examination (MMSE)[14]. Functional status was assessed using the Katz ADLs15 and Lawton IADLs[17] questionnaires. ADLs included six items (rising or lying down, feeding, dressing, bathing, toileting, urinary and fecal continence), and IADLs included eight items (using a telephone, shopping, doing housework, doing laundry, preparing meals, using transportation, taking medications, managing money). ADL scores range from 0 to 6 and IADL scores from 0 to 8, with 0 indicating total dependence and the maximum score total independence. Information on functional status was checked with close informants.

Comorbidities and Lifestyle Factors

Hypertension was defined as diagnosis of hypertension or treatment with antihypertensive drugs. Coronary heart disease was defined according to a history of acute myocardial infarction or angina pectoris or therapy with nitrates. History of transient ischemic attack or stroke, diabetes mellitus, atrial fibrillation, claudication, and Parkinson's disease was confirmed using medical documents. Diagnosis of probable Alzheimer's disease was based on international criteria[18,19]. Dementia with Lewy bodies, frontotemporal dementia, and primary progressive aphasia were classified as other neurodegenerative conditions. Alcohol abuse was defined as intake of 70 g of alcohol per day or more. Cancer was defined according to a diagnosis within the previous 5 years. Glomerular filtration rate (GFR), which is an index of renal function, was calculated using the Modification of Diet in Renal Disease Study Group formula[20]. Symptoms of anxiety and depression were self-reported or stated in medical documents. Smoking was dichotomized as never or ever (current or previous). Education was defined as years of school attended.

Medications

Number of medications was defined as the number of drugs taken chronically or cyclically. Antihypertensive drugs were defined as ATC classification codes C02 (antiadrenergics), C03 (diuretics), C07 (beta-blockers), C08 (calcium-channel blockers), and C09 (agents acting on the renin-angiotensin system)[16]. Psychotropic drugs were defined as ATC classification codes N05A (antipsychotics), N05B (anxiolytics), N05C (sleep-inducers or sedatives), and N06A (antidepressants).

Statistical Analysis

In summary statistics, categorical variables were reported as percentages and continuous variables as medians and interquartile ranges (IQRs) when skewed. Linear regression models were used to analyze associations between variables of interest. Analyses were performed in four steps. Model 1 presents unadjusted MMSE mean scores. In Model 2, a minimally adjusted model, analyses were adjusted for age, sex, and education. In Model 3, analyses were adjusted for relevant comorbidities and medication use; each variable was entered in the model separately. In Model 4, a fully adjusted model, analyses were further adjusted for renal function. The relationship between BP measures (predictors) and MMSE score (dependent variable) was examined in total population and within age, ADL, IADL, and BP control strata. Three age strata were defined (75–79, 80–84, ≥85). The total population was divided into two ADL strata (preserved ADL function (ADL score = 6); impaired ADL function (ADL score < 6)) and two IADL strata (IADL score < 5 (median); IADL score ≥5). Subjects were classified into three groups of BP control: normotension (no history of hypertension, SBP < 140 mmHg, DBP < 90 mmHg), controlled hypertension (history of hypertension, SBP < 140 mmHg, DBP < 90 mmHg), and uncontrolled hypertension (history of hypertension, SBP ≥ 140 mmHg, DBP ≥ 90 mmHg). Interaction between BP measures and age, ADLs, and IADLs in relation to cognition was assessed. Interaction terms were calculated by multiplying BP measures by age and ADL and IADL scores, using age and ADL and IADL scores as continuous variables. Sensitivity analyses were performed after exclusion of SBP and DBP outliers; outliers were subjects with SBP or DBP measurements 2 standard deviations or more below or above the

mean of the total population. All analyses were performed using SPSS version 20.0.0 (SPSS, Inc., Chicago, IL).

RESULTS

Table 1 summarizes the characteristics of participants at first visit according to tertile of SBP. The median age of the study population was 82 (range 75–101), and 70% were female. Median SBP was 145 mmHg, and median DBP was 80 mmHg. Participants with higher SBP were more likely to be female and had a higher prevalence of hypertension and antihypertensive use. Participants with higher SBP were more likely to use alpha-antiadrenergics and angiotensin-converting enzyme inhibitors or angiotensin II antagonists; participants with lower SBP used antipsychotics more frequently. Participants in the lowest tertile of SBP had the highest prevalence of Parkinson's disease (all $P < .05$).

Table 2 shows the association between BP measures and cognitive function in the total population. Higher SBP, DBP, PP, and MAP were associated with higher MMSE score in all models of adjustment (all $P < .05$). In the fully adjusted model, a 10-mmHg increase in SBP was associated with a 0.26-point higher MMSE score (95% confidence interval (CI) = 0.13–0.40), a 10-mmHg increase in PP with a 0.20-point higher MMSE score (95% CI = 0.03–0.37), a 10-mmHg increase in DBP with a 0.55-point higher MMSE score (95% CI = 0.27–0.83), and a 10-mmHg increase in MAP with a 0.50-point higher MMSE score (95% CI = 0.27–0.74). In the fully adjusted model, subjects in the lowest SBP tertile (SBP <140 mmHg) had the lowest MMSE score; subjects in the lowest and middle DBP tertiles (DBP <90 mmHg) had lower MMSE scores than those in the highest DBP tertile.

Table 3 presents the age-stratified analyses of the association between BP measures and cognitive function. Age significantly modified the association between MMSE score and SBP, DBP, and MAP (all P-values for interaction < .05 except in Model 4 for SBP ($P = .15$) and MAP ($P = .05$)). The interaction between age and PP was not significant. In all adjusted models, the association between higher SBP, DBP, and MAP and MMSE score was most pronounced in participants aged 85 and older.

The modifying effect of functional status (ADL and IADL scores) on the relationship between BP measures and MMSE score is shown in Figure 1. In the unadjusted model, all P-values for

interaction between all BP measures and ADL/IADL score were less than .05 (Figure 1). In the fully adjusted model, P-values for interaction between SBP, DBP, and MAP and ADL score were less than .10; all P-values for interaction between all BP measures and IADL score were less than .05. In all models, higher BP measures were associated with higher MMSE score in subjects with at least partial dependence in ADLs (ADL score < 6) but not in subjects with full independence (ADL score = 6). Similarly, higher BP was related to better cognitive function in subjects with worse IADL performance (IADL score < 5). Conversely, no association was observed in those with better IADL score (IADL score ≥ 5). Estimates of mean MMSE scores in Figure 1 are derived from the unadjusted model; results were similar in the fully adjusted model (data not shown).

No difference in MMSE scores was observed in the fully adjusted model between participants with normotension and those with controlled hypertension, between participants with normotension and those with uncontrolled hypertension, or between participants with controlled hypertension and those with uncontrolled hypertension (data not shown).

In sensitivity analyses after exclusion of BP outliers (n = 105), higher SBP, DBP, PP, and MAP remained associated with higher MMSE score in the total population and in subjects with impaired ADL or IADL status, even after full adjustment (all P < .05, data not shown).

DISCUSSION

Higher BP measures were associated with better cognitive function in outpatients aged 75 and older and particularly in those aged 85 and older. The association was significantly stronger in those with impaired functional status, as measured by internationally validated ADL and IADL scale scores.

Both age and functional status modified the relationship between BP and cognitive function. The correlation between SBP, DBP, and MAP and MMSE score becomes more pronounced with increasing age. In those aged 85 and older, higher BP measures were consistently associated with higher MMSE scores. Likewise, the positive association between BP measures and cognitive function was detected in participants with worse functional impairment, although this association was absent in those with better preserved functional status.

The results of this study are consistent with those of earlier reports showing that lower BP was associated with worse cognitive performance in the oldest adults[7] and in centenarians[8]. The age-dependent relationship between BP and cognitive function has been previously hypothesized[1]. The modifying effect of functional status is a novel finding. In the population-based Leiden 85-plus Study, higher SBP and PP were associated with lower annual decline in MMSE score in the oldest adults with greater physical disability, although interactions were not significant[7]. All of these studies have used population samples. The current study showed that the positive association between high BP and good cognitive performance in frail older adults can be extrapolated to the outpatient clinic.

These findings may have different biological explanations. First, cognitive impairment itself lowers BP. The central nervous system is involved in BP regulation; brain atrophy and Alzheimer-type lesions in the prefrontal areas involved in central BP regulation may cause a decline in BP[21]. Alternatively, low BP and cognitive impairment share common risk factors such as decreasing cardiac function. However, in the current study, the associations between BP measures and cognitive performance remained significant after adjustment for risk factors and comorbidities that affect cardiac function. Finally, low BP may increase the risk of worse cognitive function. Episodic or sustained hypotension, and possibly excessive treatment of hypertension, may induce brain hypoperfusion, leading to ischemia and hypoxia, which may enhance the development of neurodegenerative processes[22]. Longitudinal studies have showed that declining BP over time correlates with incident dementia and with imaging and biological markers of neurodegenerative processes. The Kungsholmen Project reported that BP markedly decreased 3 years before a dementia diagnosis and continued to decline thereafter[23]. In the Rotterdam Scan Study, elderly adults without dementia with a decline of more than 10 mmHg in DBP had more cortical atrophy than subjects with stable BP over a 20-year period.24 Longitudinal decrease in MAP was found to be associated with an increase in p-tau181, a cerebrospinal fluid biomarker of Alzheimer's disease, in subjects with hypertension[25].

Why should functional status affect the relationship between BP and cognition? Functional status may be seen as a reflection of the biological age of older adults. Of note, functional status has been shown to affect the association between BP and subsequent mortality risk. In the National Health and Nutrition Examination Survey, functional status was assessed as walking speed for a 20-foot distance in individuals aged 65 and older. High SBP (>140 mmHg) was associated with greater mortality in fast walkers, whereas the association was inversed in those

who did not manage to complete the walking test[26]. Likewise, in the population-based Longitudinal Ageing Study Amsterdam, low DBP was associated with higher all-cause mortality risk in the oldest adults and in participants with a combination of physical and cognitive dysfunction, whereas BP was not related to mortality in more-vital older individuals[27]. Moreover, in the Leiden 85-plus Study, functional status modified the association between higher BP and risk of stroke in the oldest adults[28].

Functional impairment may be a consequence of hypertension, because most of the subjects with low BP late in life had higher BP earlier in life[24]. Functional impairment thus reflects the lifelong atherosclerotic burden of elderly adults. Atherosclerotic damage stiffens brain arteries and impairs brain perfusion regulation. Therefore, subjects with more atherosclerosis are more susceptible to episodic or sustained hypotension because they have a lower critical threshold for cerebral hypoperfusion[22]. In the Kungsholmen Project, the association between SBP decline and increased risk of dementia was observed only in people with baseline SBP less than 160 mmHg or vascular disease. In subjects with vascular disease, there was a dose-response relationship between SBP decline and risk of dementia[23].

Disentangling the relationship between BP and cognition in frail older people has significant clinical implications. Given the increasing life expectancy of populations worldwide[29], dementia is a leading cause of disability[30]. Therefore, a major public health challenge is prevention of dementia through management of its modifiable risk factors. BP is a major target, but optimal BP goals are unclear in individuals aged 80 and older[31] and in frail elderly adults[32]. The Systolic Blood Pressure Intervention Trial (SPRINT), which aims to assess whether individuals aged 75 and older differ from younger individuals in their response to hypertension treatment, specifically addresses this. Moreover, the nested substudy, SPRINT Memory and cognition IN Decreased hypertension (SPRINT-MIND), is designed to evaluate the effect of treatment on age-related decline in cognition and incidence of all-cause dementia[33].

The few previous clinical trials on the prevention of dementia with antihypertensive treatment have provided conflicting results, partly because of short follow-up and the heterogeneity of antihypertensive drugs. The Systolic Hypertension in the Elderly Program[34] and the Medical Research Council[35] trials failed to show any difference in effect on cognition between placebo and active treatment with diuretics or beta-blockers as first-line antihypertensive agents. In contrast, the Systolic Hypertension in Europe[36] showed that antihypertensive therapy starting

with the dihydropyridine calcium channel blocker nitrendipine reduced the incidence of dementia by 55% over a median follow-up of 3.9 years. In the Perindopril Protection Against Recurrent Stroke Study trial[37], combined treatment with perindopril and indapamide reduced stroke-related dementia by 50%. The Hypertension in the Very Elderly Trial (HYVET)[38] failed to show a significant reduction in the incidence of dementia with treatment with indapamide and perindopril. The HYVET data, when combined in a meta-analysis with other placebo-controlled trials of antihypertensive treatment, provided evidence that antihypertensive treatment is beneficial for reducing incidence of dementia in fit elderly adults. Nevertheless, a major weakness of these trials is the inclusion of relatively healthy subjects, which limits the generalizability of results to other populations. A recent community-based study found that only 9% of the oldest adults with hypertension were eligible for inclusion in HYVET.39 As further proof of the selective recruitment of fit elderly adults, the incidence of dementia in the placebo group of the trials was lower than in population-based studies[36,40]. Evidence of the generalizability of the results of clinical trials to the population of elderly outpatients is even more limited.

A strength of this study is that it investigated the connection between BP and cognition in an unselected population of elderly outpatients. To the knowledge of the authors, this is the largest study to be performed in a general geriatric unit. Another strength is that it proves the utility of categorizing elderly adults on the basis not only of chronological age, but also of markers of biological age as ADL and IADL scores. Any trained physician can collect this information. The main limitation of this study is the cross-sectional observational design, which prevents causality relationships from being inferred. Second, the MMSE, a widely used global measure of cognitive function, might have missed variation in executive function, the domain of cognition that hypertension particularly affects. Third, a single BP measurement was used in the analyses. Because BP is highly variable in older adults, participants may have been misclassified, although is it likely that misclassification would have occurred randomly, possibly leading to underestimation of true associations. Nevertheless, the data add further evidence of low BP as a risk factor for frail older adults in an outpatient setting.

In conclusion, higher BP is associated with better cognitive function in older individuals aged over 85 and in those with impaired functional status. The optimal threshold of BP may depend on both chronological and biological age (reflected by functional status). Therefore, BP management in older adults should be personalized, taking into account functional status.

REFERENCES

1. Qiu C, Winblad B, Fratiglioni L. The age-dependent relation of blood pressure to cognitive function and dementia. Lancet Neurol. 2005; 4: 487–499.
2. Launer LJ, Masaki K, Petrovitch H et al. The association between midlife blood pressure levels and late-life cognitive function. The Honolulu-Asia Aging Study. JAMA. 1995;274:1846–1851.
3. Elias MF, Wolf PA, D'Agostino RB et al. Untreated blood pressure level is inversely related to cognitive functioning: The Framingham Study. Am J Epidemiol 1993;138:353–364.
4. Kilander L, Nyman H, Boberg M et al. Hypertension is related to cognitive impairment: A 20-year follow-up of 999 men. Hypertension 1998;31:780–786.
5. Korf ES, White LR, Scheltens P et al. Midlife blood pressure and the risk of hippocampal atrophy: the Honolulu Asia Aging Study. Hypertension. 2004;44:29–34.
6. Obisesan TO, Obisesan OA, Martins S et al. High blood pressure, hypertension, and high pulse pressure are associated with poorer cognitive function in persons aged 60 and older: The Third National Health and Nutrition Examination Survey. J Am Geriatr Soc 2008;56:501–509.
7. Sabayan B, Oleksik AM, Maier AB et al. High blood pressure and resilience to physical and cognitive decline in the oldest old: the Leiden 85-plus Study. J Am Geriatr Soc. 2012 Nov;60(11):2014-9.
8. Richmond R, Law J, Kay-Lambkin F. Higher blood pressure associated with higher cognition and functionality among centenarians in Australia. Am J Hypertens 2011;24:299–303.
9. Guo Z, Fratiglioni L, Winblad B et al. Blood pressure and performance on the mini-mental state examination in the very old: cross-sectional and longitudinal data from the Kungsholmen Project. Am J Epidemiol 1997; 145: 1106–13.
10. Farmer ME, White LR, Abbott RD et al. Blood pressure and cognitive performance: the Framingham Study. Am J Epidemiol 1987; 126: 1103–14.
11. Euser SM, van Bemmel T, Schram MT et al. The effect of age on the association between blood pressure and cognitive function later in life. J Am Geriatr Soc. 2009;57:1232–1237.
12. Muller M, Smulders YM, de Leeuw PW et al. Treatment of hypertension in the oldest old: a critical role for frailty? Hypertension. 2014 Mar;63(3):433-41.

13. Ruitenberg A, den Heijer T, Bakker SL et al. Cerebral hypoperfusion and clinical onset of dementia: the Rotterdam Study. Ann Neurol. 2005 Jun;57(6):789-94.
14. Folstein MF, Folstein SE, McHugh PR. "Mini-mental state". A practical method for grading the cognitive state of patients for the clinician. J Psychiatr Res. 1975; 12(3): 189-198.
15. Katz S, Ford AB, Moskowitz RW et al. Studies of illness in the aged. The index of ADL: a standardized measure of biological and psychosocial function. JAMA. 1963;185:914-919.
16. WHO Collaborating Centre for Drug Statistics Methodology, Guidelines for ATC classification and DDD assignment 2014. Oslo, 2013
17. Lawton MP, Brody EM. Assessment of older people: self-maintaining and instrumental activities of daily living. Gerontologist. 1969 Autumn;9(3):179-86.
18. McKhann G, Drachman D, Folstein M et al. Clinical diagnosis of Alzheimer's disease: report of the NINCDS-ADRDA Work Group under the auspices of Department of Health and Human Services Task Force on Alzheimer's Disease. Neurology. 1984 Jul;34(7):939-44.
19. McKhann GM, Knopman DS, Chertkow H et al. The diagnosis of dementia due to Alzheimer's disease: recommendations from the National Institute on Aging-Alzheimer's Association workgroups on diagnostic guidelines for Alzheimer's disease. Alzheimers Dement. 2011 May;7(3):263-9.
20. Levey AS, Bosch JP, Lewis JB et al. A more accurate method to estimate glomerular filtration rate from serum creatinine: a new prediction equation. Modification of Diet in Renal Disease Study Group. Ann Intern Med. 1999 Mar 16;130(6):461-70.
21. Skoog I, Andreasson LA, Landahl S et al. A population-based study on blood pressure and brain atrophy in 85-year-olds. Hypertension. 1998 Sep;32(3):404-9.
22. de la Torre JC. Pathophysiology of neuronal energy crisis in Alzheimer's disease. Neurodegener Dis. 2008;5(3-4):126-32.
23. Qiu C, von Strauss E, Winblad B et al. Decline in blood pressure over time and risk of dementia: a longitudinal study from the Kungsholmen project. Stroke. 2004 Aug;35(8):1810-5.
24. Heijer Td, Skoog I, Oudkerk M et al. Association between blood pressure levels over time and brain atrophy in the elderly. Neurobiol Aging. 2003 Mar-Apr;24(2):307-13.

25. Glodzik L, Rusinek H, Pirraglia E et al. Blood pressure decrease correlates with tau pathology and memory decline in hypertensive elderly. Neurobiol Aging. 2014 Jan;35(1):64-71.
26. Odden MC, Peralta CA, Haan MN et al. Rethinking the association of high blood pressure with mortality in elderly adults: the impact of frailty. Arch Intern Med. 2012;172:1162–1168.
27. Post Hospers G, Smulders YM, Maier AB et al. Relation between blood pressure and mortality risk in an older population: role of chronological and biological age. J Intern Med. 2014 Jul 8.
28. Sabayan B, van Vliet P, de Ruijter W et al. High blood pressure, physical and cognitive function, and risk of stroke in the oldest old: the Leiden 85-plus Study. Stroke. 2013 Jan;44(1):15-20.
29. World Population Ageing Report: World Population Ageing Report; 2013 (online). Available at: http://www.un.org/en/development/desa/population/publications/ageing/WorldPopulationAgeingReport2013.shtml . Accessed June 30, 2014.
30. World Health Organization and Alzheimer's Disease International. Dementia: a public health priority. 2012 (online). Available at: http://www.who.int/mental_health/publications/dementia_report_2012/en. Accessed June 30, 2014.
31. Aronow WS, Fleg JL, Pepine CJ et al; ACCF Task Force. ACCF/AHA 2011 expert consensus document on hypertension in the elderly: a report of the American College of Cardiology Foundation Task Force on Clinical Expert Consensus Documents. Circulation. 2011;123(21):2434-2506.
32. Mancia G, Fagard R, Narkiewicz K et al. 2013 ESH/ESC guidelines for the management of arterial hypertension: the Task Force for the Management of Arterial Hypertension of the European Society of Hypertension (ESH) and of the European Society of Cardiology (ESC). Eur Heart J. 2013;34:2159–2219.
33. Ambrosius WT, Sink KM, Foy CG et al. The design and rationale of a multicenter clinical trial comparing two strategies for control of systolic blood pressure: the Systolic Blood Pressure Intervention Trial (SPRINT). Clin Trials. 2014 Oct;11(5):532-46.
34. SHEP Cooperative Research Group. Prevention of stroke by antihypertensive drug treatment in older persons with isolated systolic hypertension. Final results of the Systolic Hypertension in the Elderly Program (SHEP). JAMA. 1991 Jun 26;265(24):3255-64.

35. Prince MJ, Bird AS, Blizard RA, Mann AH. Is the cognitive function of older patients affected by antihypertensive treatment? Results from 54 months of the Medical Research Council's trial of hypertension in older adults. BMJ. 1996 Mar 30;312(7034):801-5.
36. Forette F, Seux ML, Staessen JA et al. The prevention of dementia with antihypertensive treatment: new evidence from the Systolic Hypertension in Europe (Syst-Eur) study. Arch Intern Med. 2002 Oct 14;162(18):2046-52.
37. Tzourio C, Anderson C, Chapman N et al. Effects of blood pressure lowering with perindopril and indapamide therapy on dementia and cognitive decline in patients with cerebrovascular disease. Arch Intern Med. 2003 May 12;163(9):1069-75.
38. Peters R, Beckett N, Forette F et al. Incident dementia and blood pressure lowering in the Hypertension in the Very Elderly Trial cognitive function assessment (HYVET-COG): a double-blind, placebo controlled trial. Lancet Neurol. 2008 Aug;7(8):683-9.
39. Jacobs JM, Stessman J, Ein-Mor E et al. Hypertension and 5-year mortality among 85-year-olds: The Jerusalem Longitudinal Study. J Am Med Dir Assoc 2012; 13:759e1–759e6.
40. Andersen K, Launer LJ, Dewey ME et al. Gender differences in the incidence of AD and vascular dementia: The EURODEM Studies. EURODEM Incidence Research Group. Neurology. 1999 Dec 10;53(9):1992-7.

Table 1. Characteristics of study population according to tertile of systolic blood pressure (starts)

Characteristic	Total population	Systolic blood pressure tertile			p-value
	n = 1,540	Low n=431	Middle n=601	High n=508	
Demographic					
Age, years median [IQR]	82 [78, 86]	81 [78, 87]	81 [78, 85.5]	82 [79, 86]	0.222
Females, n (%)	1,075 (69.8)	288 (66.8)	404 (67.2)	383 (75.4)	0.004
Education, years, median [IQR]	6 [5, 12]	7 [5, 12]	6 [5, 11]	6 [5, 11]	0.615
Blood pressure (mmHg)					
Systolic, median [IQR]	145 [130, 160]	130 [120, 130]	140 [140, 150]	170 [160, 175]	<0.001
Diastolic, median [IQR]	80 [80, 90]	80 [70, 80]	80 [80, 90]	90 [80, 95]	<0.001
Pulse, median [IQR]	60 [50, 70]	50 [40, 55]	60 [60, 70]	80 [70, 90]	<0.001
Mean arterial, median [IQR]	103 [97, 110]	93 [87, 97]	103 [100, 107]	113 [110, 120]	<0.001
Cognitive and functional status					
MMSE, median [IQR]	26 [20, 28]	25 [17, 28]	25 [21, 29]	26 [21, 29]	0.003
ADL, median [IQR]	5.5 [4, 6]	5 [3.5, 6]	5.5 [4.5, 6]	5.5 [4.5, 6]	<0.001
IADL, median [IQR]	5 [2, 8]	4 [1, 7]	5 [3, 8]	5 [3, 8]	<0.001
Cardiovascular risk factors					
Ever smoker, n (%)	551 (35.8)	165 (38.3)	210 (34.9)	176 (34.6)	0.440
Hypertension, n (%)	1095 (71.1)	266 (61.7)	429 (71.4)	400 (78.7)	<0.001
Co-morbidities					
Diabetes mellitus, n (%)	180 (11.7)	37 (8.6)	76 (12.6)	67 (13.2)	0.059
Atrial fibrillation, n (%)	218 (14.2)	67 (15.5)	82 (13.6)	69 (13.6)	0.621
Coronary heart disease, n (%)	361 (23.4)	103 (23.9)	144 (24.0)	114 (22.4)	0.809
Claudication, n (%)	94 (6.1)	27 (6.3)	35 (5.8)	32 (6.3)	0.934
Depression/anxiety, n (%)	762 (49.5)	218 (50.6)	293 (48.8)	251 (49.4)	0.845
Stroke/TIA, n (%)	258 (16.8)	79 (18.3)	96 (16.0)	83 (16.3)	0.579
Cancer, n (%)	136 (8.8)	48 (11.1)	51 (8.5)	37 (7.3)	0.108
Alcohol abuse, n (%)	62 (4.0)	14 (3.2)	28 (4.7)	20 (3.9)	0.520
Alzheimer's dementia, n (%)	389 (25.3)	118 (27.4)	158 (26.3)	113 (22.2)	0.149
Parkinson's disease, n (%)	22 (1.4)	12 (2.8)	2 (0.3)	8 (1.6)	0.004
Other neurod., n (%)	15 (1.0)	6 (1.4)	6 (1.0)	3 (0.6)	0.459
GFR, mL/min, median [IQR]	64.9 [55.6, 84.5]	66.0 [55.5, 85.4]	64.8 [55.4, 84.0]	65.2 [55.9, 84.1]	0.765

Table 1. Characteristics of study population according to tertile of systolic blood pressure (continues)

Characteristic	Total population	Systolic blood pressure tertile			p-value
	n = 1,540	Low n=431	Middle n=601	High n=508	
Medications					
On antihypertensives, n (%)	993 (64.5)	248 (57.5)	391 (65.1)	354 (69.7)	0.001
Anti-adrenergics, n (%)	58 (3.8)	9 (2.1)	18 (3.0)	31 (6.1)	0.002
Diuretics, n (%)	344 (22.3)	104 (24.1)	141 (23.5)	99 (19.5)	0.164
Beta-block., n (%)	139 (9.0)	37 (8.6)	49 (8.2)	53 (10.4)	0.390
Calcium-channel block., n (%)	370 (24.0)	88 (20.4)	155 (25.8)	127 (25.0)	0.113
ACE-inhibitors/AA, n (%)	566 (36.8)	129 (29.9)	217 (36.1)	220 (43.3)	<0.001
Antipsychotics, n (%)	136 (8.8)	56 (13.0)	52 (8.7)	28 (5.5)	<0.001
Anxiolytics, n (%)	404 (26.2)	103 (23.9)	164 (27.3)	137 (27.0)	0.427
Hypnotics/sedatives, n (%)	116 (7.5)	39 (9.0)	40 (6.7)	37 (7.3)	0.344
Antidepressants, n (%)	197 (12.8)	60 (13.9)	72 (12.0)	65 (12.8)	0.655
N of medications, median [IQR]	3 [2, 5]	3 [2, 5]	3 [2, 5]	3.5 [2, 5]	0.219

Abbreviations: n = number; IQR = inter quartile range; mmHg: millimeter of mercury; MMSE = Mini Mental State Examination; ADL = Activities of Daily Living; IADL = Instrumental Activities of Daily Living; TIA = transient ischemic attack; neurod = neurodegenerative; GFR= glomerular filtration rate; mL/min = millilitre/minute; block = blockers; ACE = angiotensin converting enzyme inhibitors; AA = angiotensin II antagonists.

Table 2. MMSE score according to tertile of blood pressure

	Tertiles			
	Low	**Middle**	**High**	**p-value**
SBP				
n	431	601	508	
Range of SBP (mmHg)	85 - 135	140 - 150	155 - 260	
Mean SBP (SD) (mmHg)	124.3 (8.4)	144.3 (4.8)	169.5 (12.9)	
MMSE score, mean (SE)	22.2 (0.3)	23.6 (0.3)*	24.2 (0.3)*	**<0.001**
DBP				
n	307	737	496	
Range of DBP (mmHg)	45 - 75	80 - 85	90 - 130	
Mean DBP (SD) (mmHg)	69.1 (4.6)	80.7 (1.7)	93.7 (5.8)	
MMSE score, mean (SE)	21.6 (0.4)	23.6 (0.2)*	24.1 (0.3)*	**<0.001**
PP				
n	451	465	624	
Range of PP (mmHg)	20 - 55	60 - 65	70 - 130	
Mean PP (SD) (mmHg)	46.9 (6.2)	60.8 (1.8)	79.9 (11.0)	
MMSE score, mean (SE)	22.9 (0.3)	23.1 (0.3)	23.9 (0.3)*	**0.013**
MAP				
n	476	544	520	
Range of MAP (mmHg)	66.7 -98.3	100 – 108.3	110 – 173.3	
Mean MAP (SD) (mmHg)	91.3 (5.7)	102.9 (2.9)	116.9 (7.6)	
MMSE score, mean (SE)	22.3 (0.3)	23.6 (0.3)*	24.2 (0.3)*	**<0.001**

Abbreviations: mmHg: millimeter of mercury; MMSE: Mini Mental State Examination; SD: standard deviation; SE: standard error; SBP: systolic blood pressure; DBP: diastolic blood pressure; PP: pulse pressure; MAP: mean arterial pressure. MMSE scores are presented as unadjusted means (Standard Error). P-values are computed using blood pressure measures as continuous variables and are derived from the unadjusted model. *p-value<0.05 for difference between the low tertile and the middle/high tertile.

Table 3. Associations between blood pressure measures and Mini Mental State Examination score, stratified by age (starts)

	75 – 79 years (n=534)		80 – 84 years (n=497)		85+ years (n=509)		p-value for interaction
	β-coefficient (95% CI)	p-value	β-coefficient (95% CI)	p-value	β-coefficient (95% CI)	p-value	
SBP							
Model 1[a]	0.17 [-0.11; 0.44]	0.235	0.27 [-0.02; 0.56]	0.065	0.73 [0.45; 1.01]	**<0.001**	**0.001**
Model 2[b]	0.29 [0.04; 0.54]	**0.025**	0.23 [-0.06; 0.51]	0.114	0.64 [0.38; 0.90]	**<0.001**	**0.005**
Model 3[c]	0.19 [-0.04; 0.42]	0.104	0.25 [0.01; 0.49]	**0.045**	0.44 [0.22; 0.66]	**<0.001**	**0.006**
Model 4[d]	0.18 [-0.06; 0.43]	0.146	0.24 [-0.03; 0.51]	0.080	0.33 [0.10; 0.56]	**0.005**	0.146
DBP							
Model 1[a]	0.53 [-0.02; 1.09]	0.060	0.37 [-0.19; 0.94]	0.195	1.57 [0.96; 2.18]	**<0.001**	**<0.001**
Model 2[b]	0.57 [0.05; 1.08]	**0.030**	0.23 [-0.32; 0.78]	0.407	1.27 [0.71; 1.83]	**<0.001**	**0.001**
Model 3[c]	0.40 [-0.06; 0.85]	0.087	0.36 [-0.12; 0.83]	0.138	1.00 [0.53; 1.48]	**<0.001**	**0.002**
Model 4[d]	0.30 [-0.20; 0.80]	0.244	0.32 [-0.20; 0.83]	0.230	0.91 [0.42; 1.40]	**<0.001**	**0.041**

Table 3. Associations between blood pressure measures and Mini Mental State Examination score, stratified by age (continues)

PP							
Model 1[a]	0.06 [-0.29; 0.41]	0.740	0.27 [-0.09; 0.64]	0.140	0.63 [0.28; 0.98]	**<0.001**	**0.036**
Model 2[b]	0.25 [-0.08; 0.57]	0.137	0.26 [-0.09; 0.61]	0.150	0.57 [0.25; 0.89]	**0.001**	0.069
Model 3[c]	0.14 [-0.15; 0.43]	0.333	0.24 [-0.07; 0.54]	0.126	0.34 [0.07; 0.61]	**0.013**	0.061
Model 4[d]	0.18 [-0.14; 0.48]	0.268	0.23 [-0.10; 0.57]	0.170	0.20 [-0.09; 0.49]	0.172	0.438
MAP							
Model 1[a]	0.40 [-0.06; 0.85]	0.091	0.43 [-0.05; 0.90]	0.081	1.41 [0.92; 1.89]	**<0.001**	**<0.001**
Model 2[b]	0.53 [0.10; 0.95]	**0.015**	0.32 [-0.15; 0.78]	0.181	1.19 [0.74; 1.64]	**<0.001**	**0.001**
Model 3[c]	0.36 [-0.02; 0.74]	0.064	0.40 [-0.00; 0.80]	0.052	0.88 [0.50; 1.26]	**<0.001**	**0.001**
Model 4[d]	0.31 [-0.11; 0.72]	0.146	0.37 [-0.07; 0.82]	0.099	0.73 [0.33; 1.13]	**<0.001**	0.053

Abbreviations: CI: confidence interval; SBP: systolic blood pressure; DBP: diastolic blood pressure; PP: pulse pressure; MAP: mean arterial pressure. Beta-coefficients represent change in Mini Mental State Examination score for each 10 mmHg increase in blood pressure measures. [a] Model 1: unadjusted. [b] Model 2: adjusted for age, sex and education. [c] Model 3: adjusted for age, sex, education, smoke, hypertension, diabetes mellitus, atrial fibrillation, coronary heart disease, claudication, depression/anxiety, history of transient ischemic attack or stroke, Alzheimer's disease, present cancer, alcohol abuse, Parkinson's disease, other neurodegenerative conditions, number of medication, anti-adrenergics, diuretics, beta-blockers, calcium-channel blockers, angiotensin converting enzyme inhibitors/angiotensin II antagonists, antipsychotics, anxiolytics, sleep-inducers and sedatives, antidepressants. [d] Model 4: Model 3 plus glomerular filtration rate.

Figure 1. MMSE score in tertiles of blood pressure stratified for ADL and IADL

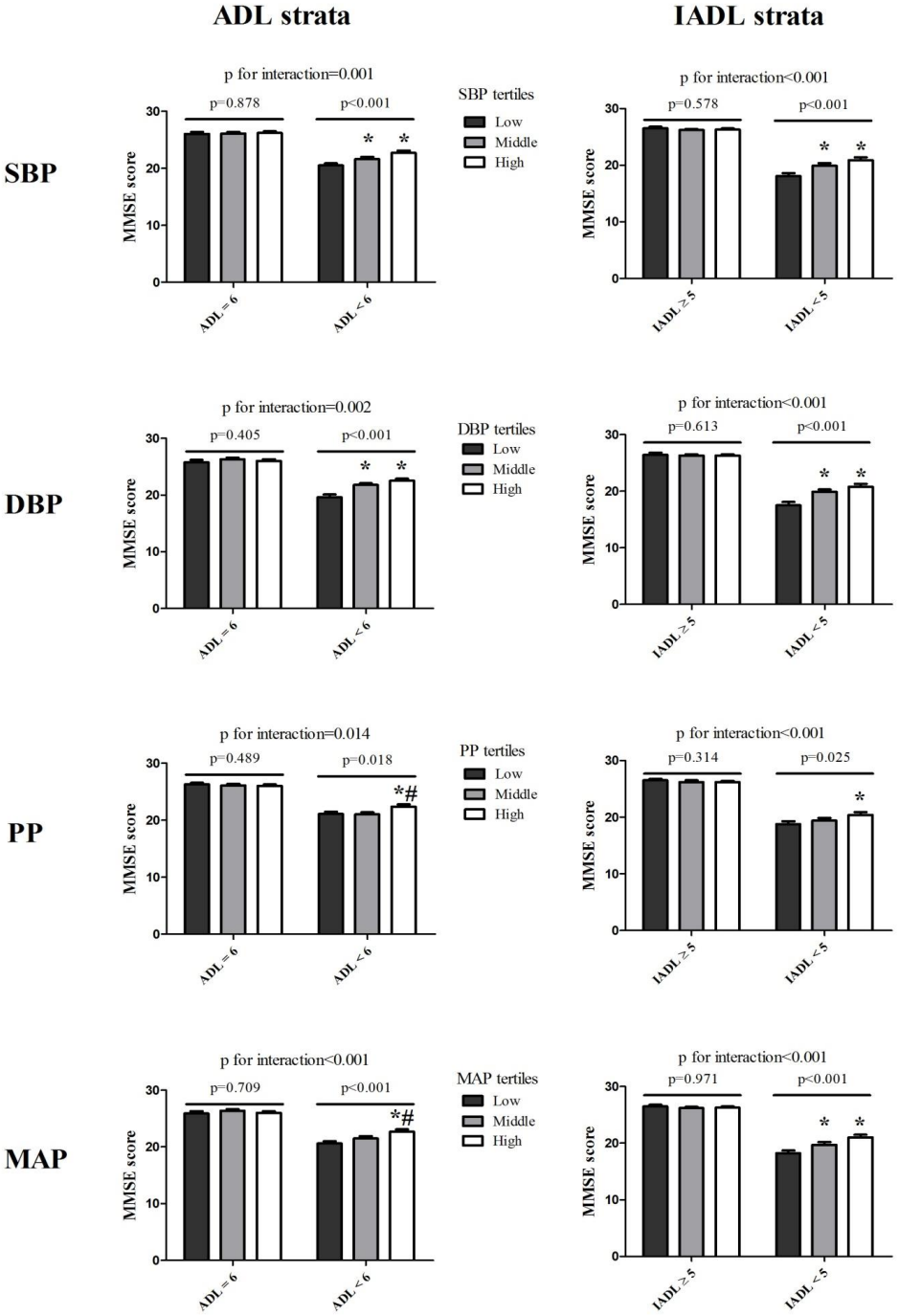


Figure 1. Bars represent unadjusted MMSE score means (with standard error). Abbreviations: MMSE = Mini Mental State Examination; ADL = Activities of Daily Living; IADL = Instrumental Activities of Daily Living; SBP = systolic blood pressure, DBP = diastolic blood pressure, PP = pulse pressure, MAP = mean arterial pressure. P-values for interaction indicate the interaction between blood pressure measures and ADL/IADL (interaction terms were calculated by multiplying continuous blood pressure measures by continuous ADL/IADL scores). The other p-values indicate the trend (linear regression). The symbol * indicates a significant difference between the low tertile and the middle/high tertile. The symbol # indicates a significant difference between the middle and high tertile. P-values for interaction, p-values for trend and differences between tertiles are computed in the unadjusted model.

Chapter 3

Blood pressure and mortality

Manuscript based on this chapter has been published as:

Ogliari G, Westendorp RG, Muller M, Mari D, Torresani E, Felicetta I, Lucchi T, Rossi PD, Sabayan B, de Craen AJ. Blood pressure and 10-year mortality risk in the Milan Geriatrics 75+ Cohort Study: role of functional and cognitive status. Age Ageing. 2015;44(6):932-7.

ABSTRACT

Background: optimal blood pressure targets in older adults are controversial.

Objective: to investigate whether the relation of blood pressure with mortality in older adults varies by age, functional and cognitive status.

Design: longitudinal geriatric outpatient cohort.

Setting: Milan Geriatrics 75+ Cohort Study.

Subjects: one thousand five hundred and eighty-seven outpatients aged 75 years and over.

Methods: the relations of systolic (SBP) and diastolic blood pressure (DBP) with mortality risk were analysed using Cox proportional hazards models. Blood pressure, Mini-Mental State Examination (MMSE) and Basic Activities of Daily Living (ADL) were assessed at baseline. All analyses were adjusted for socio-demographic factors, co-morbidities and medications.

Results: one thousand and forty-six patients died during 10-year follow-up. The relationships of SBP and DBP with mortality risk were U-shaped; SBP of 165 mmHg and DBP of 85 mmHg were associated with the lowest mortality. Patients with SBP < 120 mmHg and patients with SBP 120–139 mmHg had 1.64-fold (95% confidence intervals, CI 1.21–2.23) and 1.32-fold (95% CI 1.10–1.60) higher mortality risk than patients with SBP 160–179 mmHg (P values 0.001 and 0.004, respectively). In patients with SBP below 180 mmHg, higher SBP was associated with lower mortality in patients with impaired ADL and MMSE but not in those with preserved ADL and/or MMSE (P for interaction 0.033). Age did not modify the correlation of SBP with mortality.

Conclusions: the correlations of SBP and DBP with mortality were U-shaped. Higher SBP is related to lower mortality in subjects with impaired ADL and MMSE. ADL and MMSE may identify older subjects who benefit from higher blood pressure.

INTRODUCTION

The prevalence of hypertension increases with age, mainly due to rising systolic blood pressure (SBP)[1]. Hypertension is a leading risk factor for mortality, and its detection and control is a public health priority[2]. However, optimal treatment goals in frail older adults remain controversial[3–5].

The relation between blood pressure (BP) and mortality becomes complex in older adults. While in middle age higher BP is strongly and consistently associated with increased mortality risk, this association attenuates or even reverses when ageing[6]. A few studies showed a paradoxical increase in mortality with decreasing BP, thus suggesting a U-shaped relationship[7,8]. In addition, population-based studies indicated that chronological and biological age may affect the relation between BP and mortality, with lower BP being associated with increased mortality in the oldest and in the frailest[9–14]. An increased mortality risk in older adults with low BP may be a short-term phenomenon, attributable to co-morbidity and/or low BP in proximity of death, though reports are conflicting[11, 12]. Moreover, trials on the effect of antihypertensive drugs on mortality are conflicting; a meta-analysis showed no association between antihypertensives and overall mortality in adults ≥80 years[15]. Trials evidence on frailty impact is scarce[5].

Furthermore, the generalisability of data from trials or population-based studies to geriatric patients is debatable. Clinicians are confronted with a lack of data in outpatient populations, in which co-morbidities, functional and cognitive impairment may be more prevalent and severe and their interaction with BP and mortality more complex than in the general population.

In the Milan Geriatrics 75+ Cohort Study, we recruited older outpatients with a wide range of functional and cognitive status and prospectively followed them for 10 years. In this study, we examined whether baseline BP was associated with all-cause mortality and whether this association varied by chronological age and levels of functional and cognitive impairment.

METHODS

Study Design and Participants

The Milan Geriatrics 75+ Cohort Study is a prospective, hospital-based cohort study of the outpatients of the Geriatric Unit of 'I.R.C.C.S. Ca' Granda' in Milan, Italy. Between 3 January 2000 and 25 March 2004, 1,861 new consecutive outpatients aged ≥75 years attended a first comprehensive visit. Details of study design have been previously described[16]. After informed consent, the participants underwent a face-to-face standardised, structured, extensive medical assessment with trained physicians. After excluding participants with missing data on baseline BP (n = 200), other baseline covariates (n = 32) or mortality (n = 42), we included 1,587 participants in this study. I.R.C.C.S. Ca' Granda Ethics Committee approved the study.

Blood pressure

Physicians measured baseline arterial BP with a mercury sphygmomanometer, at heart level, in the seated position, after 5 min of rest and no vigorous exercise in the preceding 30 min. SBP and diastolic BP (DBP) were manually auscultated. Mean arterial pressure (MAP) was calculated as 1/3(SBP) + 2/3 (DBP) and pulse pressure (PP) as (SBP) − (DBP)[6].

Cognitive and functional status

Cognitive function was assessed using the 30-item Mini-Mental State Examination (MMSE)[17]. Functional status was evaluated using Katz' Activities of Daily Living (ADL) questionnaire[18]. ADL includes six items (rising or lying down, feeding, dressing, bathing, toileting, urinary and faecal continence). We defined impaired ADL as ADL score ≤ 5 and impaired MMSE as MMSE score ≤ 24 [17, 18].

Co-morbidities and life-style factors

Hypertension was defined by a previous diagnosis. Coronary heart disease (CHD), history of transient ischaemic attack (TIA) or stroke, diabetes mellitus, atrial fibrillation, claudication and heart failure were proved by medical documents. Cardiovascular disease was defined by the presence of CHD, TIA/stroke or claudication. Cancer was defined by a diagnosis within the previous 5 years. Symptoms of anxiety/depression were self-reported or stated in medical documents. Smoking was dichotomised as never or ever (current and previous). Education was defined as years of school attended.

Medications

The number of medications was the number of drugs taken chronically or cyclically. Antihypertensives were defined by Anatomical Therapeutic Chemical classification codes C02 (alpha-anti-adrenergics), C03 (diuretics), C07 (β-blockers), C08 (calcium-channel blockers) and C09 (angiotensinconverting enzyme (ACE) inhibitors/angiotensin II antagonists)[19].

Mortality

All-cause mortality was assessed by collecting data from the Register Office of Milan or other town of residence. The follow-up period was the time between baseline and either death, loss to follow-up or 10-year period.

Statistical analyses

We performed Cox regression to estimate hazard ratios (HRs) and 95% confidence intervals (CI) for the association between BP measures and mortality.

In all cohort, to test the presence of a non-linear association between BP measures and mortality, we entered BP measures and squared BP measures in the Cox regression as continuous variables. To gain clinical insight into these correlations, we run further analyses using BP measures as categorical variables. We classified participants into five SBP clinical

categories (<120, 120–139, 140–159, 160–179 and ≥180 mmHg), four DBP categories (<80, 80–89, 90–99 and ≥100 mmHg) and quintiles of MAP or PP. The categories associated with the lowest mortality risk were set as references.

In participants with SBP below 180 mmHg (without hypertensive crisis)[3], we tested the presence of a linear association between SBP and mortality. In this group, we explored the influence of chronological and biological age on the relationship between SBP and mortality. To evaluate the effect of chronological age, we performed Cox regression after categorising participants in three age strata (75–79, 80–84 and 85+ years). Furthermore, to assess the role of biological age as reflected in the functional/cognitive status, we repeated the analyses after stratifying participants according to: (i) ADL impairment (ADL score ≤ 5), (ii) MMSE impairment (MMSE score ≤ 24) or (iii) a combination of ADL/MMSE impairment (both impaired, either impaired, preserved)[17, 18]. We also carried out the analyses after stratifying participants by history of hypertension or cardiovascular disease. We tested for interaction by computing interaction terms using SBP as a continuous measure. Sensitivity analyses explored the influence of impaired cardiac function and imminent death, respectively, by excluding: (i) participants with heart failure at baseline or (ii) participants who died or were lost to follow-up in the first year. All analyses were also done for DBP.

All analyses were performed in three steps. In Model 1, analyses were adjusted for age and sex. In Model 2, they were additionally adjusted for education, smoke, hypertension, diabetes mellitus, atrial fibrillation, CHD, claudication, TIA/stroke, depression/anxiety, cancer and number of medications. In Model 3, they were further adjusted for the use of antihypertensives.

Analyses were performed using SPSS version 20.0.0 (SPSS Inc., Chicago, IL, USA).

RESULTS

Table 1 shows baseline characteristics of the study population. One thousand one hundred and fourteen (70.2%) participants were female, mean age was 82 years (range 75–101) and median SBP and DBP were 145 and 80 mmHg, respectively. One thousand and seventeen (64.1%) participants had a history of hypertension. Participants with higher SBP were more likely to be

females, to have hypertension or to be treated with alpha-anti-adrenergics or ACE inhibitors/angiotensin II antagonists, while less likely to have CHD (all $P < 0.05$) (Supplementary Table 1). Similar associations with clinical characteristics were observed for DBP (Supplementary Table 2).

After 10-year follow-up, 1,046 (65.9%) participants had died. The relationships of SBP, DBP, MAP and PP with mortality risk, respectively, were U-shaped (all $P < 0.05$). Mortality risk was lowest at SBP of 165 mmHg and at DBP of 85 mmHg. Figure 1 shows mortality risk in SBP and DBP categories. Participants with SBP < 120 mmHg and participants with SBP 120–139 mmHg had a 1.64-fold (95% CI 1.21–2.23) and a 1.32-fold (95% CI 1.10–1.60) increased mortality risk than participants with SBP 160–179 mmHg (P values 0.001 and 0.004, respectively). Participants with DBP ≥ 100 mmHg had a 1.44-fold (95% CI 1.12–1.86) increased mortality risk than participants with DBP 90–99 mmHg (P value 0.004). Mortality risk in MAP and PP quintiles is illustrated in Supplementary Figure 1.

When focusing on participants with SBP below 180 mmHg (n = 1,451), we observed an inverse linear relationship between SBP and mortality (Supplementary Table 3). In this group, each 10 mmHg higher SBP was associated with a 0.87-fold (95% CI 0.77–0.99), a 0.91-fold (95% CI 0.86–0.96) and a 0.92-fold (95% CI 0.88–0.96) decreased risk of mortality at 1-year, 5-year and 10-year follow-up, respectively (all $P < 0.05$). Age, history of hypertension or cardiovascular disease did not modify the association between SBP and mortality (P values for interaction = 0.653, 0.609 and 0.545, respectively). After stratifying for functional status, higher SBP was related to a decreased mortality risk in participants with impaired ADL, while not in those with preserved ADL (P values 0.001 and 0.085, respectively; P value for interaction = 0.093). Likewise, the association between higher SBP and decreased mortality risk was consistent in participants with impaired MMSE but not in those with preserved MMSE (P values 0.001 and 0.070, respectively; P value for interaction = 0.100). The relationship of SBP with mortality remained significant after exclusion of: (i) participants with heart failure at baseline (n = 145) or (ii) participants who died or were lost to follow-up in the first year (n = 171). In participants with SBP below 180 mmHg, no association was observed between DBP and mortality (Supplementary Table 4).

Figure 2 illustrates the relationship of SBP and DBP with 10-year mortality after stratifying participants for both functional and cognitive status. In participants with impairment in both

ADL and MMSE, each 10-mmHg rise in SBP was related to a 0.89-fold (95% CI 0.83–0.96) decreased mortality risk. Conversely, no association was observed in participants with impairment in either ADL or MMSE (HR 0.94, CI 0.87–1.02) or in participants with preserved ADL and MMSE (HR 0.95, CI 0.87–1.03). Functional and cognitive status significantly modified the relationship of SBP with mortality (P for interaction = 0.033). Likewise, higher DBP tended to be more related with decreased mortality risk in participants with impaired ADL and/or MMSE and with increased mortality risk in those with preserved ADL and MMSE.

DISCUSSION

In this large longitudinal geriatric outpatient cohort, we reported two major findings. First, the association between BP and mortality risk was U-shaped with lowest mortality at SBP of 165 mmHg. Second, the association between SBP and mortality varied by functional and cognitive status; higher SBP was correlated with increased survival especially in participants with impairment. These correlations were independent of cardiovascular risk factors, co-morbidities and medications.

Our report of a mortality nadir at SBP of 165 mmHg is in line with results from a population-based cohort of adults aged ≥ 85 years[6]. In our cohort, participants with SBP values in the optimal or normal range (<140 mmHg) presented increased mortality risk compared with those with SBP 160–179 mmHg.

Our novel finding is that the association between SBP and mortality varies by biological age as defined by functional and cognitive status in geriatric outpatients. To our knowledge, the influence of biological age has been explored only in few population-based studies[13, 14, 20]. In the National Health and Nutrition Examination Survey, the association of BP with mortality varied by walking speed; elevated SBP was associated with a greater mortality risk among faster walkers while the association was reversed in the frailest participants who did not manage to complete the walking test[13]. In the Longitudinal Ageing Study Amsterdam, higher DBP was related to lower mortality risk in participants with a combination of physical and cognitive dysfunction (low walking speed and/or low MMSE score)[14]. In the Cardiovascular Health Study, DBP ≤ 65 mmHg correlated with the highest mortality risk in people with ADL

impairment, whereas the association reversed in those without ADL impairment[20]. We did not observe similar associations, possibly due to a DBP distribution towards higher values in our cohort.

Different explanations can be proposed. First, both low SBP and higher mortality risk could result from a common underlying cause, such as imminent death or impaired cardiac function[21]. Nonetheless, our estimates did not change after exclusion of participants who died in the first year of follow-up or those with heart failure.

A second explanation is that hypoperfusion of vital organs such as the heart and the brain may link lower BP to increased mortality risk. Episodic hypotension has been associated with compromised coronary perfusion[8] and markers of brain damage[22, 23]. The ageing brain may be more vulnerable to hypoperfusion during episodes of hypotension[24]. Conversely, higher SBP has been related to lower risk of stroke in adults over 85 years with physical and or cognitive impairment[25]. Consistently, in our cohort, the association of lower SBP and higher mortality was consistent in participants with impairment.

Given the increasing life expectancy, it is crucial to explore whether the relationship of BP and mortality varies among heterogeneous old populations. Controversy persists on BP targets in adults over 80 years and in frail older adults[3, 4]. Findings from clinical trials are mixed. In the Hypertension in the Very Elderly Trial (HYVET), antihypertensive treatment, compared with placebo, reduced mortality in adults over 80 years with SBP $\geq$ 160 mmHg[26]. In HYVET, participants were treated with indapamide with or without perindopril to achieve the target BP of 150/80. Similarly, in STOP-Hypertension, therapy with β-blockers and a diuretic reduced mortality in adults aged 70–84 years, compared with placebo[27]. In contrast, in Syst-Eur, treatment starting with nitrendipine had no effect on mortality in adults over 60 years[28]. In Systolic Hypertension in the Elderly Program, treatment of isolated systolic hypertension with chlorthalidone was not associated with survival[29].

Furthermore, concern is growing on the generalisability of clinical trials. In HYVET, frailty did not modify treatment effects, but the definition of frailty did not consider the severity of impairments and dementia was an exclusion criterion[5]. The exclusion of people with severe co-morbidities or impairment from trials is proved by low mortality rates[29]. In a population-based study, only 9% of the oldest with hypertension were eligible for inclusion in HYVET[30].

As the relationship of BP with mortality differs between fit and frail older adults, as suggested by our and other reports, the outcomes of the trials so far are difficult to generalise.

The novelty of our study is to investigate the relationship between BP and mortality in an outpatient hospital-based cohort. A further asset is to show that ADL and MMSE— simple, common questionnaires—can identify adults who may benefit from higher BP. The observational design limits us in inferring causality.

In conclusion, higher SBP is correlated with decreased mortality risk in adults with functional and cognitive impairment. BP management in older adults should be personalised using functional and cognitive status as markers of biological age.

ACKNOWLEDGEMENTS

We thank Milan Register Office ('Ufficio Anagrafe').

REFERENCES

1. Lloyd-Jones DM, Evans JC, Levy D. Hypertension in adults across the age spectrum: current outcomes and control in the community. JAMA. 2005;294:466-72.
2. Lim SS, Vos T, Flaxman AD, et al. A comparative risk assessment of burden of disease and injury attributable to 67 risk factors and risk factor clusters in 21 regions, 1990-2010: a systematic analysis for the Global Burden of Disease Study 2010. Lancet. 2012;380:2224-60.
3. Mancia G, Fagard R, Narkiewicz K, et al. 2013 ESH/ESC guidelines for the management of arterial hypertension: the Task Force for the Management of Arterial Hypertension of the European Society of Hypertension (ESH) and of the European Society of Cardiology (ESC). Eur Heart J. 2013;34:2159–2219.
4. Aronow WS, Fleg JL, Pepine CJ, et al; ACCF Task Force. ACCF/AHA 2011 expert consensus document on hypertension in the elderly: a report of the American College of Cardiology Foundation Task Force on Clinical Expert Consensus Documents. Circulation. 2011;123:2434-2506.

5. Warwick J, Falaschetti E, Rockwood K, et al. No evidence that frailty modifies the positive impact of antihypertensive treatment in very elderly people: an investigation of the impact of frailty upon treatment effect in the HYpertension in the Very Elderly Trial (HYVET) study, a double-blind, placebo-controlled study of antihypertensives in people with hypertension aged 80 and over. BMC Med. 2015;13:78.
6. Lewington S, Clarke R, Qizilbash N, Peto R, Collins R; Prospective Studies Collaboration. Age-specific relevance of usual blood pressure to vascular mortality: a meta-analysis of individual data for one million adults in 61 prospective studies. Lancet. 2002;360:1903-13.
7. Molander L, Lövheim H, Norman T, Nordström P, Gustafson Y. Lower systolic blood pressure is associated with greater mortality in people aged 85 and older. J Am Geriatr Soc. 2008;56(10):1853-9.
8. Messerli FH, Mancia G, Conti CR, et al. Dogma disputed: can aggressively lowering blood pressure in hypertensive patients with coronary artery disease be dangerous? Ann Intern Med. 2006;144:884-93.
9. Satish S, Freeman DH Jr, Ray L, Goodwin JS. The relationship between blood pressure and mortality in the oldest old. J Am Geriatr Soc. 2001;49:367–374.
10. van Bemmel T, Gussekloo J, Westendorp RG, Blauw GJ. In a population based prospective study, no association between high blood pressure and mortality after age 85 years. J Hypertens. 2006;24:287–292.
11. Boshuizen HC, Izaks GJ, van Buuren S, Ligthart GJ. Blood pressure and mortality in elderly people aged 85 and older: community based study. BMJ. 1998;316:1780-4.
12. Poortvliet RK, Blom JW, de Craen AJ, et al. Low blood pressure predicts increased mortality in very old age even without heart failure: the Leiden 85-plus Study. Eur J Heart Fail. 2013;15:528-33.
13. Odden MC, Peralta CA, Haan MN, Covinsky KE. Rethinking the association of high blood pressure with mortality in elderly adults: the impact of frailty. Arch Intern Med. 2012;172:1162-8.
14. Post Hospers G, Smulders YM, Maier AB, Deeg DJ, Muller M. Relation between blood pressure and mortality risk in an older population: role of chronological and biological age. J Intern Med. 2015 Apr;277:488-97.
15. Bejan-Angoulvant T, Saadatian-Elahi M, Wright JM, et al. Treatment of hypertension in patients 80 years and older: the lower the better? A meta-analysis of randomized controlled trials. J Hypertens. 2010;28:1366-72.

16. Ogliari G, Sabayan B, Mari D, et al. Age- and functional status-dependent association of blood pressure with cognition: the Milan Geriatrics 75+ Cohort Study. J Am Geriatr Soc 2015;63: 1741–8.
17. Folstein MF, Folstein SE, McHugh PR. "Mini-mental state". A practical method for grading the cognitive state of patients for the clinician. J Psychiatr Res. 1975;12:189-198.
18. Katz S, Ford AB, Moskowitz RW, Jackson BA, Jaffe MW. Studies of illness in the aged. The index of ADL: a standardized measure of biological and psychosocial function. JAMA. 1963;185:914-919.
19. WHO Collaborating Centre for Drug Statistics Methodology, Guidelines for ATC classification and DDD assignment 2014. Oslo, 2013.
20. Peralta CA, Katz R, Newman AB, Psaty BM, Odden MC. Systolic and diastolic blood pressure, incident cardiovascular events, and death in elderly persons: the role of functional limitation in the Cardiovascular Health Study. Hypertension. 2014;64:472-80.
21. van Bemmel T, Holman ER, Gussekloo J, Blauw GJ, Bax JJ, Westendorp RG. Low blood pressure in the very old, a consequence of imminent heart failure: the Leiden 85-plus Study. J Hum Hypertens. 2009;23:27-32.
22. Skoog I, Andreasson LA, Landahl S, Lernfelt B. A population-based study on blood pressure and brain atrophy in 85-year-olds. Hypertension. 1998;32:404-9.
23. Olesen PJ, Guo X, Gustafson D, et al. A population-based study on the influence of brain atrophy on 20-year survival after age 85. Neurology. 2011;76:879-86.
24. de la Torre JC. Pathophysiology of neuronal energy crisis in Alzheimer's disease. Neurodegener Dis. 2008;5:126-32.
25. Sabayan B, van Vliet P, de Ruijter W, Gussekloo J, de Craen AJ, Westendorp RG. High blood pressure, physical and cognitive function, and risk of stroke in the oldest old: the Leiden 85-plus Study. Stroke. 2013;44:15-20.
26. Beckett NS, Peters R, Fletcher AE, et al; HYVET Study Group. Treatment of hypertension in patients 80 years of age or older. N Engl J Med. 2008;358:1887-98.
27. Dahlöf B, Lindholm LH, Hansson L, Scherstén B, Ekbom T, Wester PO. Morbidity and mortality in the Swedish Trial in Old Patients with Hypertension (STOP-Hypertension). Lancet. 1991;338:1281-5.
28. Staessen JA, Fagard R, Thijs L, et al. Randomised double-blind comparison of placebo and active treatment for older patients with isolated systolic hypertension. The Systolic Hypertension in Europe (Syst-Eur) Trial Investigators. Lancet. 1997;350:757-64.

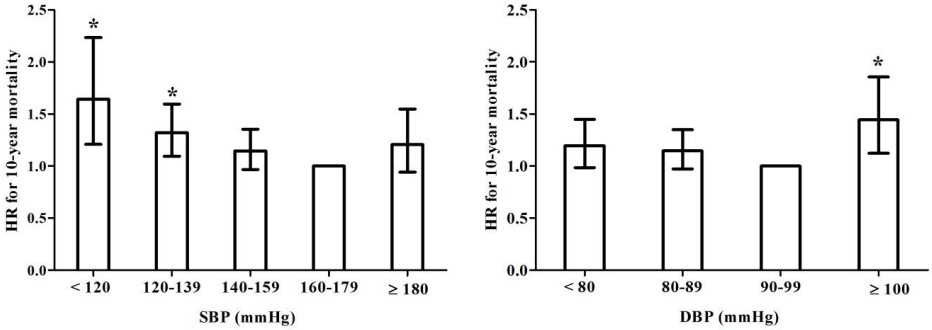
29. Kostis JB, Cabrera J, Cheng JQ, et al. Association between chlorthalidone treatment of systolic hypertension and long-term survival. JAMA. 2011;306:2588-93.
30. Jacobs JM, Stessman J, Ein-Mor E, Bursztyn M. Hypertension and 5-year mortality among 85-year-olds: the Jerusalem Longitudinal Study. J Am Med Dir Assoc. 2012;13:759.e1-6.

Table 1. Baseline characteristics of study population

Characteristic	All cohort
	n = 1,587
Demographics	
Age, years median [IQR]	82 [78; 86]
Females, n (%)	1114 (70.2)
Education, years, median [IQR]	7 [5; 11]
Blood pressure	
SBP, mmHg, median [IQR]	145 [130; 160]
DBP, mmHg, median [IQR]	80 [80; 90]
Cardiovascular risk factors	
Ever smoker, n (%)	566 (35.7)
Hypertension, n (%)	1017 (64.1)
Functional/cognitive status	
ADL score, median [IQR]	5.5 [4.5; 6]
MMSE score, median [IQR]	25 [20; 29]
Co-morbidities	
Diabetes mellitus, n (%)	194 (12.2)
Atrial fibrillation, n (%)	235 (14.8)
Coronary heart disease, n (%)	393 (24.8)
Claudication, n (%)	101 (6.4)
Depression/anxiety, n (%)	772 (48.6)
Stroke/TIA, n (%)	267 (16.8)
Cancer, n (%)	141 (8.9)
Heart failure, n (%)	153 (9.6)
Drugs	
Alpha-anti-adrenergics, n (%)	60 (3.8)
Diuretics, n (%)	369 (23.3)
Beta-blockers, n (%)	146 (9.2)
Calcium-channel blockers, n (%)	386 (24.3)
ACE-inhibitors/AA, n (%)	572 (36.0)
N of drugs, median [IQR]	3 [2; 5]

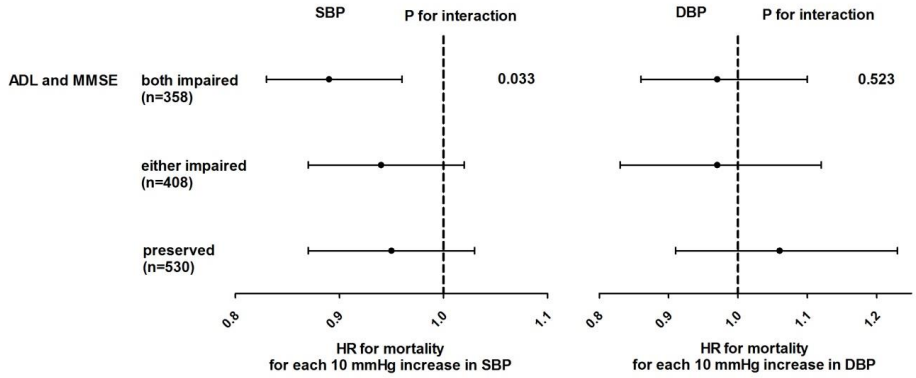
Abbreviations: n = number, IQR = inter quartile range, ADL = activities of daily living, MMSE = mini mental state examination, TIA = transient ischemic attack, ACE = angiotensin-converting-enzyme, AA = angiotensin II antagonists.

Figure 1. Risk of 10-year mortality in SBP and DBP categories



Bars represent hazard ratios (95% confidence interval). The category of SBP 160-179 mmHg and the category of DBP 90-99 mmHg were set as references in the left and right graph, respectively. The symbol * indicates a significant difference with the reference category. Analyses were adjusted for age, sex, education, smoke, hypertension, diabetes mellitus, atrial fibrillation, coronary heart disease, claudication, history of transient ischemic attack or stroke, depression/anxiety, cancer, number of medications, alpha-anti-adrenergics, diuretics, beta-blockers, calcium-channel blockers, angiotensin-converting-enzyme inhibitors/angiotensin II antagonists. Abbreviations: HR = hazard ratio, SBP = systolic blood pressure, DBP = diastolic blood pressure. Number of patients in each SBP category: SBP <120, n=74; SBP 120-139, n=376; SBP 140-159, n=647; SBP 160-179, n=354; SBP≥180, n=136. Number of patients in each DBP category: DBP <80, n=320; DBP 80-89, n=764; DBP 90-99, n=364; DBP ≥100, n=139.

Figure 2. Risk of 10-year mortality for each 10mmHg increase in SBP / DBP stratified for functional and cognitive status



Bars represent hazard ratios (95% confidence interval). Impaired ADL was defined as ADL score ≤ 5 and impaired MMSE as MMSE score ≤ 24. Analyses were adjusted for age, sex, education, smoke, hypertension, diabetes mellitus, atrial fibrillation, coronary heart disease, claudication, history of transient ischemic attack or stroke, depression/anxiety, cancer, number of medications, alpha-anti-adrenergics, diuretics, beta-blockers, calcium-channel blockers, angiotensin-converting-enzyme inhibitors / angiotensin II antagonists. Abbreviations: HR = hazard ratio, SBP = systolic blood pressure, DBP = diastolic blood pressure, ADL = activities of daily living, MMSE = mini mental state examination.

Supplementary Table 1. Baseline characteristics of study population in categories of systolic blood pressure

Characteristic	Systolic blood pressure categories (mmHg)					p-value
	<120 n=74	120 - 139 n=376	140 – 159 n=647	160 - 179 n=354	≥180 n=136	
Demographics						
Age, years median [IQR]	82 [78; 89]	82 [78; 87]	81 [78; 85]	82 [79; 87]	83 [79; 87]	0.092
Females, n (%)	54 (73.0)	252 (67.0)	433 (66.9)	276 (78.0)	99 (72.8)	0.003
Education, years, median [IQR]	6 [5; 12]	7 [5; 12]	7 [5; 10]	6 [5; 10]	8 [5; 12]	0.857
Cardiovascular risk factors						
Ever smoker, n (%)	26 (35.1)	144 (38.3)	230 (35.5)	121 (34.2)	45 (33.1)	0.757
Hypertension, n (%)	42 (56.8)	195 (51.9)	409 (63.2)	259 (73.2)	112 (82.4)	<0.001
Functional/cognitive status						
ADL score, median [IQR]	4 [2.5; 5.5]	5.5 [4; 6]	5.5 [4.5; 6]	5.5 [4.5; 6]	5.5 [4.5; 6]	<0.001
MMSE score, median [IQR]	23 [15; 27]	25 [19; 28]	25 [21; 29]	26 [21; 29]	26 [22; 29]	0.004
Co-morbidities, n (%)						
Diabetes mellitus	5 (6.8)	33 (8.8)	86 (13.3)	51 (14.4)	19 (14.0)	0.064
Atrial fibrillation	15 (20.3)	59 (15.7)	97 (15.0)	46 (13.0)	18 (13.2)	0.527
Coronary heart disease	25 (33.8)	97 (25.8)	159 (24.6)	91 (25.7)	21 (15.4)	0.041
Claudication	7 (9.5)	26 (6.9)	40 (6.2)	20 (5.6)	8 (5.9)	0.777
Depression/anxiety	39 (52.7)	183 (48.7)	313 (48.4)	180 (50.8)	57 (41.9)	0.454
Stroke/TIA	16 (21.6)	66 (17.6)	103 (15.9)	56 (15.8)	26 (19.1)	0.644
Cancer	6 (8.1)	42 (11.2)	60 (9.3)	26 (7.3)	7 (5.1)	0.200
Heart failure	14 (18.9)	33 (8.8)	70 (10.8)	28 (7.9)	8 (5.9)	0.017
Drugs, n (%)						
Alpha-anti-adrenergics	1 (1.4)	8 (2.1)	19 (2.9)	21 (5.9)	11 (8.1)	0.002
Diuretics	22 (29.7)	95 (25.3)	157 (24.3)	68 (19.2)	27 (19.9)	0.131
Beta-blockers	6 (8.1)	32 (8.5)	56 (8.7)	34 (9.6)	18 (13.2)	0.514
Calcium-channel blockers	9 (12.2)	90 (23.9)	165 (25.5)	88 (24.9)	34 (25.0)	0.161
ACE-inhibitors/AA	23 (31.1)	107 (28.5)	226 (34.9)	153 (43.2)	63 (46.3)	<0.001
N of drugs, median [IQR]	4 [2; 6]	3 [2; 5]	3 [2; 5]	4 [2; 5]	3 [1; 5]	0.092

Abbreviations: n = number, IQR = inter quartile range, ADL = activities of daily living, MMSE = mini mental state examination, TIA = transient ischemic attack, ACE = angiotensin-converting-enzyme, AA = angiotensin II antagonists. P-values were computed using chi-squared test for categorical variables and Kruskal-Wallis test for continuous variables.

Supplementary Table 2. Baseline characteristics of study population in categories of diastolic blood pressure

Characteristic	Diastolic blood pressure categories (mmHg)				p-value
	<80	80 - 89	90 – 99	≥100	
	n=320	n=764	n=364	n=139	
Demographics					
Age, years median [IQR]	82 [78; 87]	82 [78; 86]	82 [78; 86]	81 [78; 85]	0.462
Females, n (%)	212 (66.2)	537 (70.3)	260 (71.4)	105 (75.5)	0.208
Education, years, median [IQR]	6 [5; 10]	8 [5; 12]	6 [5; 10]	6 [5; 12]	0.162
Cardiovascular risk factors					
Ever smoker, n (%)	131 (40.9)	269 (35.2)	118 (32.4)	48 (34.5)	0.127
Hypertension, n (%)	174 (54.4)	470 (61.5)	262 (72.0)	111 (79.9)	<0.001
Functional/cognitive status					
ADL score, median [IQR]	5 [3.5; 6]	5.5 [4.5; 6]	5.5 [4.5; 6]	5.5 [4.5; 6]	<0.001
MMSE score, median [IQR]	25 [16; 28]	26 [21; 29]	26 [22; 29]	26 [21; 29]	0.001
Co-morbidities, n (%)					
Diabetes mellitus	39 (12.2)	91 (11.9)	48 (13.2)	16 (11.5)	0.930
Atrial fibrillation	60 (18.8)	98 (12.8)	59 (16.2)	18 (12.9)	0.064
Coronary heart disease	94 (29.4)	184 (24.1)	80 (22.0)	35 (25.2)	0.147
Claudication	30 (9.4)	47 (6.2)	19 (5.2)	5 (3.6)	0.057
Depression/anxiety	165 (51.6)	365 (47.8)	178 (48.9)	64 (46.0)	0.635
Stroke/TIA	60 (18.8)	141 (18.5)	48 (13.2)	18 (12.9)	0.065
Cancer	37 (11.6)	61 (8.0)	37 (10.2)	6 (4.3)	0.048
Heart failure	47 (14.7)	66 (8.6)	28 (7.7)	12 (8.6)	0.007
Drugs, n (%)					
Alpha-anti-adrenergics	8 (2.5)	21 (2.7)	20 (5.5)	11 (7.9)	0.004
Diuretics	81 (25.3)	193 (25.3)	64 (17.6)	31 (22.3)	0.028
Beta-blockers	33 (10.3)	59 (7.7)	42 (11.5)	12 (8.6)	0.179
Calcium-channel blockers	73 (22.8)	197 (25.8)	90 (24.7)	26 (18.7)	0.296
ACE-inhibitors/AA	100 (31.2)	272 (35.6)	136 (37.4)	64 (46.0)	0.023
N of drugs, median [IQR]	4 [2; 5]	3 [2; 5]	3 [2; 5]	3 [2; 5]	0.181

Abbreviations: n = number, IQR = inter quartile range, ADL = activities of daily living, MMSE = mini mental state examination, TIA = transient ischemic attack, ACE = angiotensin-converting-enzyme, AA = angiotensin II antagonists. P-values were computed using chi-squared test for categorical variables and Kruskal-Wallis test for continuous variables.

Supplementary Table 3. Risk of mortality for each 10mmHg increase in systolic blood pressure

	Model 1		Model 2		Model 3	
	HR [95% C.I.]	p-value	HR [95% C.I.]	p-value	HR [95% C.I.]	p-value
All participants						
at 1-year	0.86 [0.77-0.97]	0.011	0.86 [0.76-0.98]	0.019	0.87 [0.77-0.99]	0.035
at 5-year	0.90 [0.85-0.94]	<0.001	0.90 [0.85-0.95]	<0.001	0.91 [0.86-0.96]	0.001
at 10-year	0.91 [0.88-0.95]	<0.001	0.91 [0.88-0.95]	<0.001	0.92 [0.88-0.96]	<0.001
Age strata*						
75-79 years (n=500)	0.92 [0.85-1.00]	0.051	0.88 [0.81-0.97]	0.006	0.89 [0.81-0.97]	0.010
80-84 years (n=470)	0.94 [0.87-1.01]	0.083	0.95 [0.88-1.02]	0.131	0.95 [0.88-1.03]	0.204
85+ years (n=481)	0.87 [0.82-0.93]	<0.001	0.88 [0.83-0.94]	<0.001	0.88 [0.83-0.94]	<0.001
ADL strata*						
ADL≤5 (n=594)	0.90 [0.86-0.95]	<0.001	0.90 [0.86-0.96]	<0.001	0.91 [0.86-0.96]	0.001
ADL>5 (n=802)	0.95 [0.89-1.01]	0.119	0.94 [0.88-1.01]	0.079	0.94 [0.88-1.01]	0.085
MMSE strata*						
MMSE≤24 (n=586)	0.89 [0.84-0.94]	<0.001	0.90 [0.85-0.95]	<0.001	0.90 [0.85-0.96]	0.001
MMSE>24 (n=765)	0.95 [0.90-1.01]	0.098	0.94 [0.89-1.00]	0.064	0.94 [0.89-1.01]	0.070
Hypertension strata*						
Without (n=546)	0.90 [0.84-0.96]	0.002	0.89 [0.83-0.96]	0.002	0.89 [0.83-0.96]	0.003
With (n=905)	0.91 [0.86-0.95]	<0.001	0.91 [0.87-0.96]	<0.001	0.92 [0.87-0.97]	0.001
CVD strata*						
Without (n=880)	0.91 [0.86-0.96]	0.001	0.90 [0.85-0.95]	<0.001	0.91 [0.86-0.96]	0.001
With (n=571)	0.92 [0.86-0.97]	0.003	0.93 [0.87-0.98]	0.013	0.93 [0.88-0.99]	0.033
Restricted samples						
No heart failure* (n=1306)	0.91 [0.87-0.95]	<0.001	0.91 [0.87-0.95]	<0.001	0.91 [0.87-0.95]	<0.001
First year survivors* (n=1280)	0.92 [0.88-0.96]	<0.001	0.92 [0.88-0.96]	<0.001	0.92 [0.88-0.96]	<0.001

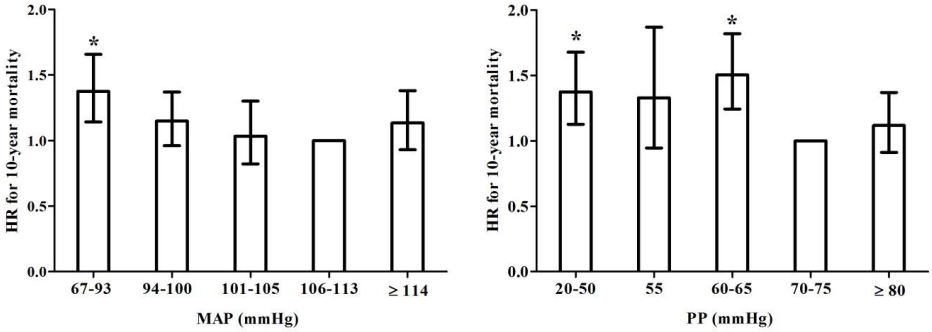
[a]Model 1: adjusted for age and sex; [b]Model 2: age, sex, education, ever smoker, number of medications, hypertension, diabetes mellitus, atrial fibrillation, coronary heart disease, claudication, history of transient ischemic attack or stroke, depression/anxiety, cancer; [c]Model 3: age, sex, education, smoke, hypertension, diabetes mellitus, atrial fibrillation, coronary heart disease, claudication, history of transient ischemic attack or stroke, depression/anxiety, cancer, number of medications, alpha-anti-adrenergics, diuretics, beta-blockers, calcium-channel blockers, angiotensin-converting-enzyme inhibitors/angiotensin II antagonists. Abbreviations: HR = hazard ratio, CI = confidence interval, SBP = systolic blood pressure, CVD = cardiovascular disease. The symbol * indicates hazard ratio for 10-year mortality risk.

Supplementary Table 4. Risk of mortality for each 10mmHg increase in diastolic blood pressure

	Model 1		Model 2		Model 3	
	HR [95% C.I.]	p-value	HR [95% C.I.]	p-value	HR [95% C.I.]	p-value
All participants						
at 1-year	0.86 [0.70-1.06]	0.164	0.85 [0.68-1.07]	0.171	0.86 [0.68-1.08]	0.186
at 5-year	0.95 [0.86-1.05]	0.294	0.98 [0.89-1.09]	0.741	0.99 [0.90-1.09]	0.839
at 10-year	0.95 [0.89-1.03]	0.190	0.98 [0.91-1.06]	0.580	0.98 [0.91-1.06]	0.634
Age strata*						
75-79 years (n=500)	0.94 [0.81-1.09]	0.428	0.94 [0.81-1.10]	0.440	0.95 [0.81-1.11]	0.528
80-84 years (n=470)	1.02 [0.90-1.16]	0.795	1.07 [0.93-1.22]	0.357	1.07 [0.94-1.23]	0.307
85+ years (n=481)	0.91 [0.81-1.02]	0.093	0.90 [0.80-1.01]	0.070	0.89 [0.79-1.00]	0.054
ADL strata*						
ADL≤5 (n=594)	0.93 [0.85-1.03]	0.186	0.95 [0.86-1.06]	0.368	0.96 [0.87-1.07]	0.454
ADL>5 (n=802)	1.00 [0.89-1.11]	0.944	1.03 [0.92-1.15]	0.617	1.03 [0.92-1.15]	0.648
MMSE strata*						
MMSE≤24 (n=586)	0.93 [0.84-1.03]	0.172	0.97 [0.87-1.08]	0.572	0.97 [0.87-1.08]	0.550
MMSE>24 (n=765)	1.03 [0.92-1.16]	0.592	1.04 [0.92-1.17]	0.552	1.04 [0.92-1.17]	0.521
Hypertension strata*						
Without (n=546)	0.92 [0.81-1.05]	0.224	0.96 [0.84-1.10]	0.577	0.96 [0.84-1.10]	0.576
With (n=905)	0.95 [0.87-1.04]	0.248	0.97 [0.88-1.06]	0.481	0.97 [0.89-1.07]	0.565
CVD strata*						
Without (n=880)	0.98 [0.89-1.09]	0.758	0.98 [0.89-1.09]	0.732	0.98 [0.89-1.09]	0.747
With (n=571)	0.93 [0.83-1.04]	0.181	0.98 [0.87-1.10]	0.721	0.98 [0.87-1.10]	0.718
Restricted samples						
No heart failure* (n=1306)	0.96 [0.88-1.04]	0.297	0.98 [0.90-1.06]	0.570	0.98 [0.90-1.07]	0.610
First year survivors* (n=1280)	0.97 [0.89-1.05]	0.387	1.00 [0.92-1.08]	0.965	1.00 [0.92-1.08]	0.996

[a]Model 1: adjusted for age and sex; [b]Model 2: age, sex, education, ever smoker, number of medications, hypertension, diabetes mellitus, atrial fibrillation, coronary heart disease, claudication, history of transient ischemic attack or stroke, depression/anxiety, cancer; [c]Model 3: age, sex, education, smoke, hypertension, diabetes mellitus, atrial fibrillation, coronary heart disease, claudication, history of transient ischemic attack or stroke, depression/anxiety, cancer, number of medications, alpha-anti-adrenergics, diuretics, beta-blockers, calcium-channel blockers, angiotensin-converting-enzyme inhibitors/angiotensin II antagonists. Abbreviations: HR = hazard ratio, CI = confidence interval, DBP = systolic blood pressure, CVD = cardiovascular disease. The symbol * indicates that we report hazard ratio for 10-year mortality risk.

Supplementary Figure 1. Risk of 10-year mortality in MAP and PP quintiles



Bars represent hazard ratios (95% confidence interval). The category of MAP 106-113 mmHg and the category of PP 70-75 mmHg were set as references in the left and right graph, respectively. The symbol * indicates a significant difference with the reference category. Analyses were adjusted for age, sex, education, smoke, hypertension, diabetes mellitus, atrial fibrillation, coronary heart disease, claudication, history of transient ischemic attack or stroke, depression/anxiety, cancer, number of medications, alpha-anti-adrenergics, diuretics, beta-blockers, calcium-channel blockers, angiotensin-converting-enzyme inhibitors/angiotensin II antagonists. Abbreviations: HR = hazard ratio, MAP = mean arterial blood pressure, PP = pulse pressure. Number of patients in MAP quintiles from first to fifth: n=325; n=407; n=171; n=405; n=279. Number of patients in PP quintiles from first to fifth: n=406; n=70; n=469; n=291; n=351.

Chapter 4

Thyroid status and mortality

Manuscript based on this chapter has been submitted as:

Ogliari G, Smit RA, van der Spoel E, Mari D, Torresani E, Felicetta I, Lucchi TA, Rossi PD, van Heemst D, de Craen AJ, Westendorp RG. Thyroid status and mortality risk in euthyroid older adults: sex-differences in the Milan Geriatrics 75+ Cohort Study.

ABSTRACT

Background: Optimal thyroid status in old age is controversial. This study investigated the longitudinal association between thyroid parameters and 10-year all-cause mortality risk in euthyroid older outpatients according to sex and age.

Methods: Baseline thyrotropin (TSH), free thyroxine (fT4) and free triiodothyronine (fT3) were assessed in the Milan Geriatrics 75+ Cohort Study. 338 men and 630 women aged over 75 years were euthyroid. Hazard ratios (HRs) and 95% confidence intervals (CI) were calculated for the associations of TSH, fT4 and fT3 with mortality risk using Cox regression. Analyses were stratified by sex and adjusted for socio-demographic factors and co-morbidities.

Results: 245 men and 382 women died during follow-up. After adjustment, each 1 mU/L higher TSH was associated with decreased mortality risk in men (HR 0.81, 95% CI 0.68-0.96), but not in women (HR 1.09, 95% CI 0.96-1.23) (p for sex-difference = 0.002). Each 1 ng/L higher fT4 was associated with increased mortality risk in men (HR 1.12, 95% CI 1.03-1.21), whereas not in women (HR 0.99, 95% CI 0.94-1.04) (p for sex-difference = 0.008). Each 1 pg/mL higher fT3 was associated with decreased mortality risk in both men (HR 0.78, 95% CI 0.55-1.10) and women (HR 0.78, 95% CI 0.62-0.99). The inverse association between TSH and mortality was most pronounced in men aged over 85 years.

Conclusions: Among euthyroid older outpatients, higher TSH and lower fT4 were associated with decreased mortality risk in men but not in women. When assessing thyroid status, sex and age should be taken into account.

INTRODUCTION

Thyroid status can be assessed by measuring serum thyrotropin (TSH), free thyroxine (fT4) and free triiodothyronine (fT3). Optimal thyroid status in old age, particularly the normal TSH reference range, is controversial[1,2]. Lowering TSH upper reference limit from 4.00 to 2.50 mU/L is highly debated[3], as TSH distribution progressively shifts towards higher values with aging[4]. This shift may arise from a higher prevalence of occult thyroid disease; indeed, euthyroid adults with higher TSH have an increased risk of hypothyroidism[5]. Alternatively, this shift may result from selective survival of individuals with constitutively lower thyroid status. Indeed, exceptionally long-lived adults and their offspring exhibit higher normal TSH with unchanged fT4, possibly indicative of a different set-point of the pituitary-thyroid axis[6]. A genetic influence on thyroid status is also supported by twin studies[7] and by the observation that intra-individual variation in thyroid status is smaller than inter-individual variation[8]. In addition, sex may modulate the effect of several genetic variants for TSH and fT4 levels[9].

TSH, fT4 and fT3 have profound and pleiotropic effects on aging individuals, by influencing metabolism, cardiovascular function and mental health[10]. These effects may differ in men and women[10,11]. Furthermore, the relationship between TSH and mortality risk in euthyroid adults is unclear, with some studies reporting no association[12,13] and others an inverse association[14-18]. The relationship of fT4 and fT3 with mortality risk is ambiguous[12,15,19]. Finally, most current evidence is from population-based studies on adults with wide age ranges, which limits their generalizability. Data are lacking on older outpatients, a potentially diverse population, whom clinicians encounter in everyday clinical practise. Older outpatients may present a higher burden of comorbidities, in a complex interplay with thyroid status.

Therefore, we assessed the association between thyroid status and mortality risk in euthyroid older men and women enrolled in the Milan Geriatrics 75+ Cohort Study, a longitudinal geriatric outpatient cohort. Furthermore, we investigated whether it differs by sex and age.

METHOD

Study Design and Participants

The Milan Geriatrics 75+ Cohort Study is a prospective hospital-based cohort study of the outpatients of the Geriatric Unit of ''I.R.C.C.S. Ca' Granda'' in Milan, Italy. Between 3 January 2000 and 25 March 2004, 1861 new consecutive outpatients aged 75 years and over attended a first face-to-face, standardized, structured, comprehensive visit with trained physicians, after informed consent. Details of study design were previously described[20].

To explore the association between the natural course of euthyroid function, unmodified by medical intervention, and mortality, we excluded participants on thyroid medications (n=74), and those with baseline TSH < 0.20 mU/L or > 4.00 mU/L or missing (n=768). Additionally, we excluded participants with missing data on mortality at follow-up (n=51). Therefore, we included 968 euthyroid participants in the present analysis. These included participants were younger and more likely to be men, smokers and to have depression/anxiety compared to the excluded participants (data not shown). Of the included participants, 761 and 708 participants, respectively, had available data on fT4 and fT3; we performed our analyses on fT4 and fT3 in those with values within the reference range (n=736 and n=651, respectively). The study was approved by I.R.C.C.S. Ca' Granda Ethics Committee.

Thyroid parameters

Blood for baseline measurements was drawn in the morning, after an overnight fast. TSH, fT4 and fT3 were measured in serum using chemiluminescent assays (Immulite 2000, Medical Systems). IRCCS Ca' Granda Laboratory reference ranges were 0.20-4.00 mU/L for TSH, 8.0-18.0 ng/L for fT4 and 2.0-4.8 pg/mL for fT3.

Co-morbidities and life-style factors

Baseline data on history of hypertension, diabetes mellitus, coronary heart disease (CHD), transient ischemic attack (TIA) or stroke, atrial fibrillation, claudication and heart failure were obtained from medical documents. Cancer was defined as a diagnosis within the previous five years. Symptoms of anxiety/depression were self-reported or stated in medical documents.

Smoking was dichotomized as never or ever (current and previous). Education was defined as years of school attended. Number of medications was the number of drugs taken chronically or cyclically.

Mortality

All-cause mortality was assessed through the Register Office of Milan or other town of residence. The follow-up period was the time between baseline and either death, loss to follow-up or 10-year period.

Statistical analyses

Baseline characteristics were reported as mean (standard deviation, SD) for continuous variables and number (percentage) for categorical variables. Differences in baseline characteristics between sexes or across TSH quartiles were assessed using Student's t-test, one-way ANOVA or chi-square test where appropriate. We checked whether fT4 was inversely associated with the logarithm of TSH in our cohort, as previously shown in the literature[21], using linear regression.

We performed Cox regression to estimate hazard ratios (HRs) and 95% confidence intervals (CI) for the association of TSH, fT4 and fT3, respectively, with mortality risk in men and women, separately. First, we tested the presence of linear associations between thyroid parameters and mortality risk. Second, we checked the presence of non-linear associations, by entering thyroid parameters and squared thyroid parameters in the Cox regression as continuous variables. Third, we performed additional analyses using quartiles of TSH, fT4 and fT3. Finally, we ran further analyses using three categories of TSH values, which were defined according to clinical cut-offs (TSH 0.20-0.39; TSH 0.40-2.50; TSH 2.51-4.00 mU/L)[2,22].

To explore sex-differences in the relationship between thyroid parameters and mortality risk, we computed interaction terms by multiplying thyroid parameters, as continuous variables, by sex.

Furthermore, we examined the association between thyroid parameters and mortality risk within three age strata (75–79, 80–84, ≥85 years). We tested for interaction between thyroid

parameters and age, in men and women, separately. Moreover, we checked for sex-differences within age strata.

We performed sensitivity analyses after exclusion of participants who were on medications potentially affecting thyroid function (amiodar or lithium) or who had a history of thyroid disease. Furthermore, we performed sensitivity analyses for the association of TSH with mortality risk restricted to those participants with both fT4 and fT3 within the reference range.

All analyses were performed in two steps. In Model 1, analyses were adjusted for age. In Model 2, they were additionally adjusted for education, smoking, hypertension, diabetes mellitus, atrial fibrillation, CHD, claudication, stroke/TIA, depression/anxiety, cancer and number of medications. Analyses were performed using SPSS version 20.0.0 (SPSS Inc., Chicago, IL).

RESULTS

Table 1 shows the baseline characteristics of the participants by sex and across quartiles of TSH. In the cohort, mean age was 82 years (range 75-98) and 630 (65.1%) participants were women. Mean TSH, fT4 and fT3 were 1.7 mU/L, 12.2 ng/L and 2.9 pg/mL, respectively, and did not differ by sex (all p-values > 0.05). Men were more educated, more likely to be smokers, to have atrial fibrillation, CHD, claudication, stroke/TIA or cancer, while less likely to have depression/anxiety compared to women (all p-values < 0.05). Baseline characteristics of men did not differ across quartiles of TSH. In contrast, women with higher TSH were more likely to have a history of stroke/TIA (p-value = 0.004).

We observed an inverse relationship between fT4 and the logarithm of TSH in both men (β= -0.144, 95% CI -0.291;-0.002, p=0.054) and women (β= -148, 95% CI -0.248;-0.047, p=0.004).

After 10-year follow-up, 245 (72.5%) men and 382 (60.6%) women had died. At 10-year follow-up, higher TSH and lower fT4 were linearly associated with decreased mortality risk in men, whereas higher fT3 was linearly associated with decreased mortality risk in women (all p-values < 0.05, Figure 1). At 10-year follow-up, no U-shaped associations were observed (all p-values for quadratic associations >0.05).

In contrast, the association between TSH and 5-year mortality risk in women was U-shaped, even after full adjustment (p-value for quadratic association = 0.009, Supplementary Figure 1).

All other associations at 1-year and 5-year follow-up were similar to those observed at 10-year follow-up (Supplementary Table 1).

Figure 2 shows the effect of sex on the associations of TSH, fT4 and fT3 with 10-year mortality risk. Sex significantly modified the associations of TSH and fT4 with mortality. After full adjustment, each 1 mU/L higher TSH was associated with a 0.81-fold (95% CI 0.68-0.96, p = 0.013) decreased mortality risk in men, whereas not in women (HR 1.09, 95% CI 0.96-1.23, p = 0.206) (p for sex-difference = 0.002). Likewise, each 1 ng/L higher fT4 was associated with a 1.12-fold (95% CI 1.03-1.21, p = 0.008) increased mortality risk in men, whereas not in women (HR 0.99, 95% CI 0.94-1.04, p = 0.660) (p for sex-difference = 0.008). No sex-difference was observed in the relationship between fT3 and mortality. Each 1 pg/mL higher fT3 was associated with decreased mortality risk in men (HR 0.78, 95% CI 0.55-1.10, p =0.151) and women (HR 0.78, 95% CI 0.62-0.99, p = 0.037).

Figure 3 illustrates the influence of age on the association between TSH and mortality risk at 10-year follow-up. After full adjustment, each 1 mU/L increase in TSH was associated with a 0.77-fold (95% CI 0.55-1.08, p = 0.125) and with a 0.63-fold (95% CI 0.46-0.87, p = 0.006) decreased mortality risk in men aged 80-84 years and 85 years and over, respectively. In contrast, the association tended to revert in men aged 75-79 years (HR 1.12, 95% CI 0.82-1.52, p = 0.495). In men, interaction by age was significant (p = 0.006). In women, we observed neither association between TSH and mortality risk in any age strata nor interaction by age.

Sex-differences in the relationship between TSH and mortality risk were not present in participants aged 75-79 years (p = 0.769), whereas they appeared in those aged 80-84 (p = 0.042) and 85 years and over (p = 0.004) (Figure 3).

We found no interaction by age in the relationships of fT4 and fT3 with mortality risk in either men or women (all p > 0.05, data not shown).

Figure 4 shows the association between clinical categories of TSH and 10-year mortality risk by sex. After full adjustment, men with TSH 2.51-4.00 mU/L had a 0.61-fold (95% CI 0.41-0.92, p = 0.017) decreased mortality risk than men in the middle category. In contrast, women with TSH 2.51-4.00 mU/L had a 1.19-fold (95% CI 0.90-1.56, p = 0.221) increased mortality risk than women in the middle category.

The results did not materially change in sensitivity analyses after exclusion of participants taking amiodar (n=22), lithium (n=1) or with previous thyroid disease (n=12) (data not shown). Likewise, the association between TSH and mortality risk remained essentially unchanged when restricting the analyses to participants with both fT4 and fT3 within the reference range (n=632) (data not shown).

DISCUSSION

Among euthyroid older adults in an outpatient setting, higher TSH and lower fT4 were associated with decreased mortality risk in men, but not in women. The associations of TSH and fT4 with mortality risk significantly differed by sex. The inverse association between TSH and mortality risk was most pronounced in men aged 85 years and over. All associations were independent of cardiovascular risk factors and comorbidities.

Our finding of an inverse relationship between TSH and mortality risk in men is in line with previous population-based studies in older adults[14-18], whereas others showed no association[12,13]. The discrepancies among studies may result from differences in the age- and sex- structure of the studied populations. Indeed, the novelty of our study is to report sex-differences in the relationship between TSH and mortality risk.

Why does sex modify the relationship between thyroid status and mortality? First, women compared to men have higher prevalence and incidence of subclinical and overt thyroid dysfunctions, which have been associated with an excess of mortality[23-25]. TSH values at the upper and lower limits of our laboratory reference range may reflect occult thyroid diseases in women, while not in men. Our finding of a U-shaped relationship between TSH and mortality risk at 5-year follow-up only in women is consistent with this hypothesis. Second, sex modifies the relationship between morbidity and mortality[26]. Women live longer than men, by surviving diseases that are fatal in men[26].

High normal thyroid status, as characterised by lower TSH and higher fT4 within the reference range, has been linked to adverse health outcomes[10]. These may result from different pathophysiological mechanisms, including increased metabolic rate and altered cardiovascular hemodynamic[27]. High normal thyroid status has been linked to increased heart rate and incident atrial fibrillation, which in turn are associated with functional decline and mortality[22,28-29].

Higher fT4 has also been directly related to frailty in euthyroid community-dwelling older men[19,30].

Furthermore, high normal thyroid status may affect brain structure and function. High thyroid status may favour thromboembolism and brain vascular damage through a combination of atrial fibrillation, endothelial dysfunction and hypercoagulability[23]. Alternatively, it may directly cause neurodegeneration through increased oxidative stress[23]. However, controversy persists on the association between thyroid status and cognitive impairment and dementia, which, in turn, have been associated with increased mortality risk[31-34].

High normal thyroid status may be particularly detrimental in older adults with cardiovascular comorbidities[27]. Consistent with this hypothesis, in our cohort, lower TSH and higher fT4 were associated with increased mortality risk in men, especially the oldest men, who presented more cardiovascular comorbidities compared to women. However, sex-differences in our study remained significant after adjustment for comorbidities.

An alternative explanation to our findings may be that the set-point of the pituitary-thyroid axis is shifted towards higher TSH values in adults with genetic predisposition to longevity[6]. Men aged over 85 years in our study had above-average life-expectancy, thus suggesting a genetic longevity trait[35]. Animal studies have suggested a causal relationship between lower thyroid status and extended life span[36]. Lower thyroid status may extend life span by lowering metabolic rate and core body temperature, which in turn results in lower generation of reactive oxygen species and oxidative stress[36]. Other mechanisms may include effects on membrane composition, inflammation and stem cell renewal[37].

Our finding of an association between lower fT3 and increased mortality risk is in line with The Aging in the Chianti Area Study, which included Italians aged 65 years and over[15]. Lower fT3 in euthyroid older individuals may be indicative of non-thyroidal systemic illnesses[15,38].

Our study has relevant clinical implications. First, clinicians should take into account both sex and age when assessing thyroid status. Furthermore, we reported that older men with TSH 2.51 – 4.00 mU/L had a 0.61-fold decreased mortality risk than those with TSH 0.40 – 2.50 mU/L. This observational finding conflicts with the indication of lowering TSH upper reference limit, at least in older men[2].

Moreover, clinical trials on the clinical benefits or harms of lowering TSH upper reference limit in older adults are lacking[39]. Furthermore, clinical trials recruit selected populations, which limits their generalizability[39].

A major strength of our study is our unselected population of older geriatric outpatients, which makes our findings generalizable in common clinical practice. A further asset is the longitudinal design, with a long follow-up. However, the observational nature of our study limits us in inferring causality. Furthermore, a single measurement of thyroid status was used in the analyses, potentially leading to misclassification of subjects. However, previous research has demonstrated that intra-individual variability of thyroid status is narrow and less than inter-individual variability[8]. In addition, this misclassification would be random and merely lead to underestimation of true associations.

In conclusion, higher TSH and lower fT4 within the reference ranges were associated with decreased mortality risk in men but not in women. Our findings add to the current debate on TSH reference limits. Further research is needed to establish whether the relationship between thyroid status and mortality is causal.

ACKNOWLEDGEMENTS

We thank Milan Register Office.

REFERENCES

1. Jonklaas J, Bianco AC, Bauer AJ, et al. Guidelines for the treatment of hypothyroidism: prepared by the american thyroid association task force on thyroid hormone replacement. Thyroid. 2014;24:1670-751.
2. Pearce SH, Brabant G, Duntas LH, et al. 2013 ETA Guideline: Management of Subclinical Hypothyroidism. Eur Thyroid J. 2013;2:215-28.
3. Laurberg P, Andersen S, Carlé A, Karmisholt J, Knudsen N, Pedersen IB. The TSH upper reference limit: where are we at? Nat Rev Endocrinol. 2011;7:232-239.

4. Surks MI, Hollowell JG. Age-specific distribution of serum thyrotropin and antithyroid antibodies in the US population: implications for the prevalence of subclinical hypothyroidism. J Clin Endocrinol Metab. 2007;92:4575-82.
5. Åsvold BO, Vatten LJ, Midthjell K, Bjøro T. Serum TSH within the reference range as a predictor of future hypothyroidism and hyperthyroidism: 11-year follow-up of the HUNT Study in Norway. J Clin Endocrinol Metab. 2012;97:93-99.
6. Atzmon G, Barzilai N, Surks MI, Gabriely I. Genetic predisposition to elevated serum thyrotropin is associated with exceptional longevity. J Clin Endocrinol Metab. 2009;94:4768-75.
7. Hansen PS, Brix TH, Sørensen TI, Kyvik KO, Hegedüs L. Major genetic influence on the regulation of the pituitary-thyroid axis: a study of healthy Danish twins. J Clin Endocrinol Metab. 2004;89:1181-7.
8. Andersen S, Pedersen KM, Bruun NH, Laurberg P. Narrow individual variations in serum T(4) and T(3) in normal subjects: a clue to the understanding of subclinical thyroid disease. J Clin Endocrinol Metab. 2002;87:1068-72.
9. Porcu E, Medici M, Pistis G, et al. A meta-analysis of thyroid-related traits reveals novel loci and gender-specific differences in the regulation of thyroid function. PLoS Genet. 2013;9:e1003266.
10. Taylor PN, Razvi S, Pearce SH, Dayan CM. Clinical review: A review of the clinical consequences of variation in thyroid function within the reference range. J Clin Endocrinol Metab. 2013;98:3562-71.
11. Asvold BO, Bjoro T, Nilsen TI, Gunnell D, Vatten LJ. Thyrotropin levels and risk of fatal coronary heart disease: the HUNTstudy. Arch Intern Med. 2008;168:855–860.
12. Zhang Y, Chang Y, Ryu S, et al. Thyroid hormones and mortality risk in euthyroid individuals: the Kangbuk Samsung health study. J Clin Endocrinol Metab. 2014;99:2467-76.
13. Ittermann T, Haring R, Sauer S, et al. Decreased serum TSH levels are not associated with mortality in the adult northeast German population. Eur J Endocrinol. 2010;162:579–585.
14. Parle JV, Maisonneuve P, Sheppard MC, Boyle P, Franklyn JA. Prediction of all-cause and cardiovascular mortality in elderly people from one low serum thyrotropin result: a 10-year cohort study. Lancet. 2001;358:861–865.

15. Ceresini G, Ceda GP, Lauretani F, et al. Thyroid Status and 6-Year Mortality in Elderly People Living in a Mildly Iodine-Deficient Area: The Aging in the Chianti Area Study. J Am Geriatr Soc. 2013;61:868-74.
16. Gussekloo J, van Exel E, de Craen AJ, Meinders AE, Frölich M, Westendorp RG. Thyroid status, disability and cognitive function, and survival in old age. JAMA. 2004;292:2591–2599.
17. Pereg D, Tirosh A, Elis A, et al. Mortality and coronary heart disease in euthyroid patients. Am J Med. 2012;125:826 e827–e812.
18. Cappola AR, Arnold AM, Wulczyn K, Carlson M, Robbins J, Psaty BM. Thyroid function in the euthyroid range and adverse outcomes in older adults. J Clin Endocrinol Metab. 2015;100:1088-96.
19. van den Beld AW, Visser TJ, Feelders RA, Grobbee DE, Lamberts SW. Thyroid hormone concentrations, disease, physical function, and mortality in elderly men. J Clin Endocrinol Metab. 2005;90:6403–6409.
20. Ogliari G, Sabayan B, Mari D, et al. Age- and Functional Status-Dependent Association Between Blood Pressure and Cognition: The Milan Geriatrics 75+ Cohort Study. J Am Geriatr Soc 2015;63:1741-8.
21. Spencer CA, LoPresti JS, Patel A, et al. Applications of a new chemiluminometric thyrotropin assay to subnormal measurement. J Clin Endocrinol Metab. 1990;70:453-60.
22. Selmer C, Olesen JB, Hansen ML, et al. The spectrum of thyroid disease and risk of new onset atrial fibrillation: a large population cohort study. BMJ. 2012;345:e7895.
23. Cooper DS, Biondi B. Subclinical thyroid disease. Lancet. 2012;379:1142–1154.
24. Roberts CG, Ladenson PW. Hypothyroidism. Lancet. 2004;363:793–803.
25. Cooper DS. Hyperthyroidism. Lancet. 2003;362:459–468.
26. Hubbard RE, Rockwood K. Frailty in older women. Maturitas. 2011;69:203-7.
27. Klein I, Ojamaa K. Thyroid hormone and the cardiovascular system. N Engl J Med. 2001;344:501-9.
28. Palatini P, Benetos A, Julius S. Impact of increased heart rate on clinical outcomes in hypertension: implications for antihypertensive drug therapy. Drugs. 2006;66:133-144.
29. Ogliari G, Mahinrad S, Stott DJ, et al. Resting heart rate, heart rate variability and functional decline in old age. CMAJ. 2015;187:E442-9.
30. Yeap BB, Alfonso H, Chubb SA, et al. Higher free thyroxine levels are associated with frailty in older men: the Health In Men Study. Clin Endocrinol (Oxf). 2012;76:741-8.

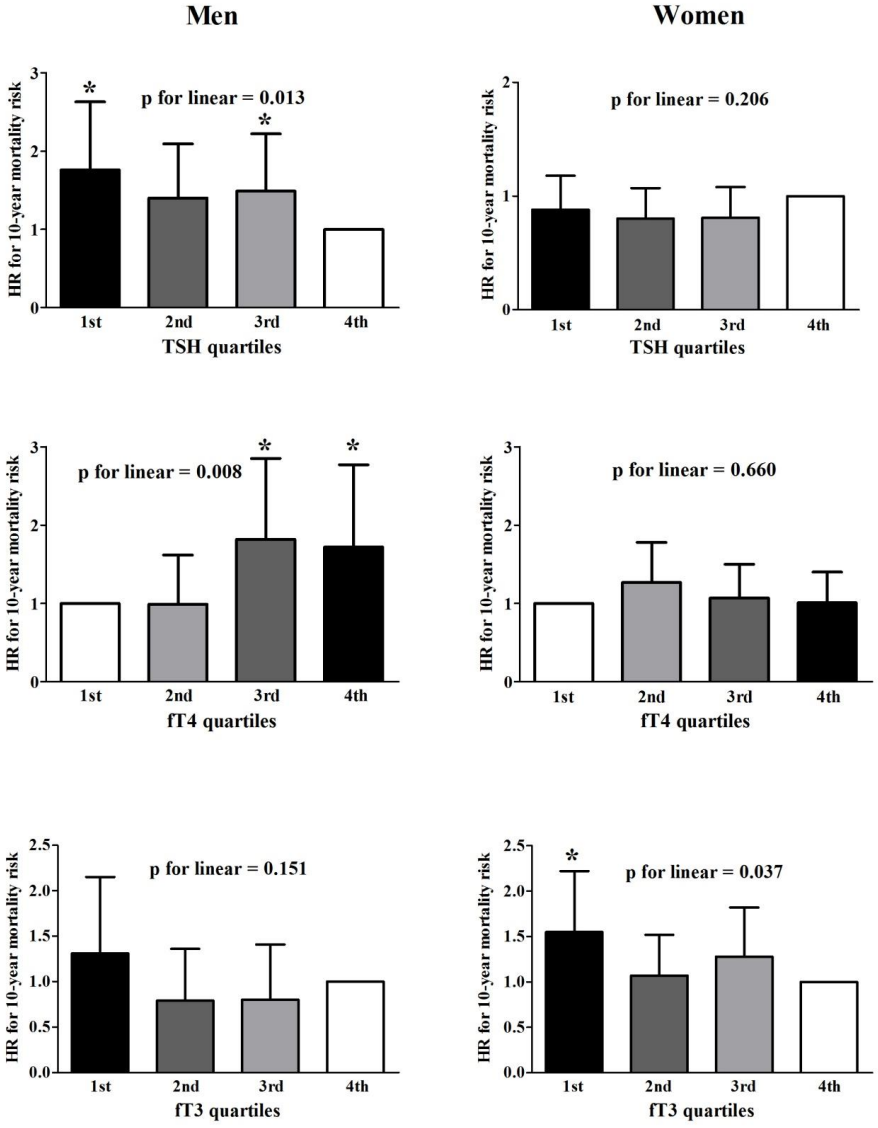
31. Tan ZS, Beiser A, Vasan RS, et al. Thyroid function and the risk of Alzheimer disease: the Framingham Study. Arch Intern Med. 2008;168:1514-20.
32. de Jong FJ, Masaki K, Chen H, et al. Thyroid function, the risk of dementia and neuropathologic changes: the Honolulu-Asia aging study. Neurobiol Aging. 2009;30:600-6.
33. Yeap BB, Alfonso H, Chubb SA, et al. Higher free thyroxine levels predict increased incidence of dementia in older men: the Health in Men Study. J Clin Endocrinol Metab. 2012;97:E2230-7.
34. World Health Organization and Alzheimer's Disease International. Dementia: A Public Health Priority, 2012 [on-line]. Available at http://www.who.int/mental_health/publications/dementia_report_2012/en. Accessed October 27, 2015.
35. Franceschi C, Motta L, Valensin S, et al. Do men and women follow different trajectories to reach extreme longevity? Italian Multicenter Study on Centenarians (IMUSCE). Aging (Milano). 2000;12:77-84.
36. Buffenstein R, Pinto M. Endocrine function in naturally long-living small mammals. Mol Cell Endocrinol. 2009;299:101-11.
37. Bowers J, Terrien J, Clerget-Froidevaux MS, et al. Thyroid hormone signaling and homeostasis during aging. Endocr Rev. 2013;34:556-89.
38. Adler SM, Wartofsky L. The nonthyroidal illness syndrome. Endocrinol Metab Clin North Am. 2007;36:657-72, vi.
39. Villar HC, Saconato H, Valente O, Atallah AN. Thyroid hormone replacement for subclinical hypothyroidism. Cochrane Database Syst Rev. 2007;(3):CD003419.

Table 1. Characteristics of study population at baseline

Characteristics	Quartiles of TSH (mU/L)					p-value
	All	First	Second	Third	Fourth	
		0.24-1.02	1.03-1.55	1.56-2.16	2.17-3.98	
Men	n=338	n=89	n=89	n=89	n=71	
Demographics, years, mean (SD)						
Age	81.6 (4.6)	81.5 (4.3)	81.2 (4.4)	81.6 (4.8)	82.3 (4.9)	0.523
Education	9.8 (5.0)	9.3 (4.6)	9.6 (5.0)	9.7 (5.2)	11.2 (5.0)	0.101
Risk factors/ comorbidities, n (%)						
Ever smoker	228 (67.5)	64 (71.9)	61 (68.5)	56 (62.9)	47 (66.2)	0.629
Hypertension	222 (65.7)	62 (69.7)	55 (61.8)	57 (64.0)	48 (67.6)	0.695
Diabetes mellitus	53 (15.7)	15 (16.9)	16 (18.0)	12 (13.5)	10 (14.1)	0.823
Atrial fibrillation	62 (18.3)	9 (10.1)	18 (20.2)	19 (21.3)	16 (22.5)	0.132
Coronary heart disease	98 (29.0)	26 (29.2)	23 (25.8)	26 (29.2)	23 (32.4)	0.842
Claudication	34 (10.1)	13 (14.6)	6 (6.7)	10 (11.2)	5 (7.0)	0.265
Depression or anxiety	136 (40.2)	46 (51.7)	35 (39.3)	30 (33.7)	25 (35.2)	0.066
Stroke or TIA	66 (19.5)	19 (21.3)	21 (23.6)	15 (16.9)	11 (15.5)	0.519
Cancer	53 (15.7)	15 (16.9)	11 (12.4)	15 (16.9)	12 (16.9)	0.799
Heart failure	35 (10.4)	5 (5.6)	11 (12.4)	7 (7.9)	12 (16.9)	0.093
N of drugs, mean (SD)	3.8 (2.4)	3.8 (2.5)	3.7 (2.3)	3.4 (2.4)	4.1 (2.4)	0.322
fT4 (ng/L), mean (SD)	12.0 (2.1)	12.3 (2.2)	12.1 (1.9)	12.0 (1.9)	11.4 (2.2)	0.147
fT3 (pg/mL), mean (SD)	2.9 (0.5)	2.9 (0.5)	2.9 (0.6)	2.9 (0.5)	2.9 (0.6)	0.995
Women	n=630	n=152	n=152	n=155	n=171	
Demographics, years, mean (SD)						
Age	82.2 (4.9)	82.4 (4.6)	82.2 (4.8)	81.4 (5.2)	82.8 (4.8)	0.074
Education	6.9 (3.8)	6.3 (3.3)	7.2 (3.7)	6.9 (3.8)	7.3 (4.3)	0.082
Risk factors/ comorbidities, n (%)						
Ever smoker	146 (23.2)	29 (19.1)	33 (21.7)	41 (26.5)	43 (25.1)	0.404
Hypertension	445 (70.6)	113 (74.3)	100 (65.8)	109 (70.3)	123 (71.9)	0.412
Diabetes mellitus	74 (11.7)	17 (11.2)	17 (11.2)	19 (12.3)	21 (12.3)	0.981
Atrial fibrillation	80 (12.7)	13 (8.6)	19 (12.5)	21 (13.5)	27 (15.8)	0.268
Coronary heart disease	132 (21.0)	33 (21.7)	25 (16.4)	29 (18.7)	45 (26.3)	0.148
Claudication	30 (4.8)	7 (4.6)	7 (4.6)	4 (2.6)	12 (7.0)	0.313
Depression or anxiety	356 (56.5)	80 (52.6)	88 (57.9)	94 (60.6)	94 (55.0)	0.514
Stroke or TIA	83 (13.2)	13 (8.6)	21 (13.8)	14 (9.0)	35 (20.5)	**0.004**
Cancer	41 (6.5)	7 (4.6)	15 (9.9)	7 (4.5)	12 (7.0)	0.186
Heart failure	50 (7.9)	12 (7.9)	14 (9.2)	10 (6.5)	14 (8.2)	0.845
N of drugs, mean (SD)	3.5 (2.3)	3.3 (2.2)	3.4 (2.3)	3.4 (2.3)	3.9 (2.3)	0.115
fT4 (ng/L), mean (SD)	12.3 (2.1)	12.6 (1.9)	12.3 (2.2)	12.5 (2.3)	11.8 (2.1)	**0.026**
fT3 (pg/mL), mean (SD)	3.0 (0.5)	2.9 (0.5)	3.0 (0.5)	3.0 (0.6)	3.0 (0.6)	0.456

P-values were calculated using ANOVA or chi-square test where appropriate. Abbreviations: SD: standard deviation, n: number, TSH: thyrotropin, fT4: free thyroxine, fT3: triiodothyronine, TIA: transient ischemic attack.

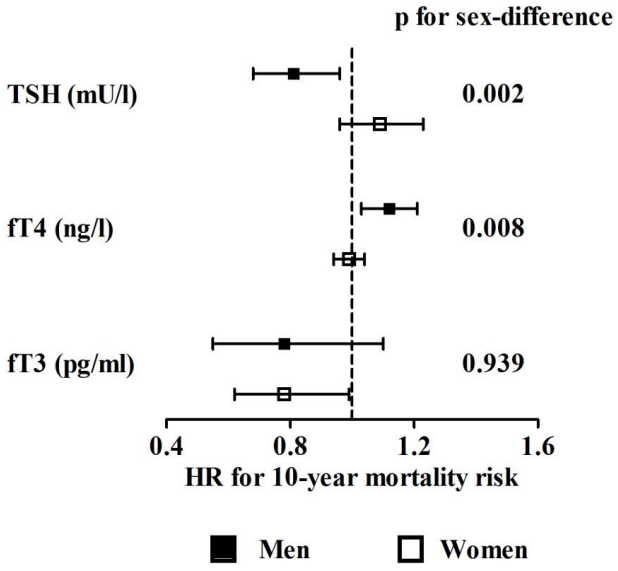
Figure 1. Association of quartiles of TSH, fT4 and fT3 with 10-year mortality risk by sex



Bars represent hazard ratios (95% confidence interval). The fourth TSH quartile, the first fT4 quartile and the fourth fT3 quartile were set as reference categories. The symbol * indicates a significant difference with the reference. P-values were computed using continuous TSH, fT4 and fT3. Analyses were adjusted for age, education, smoking, hypertension, diabetes mellitus, atrial fibrillation, CHD, claudication, heart failure, depression/anxiety, stroke/TIA, cancer, number of medications. Ranges for TSH quartiles are first: 0.24-1.02, second: 1.03-1.55, third: 1.56-2.16 and fourth: 2.17-3.98 mU/L. Ranges for fT4 quartiles are first: 8.1-10.6,

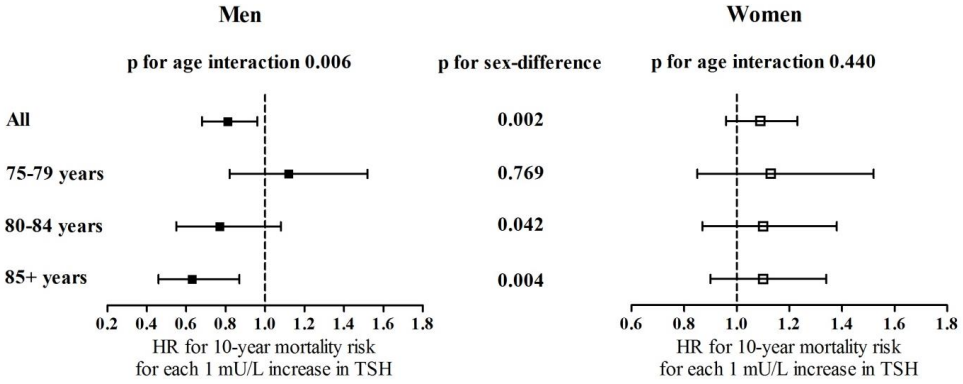
second: 10.7-11.9, third: 12.0-13.5 and fourth: 13.6-18.0 ng/L. Ranges for fT3 quartiles are first: 2.00-2.54, second: 2.55-2.90, third: 2.91-3.29 and fourth: 3.30-4.64 pg/mL. Abbreviations: HR: hazard ratio, TSH: thyrotropin, fT4: free thyroxine, fT3: triiodothyronine.

Figure 2. Association of TSH, fT4 and fT3 with 10-year mortality risk by sex



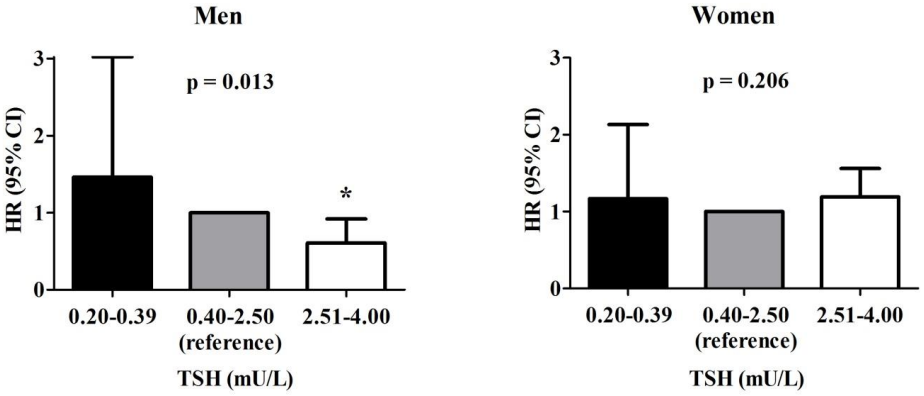
Bars represent hazard ratios (95% confidence interval) for 10-year mortality risk for each 1 mU/L increase in TSH, 1 ng/L increase in fT4 and 1 pg/mL increase in fT3. Analyses were adjusted for age, education, smoking, hypertension, diabetes mellitus, atrial fibrillation, CHD, claudication, heart failure, depression/anxiety, stroke/TIA, cancer, number of medications. Abbreviations: HR: hazard ratio, TSH: thyrotropin, fT4: free thyroxine, fT3: triiodothyronine.

Figure 3. Association between TSH and 10-year mortality risk by sex and age



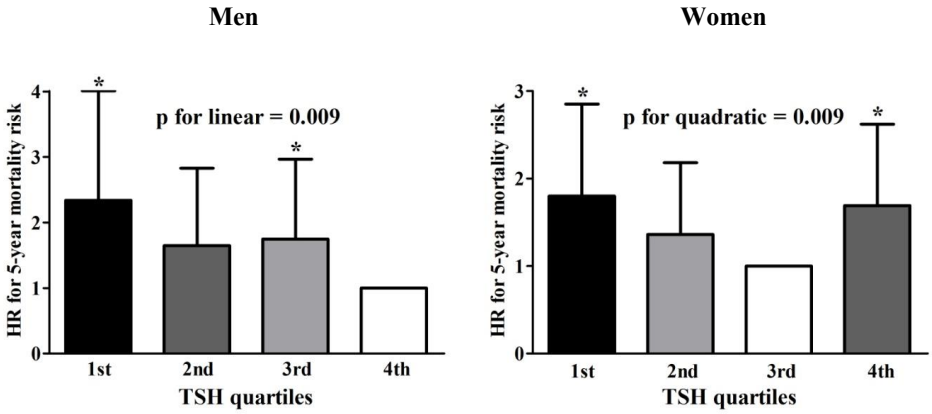
Bars represent hazard ratios (95% confidence interval). Analyses were adjusted for age, education, smoking, hypertension, diabetes mellitus, atrial fibrillation, CHD, claudication, heart failure, depression/anxiety, stroke/TIA, cancer, number of medications. P-values for age interaction were computed using TSH and age as continuous variables. Abbreviations: HR: hazard ratio, TSH: thyrotropin.

Figure 4. Association between TSH categories and 10-year mortality risk by sex



Bars represent hazard ratios (95% confidence interval). Analyses were adjusted for age, education, smoking, hypertension, diabetes mellitus, atrial fibrillation, CHD, claudication, heart failure, depression/anxiety, stroke/TIA, cancer, number of medications. Abbreviations: HR: hazard ratio, TSH: thyrotropin. The symbol * indicates a significant difference with the reference category. P-values were calculated for continuous TSH.

Supplementary Figure 1. Association of quartiles of TSH with 5-year mortality risk by sex



Bars represent hazard ratios (95% confidence interval). The fourth and the third TSH quartile were set as reference category for men and women, respectively. The symbol * indicates a significant difference with the reference. P-values for linear and for quadratic association were computed using TSH and squared TSH as continuous measures, respectively. Analyses were adjusted for age, education, smoking, hypertension, diabetes mellitus, atrial fibrillation, CHD, claudication, heart failure, depression/anxiety, stroke/TIA, cancer, number of medications. Ranges for TSH quartiles are first: 0.24-1.02, second: 1.03-1.55, third: 1.56-2.16 and fourth: 2.17-3.98 mU/L. Abbreviations: HR: hazard ratio, TSH: thyrotropin.

Supplementary Table 1. Association of TSH, fT4 and fT3 with mortality risk at different follow-ups

	Men			Women		
	HR [95% CI]	p-value for linear	p-value for quadratic	HR [95% CI]	p-value for linear	p-value for quadratic
TSH						
At 1-year	0.61 [0.35; 1.07]	0.086	0.695	0.77 [0.50; 1.18]	0.227	0.099
At 5-year	0.74 [0.59; 0.93]	**0.009**	0.818	1.05 [0.87; 1.25]	0.630	**0.009**
At 10-year	0.81 [0.68; 0.96]	**0.013**	0.977	1.09 [0.96; 1.23]	0.206	0.074
fT4						
At 1-year	1.08 [0.85; 1.38]	0.508	0.100	1.03 [0.85; 1.24]	0.766	0.335
At 5-year	1.12 [1.01; 1.24]	**0.033**	0.390	0.99 [0.91; 1.07]	0.775	0.622
At 10-year	1.12 [1.03; 1.21]	**0.008**	0.521	0.99 [0.94; 1.04]	0.660	0.909
fT3						
At 1-year	0.50 [0.14; 1.79]	0.287	0.600	0.45 [0.19; 1.09]	0.077	0.461
At 5-year	0.62 [0.38; 1.00]	**0.050**	0.321	0.59 [0.41; 0.84]	**0.004**	0.930
At 10-year	0.78 [0.55; 1.10]	0.151	0.931	0.78 [0.62; 0.99]	**0.037**	0.827

Hazard ratios (95% confidence intervals) are for each 1 mU/L increase in TSH, 1 ng/L increase in fT4 and 1 pg/mL increase in fT3. All analyses were adjusted for age, education, smoking, hypertension, diabetes mellitus, atrial fibrillation, CHD, claudication, heart failure, depression/anxiety, stroke/TIA, cancer, number of medications. P-values for linear and quadratic associations were calculated using TSH, fT4 and fT3 as continuous variables in Cox-regression. Abbreviations: HR: hazard ratio, CI: confidence interval, TSH: thyrotropin, fT4: free thyroxine, fT3: triiodothyronine.

Chapter 5

Heart rate, heart rate variability and functional decline

Manuscript based on this chapter has been published as:

Ogliari G*, Mahinrad S*, Stott DJ, Jukema JW, Mooijaart SP, Macfarlane PW, Clark EN, Kearney PM, Westendorp RG, de Craen AJ, Sabayan B. Resting heart rate, heart rate variability and functional decline in old age. CMAJ. 2015;187(15):E442-9.

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ABSTRACT

Background: Heart rate and heart rate variability, markers of cardiac autonomic function, have been related to cardiovascular diseases (CVD). We investigated whether heart rate and heart rate variability are associated with functional status in older subjects, independent of CVD.

Methods: 5042 participants, mean age 75.3 years, were enrolled in PROSPER (PROspective Study of Pravastatin in the Elderly at Risk). Heart rate and heart rate variability (SDNN) were derived from baseline 10-second electrocardiograms. Functional status in basic (ADL) and instrumental (IADL) activities of daily living was measured using Barthel and Lawton scales, at baseline and during follow-up. Mean follow-up was 3.2 years.

Results: At baseline, higher heart rate was associated with worse ADL and IADL, while lower SDNN was related to worse IADL (all p-values <0.05). Participants in the highest tertile of heart rate (range 71-117 beats/minute) had 1.79-fold (95% confidence interval (CI) 1.45-2.22) and 1.35-fold (95% CI 1.12-1.63) higher risk of decline in ADL and IADL, respectively (p for trend <0.001 and 0.001, respectively). Participants in the lowest tertile of SDNN (range 1.70-13.30 milliseconds) had 1.21-fold (95% CI 1.00-1.46) and 1.25-fold (95% CI 1.05-1.48) higher risk of decline in ADL and IADL, respectively (both p for trends <0.05). All associations were independent of sex, medications, cardiovascular risk factors and co-morbidities.

Interpretation: Higher resting heart rate and lower heart rate variability are associated with worse functional status and with higher risk of future functional decline in older subjects independent of CVD. Cardiac autonomic function correlates with the development of functional decline.

INTRODUCTION

Elevated heart rate and reduced heart rate variability — the beat-to-beat variation in heart rate intervals — both reflect an altered balance of the autonomic nervous system tone characterized by increased sympathetic and/or decreased parasympathetic activity[1-3]. Sympathetic overactivity has been linked to a procoagulant state and also to risk factors for atherosclerosis, including metabolic syndrome, obesity and subclinical inflammation[2-4]. Moreover, increased heart rate is related to atherosclerosis, not only as an epiphenomenon of sympathetic overactivity, but also through hemodynamic mechanisms, such as high pulsatile shear stress, which leads to endothelial dysfunction[5].

Atherosclerosis has been linked to increased risk of functional decline in older people via cardiovascular events[6]. As the world population is aging, the burden of functional disability is expected to increase[6]. It has been hypothesized that heart rate and heart rate variability are markers of frailty, an increased vulnerability to stressors and functional decline[7]. However, the direct link between these two parameters and risk of functional decline has not been fully established, and it is uncertain whether this association is independent of cardiovascular comorbidities.

In this study, we examined whether heart rate and heart rate variability were cross-sectionally and longitudinally associated with functional status in older adults at high risk of cardiovascular disease, independent of cardiovascular risk factors and comorbidities.

METHODS

Study design and participants

The data in this study were obtained from the Prospective Study of Pravastatin in the Elderly at Risk (PROSPER), a randomized controlled trial on the effect of pravastatin in a cohort of older men and women (70–82 yr) with pre-existing vascular disease or risk factors thereof. A total of 5804 individuals were recruited from 3 collaborating centres in Ireland, Scotland and the Netherlands. Details of study design, population recruitment and characteristics have been previously reported[8,9]. Exclusion criteria included physical or mental inability to attend clinic visits, poor cognitive function at baseline (Mini Mental State Examination score < 24),

advanced heart failure (New York Heart Association functional class III or IV), electrocardiographic (ECG) evidence of atrial fibrillation or other major arrhythmias and implanted cardiac pacemakers. Participants were followed up for a mean of 3.2 years.

From the original population, we excluded 150 participants with missing heart rate and/or heart rate variability measurements at baseline, 489 participants with cardiac rhythm not generated by sinoatrial node and 123 participants with missing data on functional status at baseline or during follow-up. We included participants from both the pravastatin and placebo arms because the PROSPER study group had previously shown that pravastatin did not affect functional status during follow-up[9]. Hence, 5042 participants were included in the present study.

The PROSPER study complied with the Declaration of Helsinki and was approved by the medical ethics committees of the 3 centres. All participants provided written informed consent.

Measurement of heart rate and heart rate variability

We measured resting heart rate and heart rate variability from a 10-second, 12-lead ECG, recorded in the morning of the first enrolment visit to limit circadian variability. All ECGs were transmitted electronically for storage at the University of Glasgow ECG Core Laboratory based at Glasgow Royal Infirmary, Scotland, and interpreted using the same software[10].

We computed the standard deviation of normal-to- normal RR intervals (SDNN), one of the most frequently used and easily calculated indices of heart rate variability, by deriving it from normal-to-normal RR intervals[11]. Normal-to-normal RR intervals were defined as the time between two successive normally conducted QRS complexes.

Functional status

Functional status was assessed using two questionnaires: the Barthel Index[12] and the Lawton Instrumental Activities of Daily Living Scale (IADL)[13]. The Barthel Index measures performance in basic activities of daily living (ADL) and consists of 10 items: fecal continence, urinary continence, grooming, toilet use, feeding, transfers (e.g., from chair to bed), walking, dressing, climbing stairs and bathing. The Lawton IADL evaluates more complex instrumental activities and includes 7 items: doing housework, taking medication as prescribed, managing

money, shopping, using a phone or other forms of communication, using technology and taking transportation within the community. Scores for ADLs and IADLs range from 0 to 20 and from 0 to 14, respectively, with higher scores indicating higher independence and better functional status. Functional status using the 2 questionnaires was measured at baseline; after 9, 18 and 30 months; and at the end of the study, which varied between 36 and 42 months. Based on changes in functional status scores during follow-up, participants were classified as either declining or not declining in ADL and IADL.

Statistical analysis

We used SPSS version 20 for all the analyses. We reported baseline characteristics of participants as number of participants (percentage) for categorical variables and as mean (standard deviation) for continuous variables. We tested differences in baseline characteristics first across heart rate tertiles and then across SDNN tertiles, using analysis of variance for continuous variables and $\chi 2$ test for categorical variables.

Linear regression analyses tested the cross-sectional associations of heart rate and SDNN with functional status. Dependent variables were the scores on each of the 2 functional status tests. We computed *p* values for trend using tertiles of heart rate and SDNN.

We performed binary logistic regression analyses to investigate longitudinal associations of heart rate and SDNN with risk of decline in functional status. Independent variables were heart rate and SDNN. The outcome variable was the risk of declining in each of the functional status tests. We calculated odds ratios (ORs) and 95% confidence intervals (CIs) in tertiles of heart rate and SDNN, respectively. The reference categories were the lowest tertile of heart rate and the highest tertile of SDNN. We calculated *p* values for trend using tertiles of heart rate and SDNN.

We performed all cross-sectional and longitudinal analyses in two steps. In the first step, analyses were adjusted for age, sex, country of enrolment and education (minimally adjusted model). In the second step, we further adjusted for cardiovascular risk factors (smoking status, body mass index [BMI], history of hypertension, history of diabetes mellitus), cardiovascular morbidities (history of myocardial infarction, history of stroke or transient ischemic attack, history of claudication), use of medications (diuretics, angiotensin-converting enzyme

inhibitors, angiotensin receptor blockers, β-blockers, calcium channel blockers, nitrates, acetylsalicylic acid, anticoagulants) and statin treatment group. In the longitudinal analyses we also adjusted for baseline functional status (fully adjusted model).

To test whether the association of heart rate and SDNN with functional status is independent of β-blocker use, we repeated the longitudinal analyses after exclusion of participants taking β-blockers. Furthermore, we repeated the longitudinal analyses after stratifying the participants by sex, history of hypertension, history of vascular diseases, use of β-blockers, calcium channel blockers or statin treatment to explore the potential modifying effect of these covariates. We computed interaction terms by multiplying heart rate and SDNN, as continuous variables, per these covariates.

To explore the influence of vascular events on the longitudinal associations, we performed sensitivity analyses from which we excluded the following: 1) participants with incident stroke, 2) participants with incident coronary events and 3) participants who were admitted to hospital for heart failure during follow-up. Furthermore, to check whether the longitudinal associations are affected by baseline functional status or by duration of follow-up, we performed sensitivity analyses including only 1) participants with maximum functional status at baseline and 2) participants who completed 36 months of follow-up.

To check whether the association between SDNN and functional status is independent of heart rate, we repeated the analyses after standardizing SDNN for heart rate (dividing SDNN by heart rate)[14].

Finally, we repeated the longitudinal analyses by dividing the participants in the lowest tertile of heart rate into two groups of participants with a heart rate of less than 50 beats/min and participants with a heart rate of 50–60 beats/min.

RESULTS

The mean age of the study population was 75.3 years. A total of 2619 (51.9%) participants were female (Table 1). The median resting heart rate and SDNN were 65 beats/min and 18.6 ms, respectively. Participants with a higher resting heart rate were older, were more likely to be female and current smokers, and had a higher BMI and a higher prevalence of diabetes

mellitus. In contrast, participants with a lower resting heart rate used β-blockers more frequently and had a higher prevalence of myocardial infarction (all p values < 0.05) (Supplementary Table 1). Participants with lower heart rate variability as measured by SDNN had a higher BMI, a higher prevalence of diabetes mellitus and less frequently used β-blockers (all p values < 0.05) (Supplementary Table 2).

Table 2 shows the associations of resting heart rate and SDNN with functional status at baseline. In the minimally adjusted model, participants with a higher resting heart rate had a worse performance in both functional status scales (p for trend < 0.05, for both). These associations remained significant in the fully adjusted model (p for trend < 0.05, for both). Likewise, participants with lower SDNN had a worse performance in both functional status scales in the minimally adjusted model (p for trend < 0.05, for both). After full adjustment, the same association persisted between SDNN and IADL (p for trend = 0.03). The same trend was observed between SDNN and ADL, although it did not reach significance (p for trend = 0.11).

During a mean follow-up of 3.2 years, 779 participants (15.5%) declined in ADL score and 1128 participants (22.4%) declined in IADL score. Among the participants who declined in ADL score, 406 (52.1%) declined 1 point, 141 (18.1%) 2 points and 232 (29.8%) 3 or more points. Among the participants who declined in IADL score, 402 (35.6%) declined 1 point, 224 (19.9%) 2 points and 502 (44.5%) 3 or more points.

Figure 1 shows the longitudinal associations of resting heart rate and SDNN with risk of decline in functional status after full adjustment. Participants with a resting heart rate in the highest tertile had a 1.79-fold (95% CI 1.45–2.22) and a 1.35-fold (95% CI 1.12–1.63) higher risk of decline in ADL and IADL scores, respectively (p for trend < 0.001 and 0.001, respectively). Participants with SDNN in the lowest tertile had 1.21-fold (95% CI 1.00–1.46) and 1.25-fold (95% CI 1.05–1.48) higher risk of decline in ADL and IADL scores, respectively (p for trend < 0.05, for both groups). These associations were similar in the minimally adjusted model (p for trend < 0.05, for all groups) (Supplementary Table 3).

Table 3 shows the sensitivity analyses after exclusion of the 1320 participants receiving treatment with β-blockers. Higher resting heart rate and lower SDNN remained significantly related to a higher risk of decline for both ADL and IADL in the fully adjusted model (p for trend < 0.05, for all groups). To clarify whether cardiovascular events during follow-up might affect the longitudinal associations between resting heart rate/SDNN and risk of decline in

functional status, we performed a series of sensitivity analyses after exclusion of 1) participants with incident stroke during follow-up (n = 220); 2) participants with incident coronary events during follow-up (n = 541); and 3) participants who were admitted to hospital for heart failure during follow-up (n = 196). Results did not materially change (Supplementary Table 4, 5 and 6).

To explore whether poor functional status at baseline might affect the longitudinal relation between resting heart rate/SDNN and risk of decline in functional status, we performed further sensitivity analyses including only participants with maximum functional status scores at baseline (n = 4343 participants with maximum ADL score, n = 4129 participants with maximum IADL score). Results did not materially change (Supplementary Table 7).

To test whether short duration of follow-up might affect the results, we repeated the longitudinal analyses including only participants who completed 36 months of follow-up (n = 4552). The longitudinal associations between resting heart rate/SDNN and risk of decline in functional status remained significant (Supplementary Table 8).

The associations of resting heart rate and SDNN with functional decline were not modified by sex, history of hypertension or vascular diseases, use of β-blockers, calcium channel blockers or statin treatment (p for interaction > 0.05, for all groups) (Supplementary Figure 1 for heart rate; data not shown for SDNN). In an extra analysis, we tested whether the observed associations were independent of baseline cognitive function as assessed by the Mini Mental State Examination. The associations did not materially change after adjustment for baseline cognitive function (data not shown). Likewise, these associations remained unchanged when we standardized SDNN for heart rate (Supplementary Table 9 and 10). Furthermore, we observed no difference in risk of functional decline between participants with a heart rate of less than 50 beats/min (n = 284) and those with a heart rate of 50–60 beats/min (n = 1365). Participants in the highest tertile had a higher risk of functional decline compared with the participants in the group with a heart rate of 50–60 beats/min (Supplementary Table 11).

INTERPRETATION

In our study, higher resting heart rate and lower heart rate variability were associated with worse functional performance at baseline and with higher risk of future functional decline in older adults at high cardiovascular risk. These associations were independent of cardiovascular risk factors, cardiovascular morbidities and use of medications.

The results of our study are in line with the results of the Prevention Regimen for Effectively Avoiding Second Stroke (PRoFESS) trial, which showed that higher heart rate was related to worse functional outcomes in patients with a recurrent stroke[15]. Our results are also consistent with findings from the Women's Health and Aging Study-I (WHAS-I), which showed a cross-sectional association between lower heart rate variability and frailty in disabled older women living in the community[16]. Our study extends the findings of WHAS-I to older adults at risk for cardiovascular disease with preserved functional status. Furthermore, we showed that the association of heart rate variability with functional decline was independent of sex.

Different pathophysiological mechanisms may underlie these associations. First, higher heart rate and lower heart rate variability have been consistently associated with incident cardiovascular events in previous studies[1-3]. In this study, the strength of the associations between heart rate/heart rate variability and functional decline did not materially change after exclusion of participants with incident cardiovascular events. This might suggest that mechanisms other than macrovascular damage play roles in the association between heart rate/heart rate variability and functional decline. Second, lower heart rate is associated with better cardiovascular fitness, which is a protective factor for brain aging and functional decline[17]. In particular, lower heart rate is related to less myocardial oxygen consumption and more prolonged time available for diastolic heart chamber filling and coronary perfusion[18]. Furthermore, higher heart rate has been suggested to increase pulsatile shear stress, which leads to endothelial dysfunction and accelerated atherosclerosis[5,19]. In this setting, use of ivabradine, a pure heart rate-lowering agent, in relation to cardiovascular outcomes has been tested with conflicting results[20-22]. Third, heart rate and heart rate variability reflect the autonomic nervous system's control over cardiac function. Cardiac autonomic control regulates the interaction between circulation and respiration. Higher heart rate variability in synchrony with respiration improves the efficiency of gas exchange at the level of the lung via efficient ventilation and

perfusion matching[23]. Furthermore, cardiac autonomic control keeps blood pressure constant within a certain range to maintain adequate perfusion to vital organs, including the brain. A preserved cardiac autonomic control buffers variations in blood pressure in response to stressors. Indeed, participants with lower heart rate variability present higher blood pressure variability in response to psychological challenge or tilt test[24,25]. Higher blood pressure variability is associated with atherosclerosis[26] and silent brain damage[27]. Finally, the autonomic nervous system is connected to regions of the central nervous system[28,29], which are involved in mood regulation. Lower heart rate variability has been associated with depression[30,31], which is a cause of disability[6].

Strengths and limitations

A strength of our study was the longitudinal design, which allowed us to show that high heart rate and low heart rate variability preceded the decline in functional status. We also showed that this association was independent of potential confounders such as vascular diseases and use of antihypertensive and cardioprotective medications. However, causality cannot be inferred given the observational nature of this study. Further strengths are the large study population of older adults and the multicentre design.

A limitation of our study was that all participants were older adults at high cardiovascular risk, which may limit the generalizability of our findings. Nevertheless, a considerable number of older adults carry high loads of cardiovascular pathologies and comorbidities. Moreover, we categorized our participants into the clinically distinguishable groups of those who declined and those who did not decline, although this categorization may result in loss of information. Another possible limitation is the use of a 10-second ECG; nonetheless, we were able to show a significant association of resting heart rate and heart rate variability with functional status even by using a short ECG recording, which is more feasible in clinical practice than longer recordings. Heart rate variability measured from standard 10-second ECG recordings correlates with heart rate variability measured from longer ECG recordings[11].

Conclusion

We found that higher resting heart rate and lower heart rate variability were associated with worse functional status in older adults, independent of cardiovascular risk factors and comorbidities. This study provides insight into the role of cardiac autonomic function in the development of functional decline. Because functional disability has a long preclinical phase, it is crucial to identify potential interventions to delay it. Further research is needed to establish whether heart rate and heart rate variability are risk markers and/or potentially modifiable risk factors for functional decline. Pharmacologic and nonpharmacologic interventions (e.g., drugs with antiadrenergic properties, physical exercise, nervus vagus stimulation) aimed at modulating cardiac autonomic function may be beneficial in preserving functional status. It is well established that physical activity is a key contributor in autonomic regulation and is linked with preservation of functional status[32,33]. However, future studies are needed to test the influence of physical activity on functionality through autonomic regulation in older adults.

REFERENCES

1. Fox K, Borer JS, Camm AJ, et al. Resting heart rate in cardiovascular disease. J Am Coll Cardiol 2007;50:823-30.
2. Heart rate variability. Standards of measurement, physiological interpretation, and clinical use. Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology. Eur Heart J 1996;17:354-81.
3. Palatini P, Benetos A, Julius S. Impact of increased heart rate on clinical outcomes in hypertension: implications for antihypertensive drug therapy. Drugs 2006;66:133-44.
4. Sajadieh A, Nielsen OW, Rasmussen V, et al. Increased heart rate and reduced heart-rate variability are associated with subclinical inflammation in middle-aged and elderly subjects with no apparent heart disease. Eur Heart J 2004;25:363-70.
5. Custodis F, Schirmer SH, Baumhakel M, et al. Vascular pathophysiology in response to increased heart rate. J Am Coll Cardiol 2010;56:1973-83.
6. Mathers C, Fat DM, Boerma J. The global burden of disease: 2004 update. Geneva: World Health Organization; 2008.

7. Chaves PH, Varadhan R, Lipsitz LA, et al. Physiological complexity underlying heart rate dynamics and frailty status in community dwelling older women. J Am Geriatr Soc 2008;56:1698-703.
8. Shepherd J, Blauw GJ, Murphy MB, et al. The design of a prospective study of Pravastatin in the Elderly at Risk (PROSPER). PROSPER Study Group. PROspective Study of Pravastatin in the Elderly at Risk. Am J Cardiol 1999;84:1192-7.
9. Shepherd J, Blauw GJ, Murphy MB, et al. Pravastatin in elderly individuals at risk of vascular disease (PROSPER): a randomised controlled trial. Lancet 2002;360:1623-30.
10. Macfarlane P, Devine B, Clark E, editors. The university of Glasgow (Uni-G) ECG analysis program. Computers in Cardiology 2005;32:451-4.
11. Hamilton RM, McKechnie PS, Macfarlane PW. Can cardiac vagal tone be estimated from the 10-second ECG? Int J Cardiol 2004;95:109-15.
12. Mahoney FI, Barthel DW. Functional evaluation: the Barthel index. Md State Med J 1965;14:61-5.
13. Lawton MP. The functional assessment of elderly people. J Am Geriatr Soc 1971;19:465-81.
14. Sacha J. Why should one normalize heart rate variability with respect to average heart rate. Front Physiol 2013;4:306.
15. Böhm M, Cotton D, Foster L, et al. Impact of resting heart rate on mortality, disability and cognitive decline in patients after ischaemic stroke. Eur Heart J 2012;33:2804-12.
16. Varadhan R, Chaves PH, Lipsitz LA, et al. Frailty and impaired cardiac autonomic control: new insights from principal components aggregation of traditional heart rate variability indices. J Gerontol A Biol Sci Med Sci 2009;64:682-7.
17. Jefferson AL, Himali JJ, Beiser AS, et al. Cardiac index is associated with brain aging: the Framingham Heart Study. Circulation 2010;122:690-7.
18. Nelson RR, Gobel FL, Jorgensen CR, et al. Hemodynamic predictors of myocardial oxygen consumption during static and dynamic exercise. Circulation 1974;50:1179-89.
19. Chatzizisis YS, Coskun AU, Jonas M, et al. Role of endothelial shear stress in the natural history of coronary atherosclerosis and vascular remodeling: molecular, cellular, and vascular behavior. J Am Coll Cardiol 2007;49:2379-93.
20. Fox K, Ford I, Steg PG, et al. Ivabradine for patients with stable coronary artery disease and left-ventricular systolic dysfunction (BEAUTIFUL): a randomised, double-blind, placebo-controlled trial. Lancet 2008;372:807-16.

21. Fox K, Ford I, Steg PG, et al. Ivabradine in stable coronary artery disease without clinical heart failure. N Engl J Med 2014; 371:1091-9.
22. Swedberg K, Komajda M, Bohm M, et al. Ivabradine and outcomes in chronic heart failure (SHIFT): a randomised placebocontrolled study. [published erratum Lancet 2010;376:1988] Lancet 2010;376:875-85.
23. Yasuma F, Hayano J. Respiratory sinus arrhythmia: Why does the heartbeat synchronize with respiratory rhythm? Chest 2004; 125:683-90.
24. Sloan RP, Demeersman RE, Shapiro PA, et al. Cardiac autonomic control is inversely related to blood pressure variability responses to psychological challenge. Am J Physiol 1997; 272:H2227-32.
25. Sloan RP, DeMeersman RE, Shapiro PA, et al. Blood pressure variability responses to tilt are buffered by cardiac autonomic control. Am J Physiol 1997;273:H1427-31.
26. Shintani Y, Kikuya M, Hara A, et al. Ambulatory blood pressure, blood pressure variability and the prevalence of carotid artery alteration: the Ohasama study. J Hypertens 2007;25:1704-10.
27. Gómez-Angelats E, de La Sierra A, Sierra C, et al. Blood pressure variability and silent cerebral damage in essential hypertension. Am J Hypertens 2004;17:696-700.
28. Castle M, Comoli E, Loewy AD. Autonomic brainstem nuclei are linked to the hippocampus. Neuroscience 2005;134:657-69.
29. Mujica-Parodi LR, Korgaonkar M, Ravindranath B, et al. Limbic dysregulation is associated with lowered heart rate variability and increased trait anxiety in healthy adults. Hum Brain Mapp 2009; 30:47-58.
30. Kemp AH, Quintana DS, Gray MA, et al. Impact of depression and antidepressant treatment on heart rate variability: a review and meta-analysis. Biol Psychiatry 2010;67:1067-74.
31. Licht CM, de Geus EJ, Zitman FG, et al. Association between major depressive disorder and heart rate variability in the Netherlands Study of Depression and Anxiety (NESDA). Arch Gen Psychiatry 2008;65:1358-67.
32. Soares-Miranda L, Sattelmair J, Chaves P, et al. Physical activity and heart rate variability in older adults: the Cardiovascular Health Study. Circulation 2014;129:2100-10.
33. Peterson MJ, Giuliani C, Morey MC, et al. Health, Aging and Body Composition Study Research Group. Physical activity as a preventative factor for frailty: the health, aging, and body composition study. J Gerontol A Biol Sci Med Sci 2009;64:61-8.

Table 1. Characteristics of the study population at baseline

Characteristic	No. (%) of participants*
Socio-demographics	
Age, years, mean ± SD	75.3 ± 3.3
Female sex	2619 (51.9)
Age left school, years, mean ± SD	15.1 ± 2.1
Cardiovascular risk factors	
History of hypertension	3127 (62.0)
History of stroke or TIA	552 (10.9)
History of MI	662 (13.1)
History of claudication	336 (6.7)
History of diabetes mellitus	517 (10.3)
Current smoking	1334 (26.5)
BMI, kg/m ^{2} , mean ± SD	26.8 ± 4.2
Medications	
Beta-blockers	1320 (26.2)
Calcium channel blockers	1275 (25.3)
Statins	2504 (49.7)

Abbreviations: HR: Heart Rate; SD: Standard Deviation; TIA: Transient Ischemic Attack; MI: Myocardial Infarction; BMI: Body Mass Index. *Unless stated otherwise.

Table 2. Baseline functional status in tertiles of resting heart rate and SDNN

	Tertiles of HR/SDNN			
	Low	Middle	High	**P for trend**
Heart Rate	n=1649	n=1742	n=1651	
HR range, beats/min	34-60	61-70	71-117	
ADL score				
Model 1	19.79 (0.02)	19.78 (0.02)	19.71 (0.02)	0.004
Model 2	19.27 (0.25)	19.26 (0.25)	19.21 (0.25)	0.02
IADL score				
Model 1	13.67 (0.03)	13.62 (0.02)	13.52 (0.03)	<0.001
Model 2	12.94 (0.34)	12.89 (0.34)	12.80 (0.33)	<0.001
SDNN	n=1689	n=1670	n=1683	
SDNN range, ms	1.70-13.30	13.40-26.50	26.60-422.60	
ADL score				
Model 1	19.73 (0.02)	19.75 (0.02)	19.80 (0.02)	0.01
Model 2	19.23 (0.25)	19.24 (0.25)	19.27 (0.25)	0.11
IADL score				
Model 1	13.55 (0.03)	13.62 (0.03)	13.65 (0.02)	0.004
Model 2	12.84 (0.33)	12.90 (0.34)	12.91 (0.34)	0.03

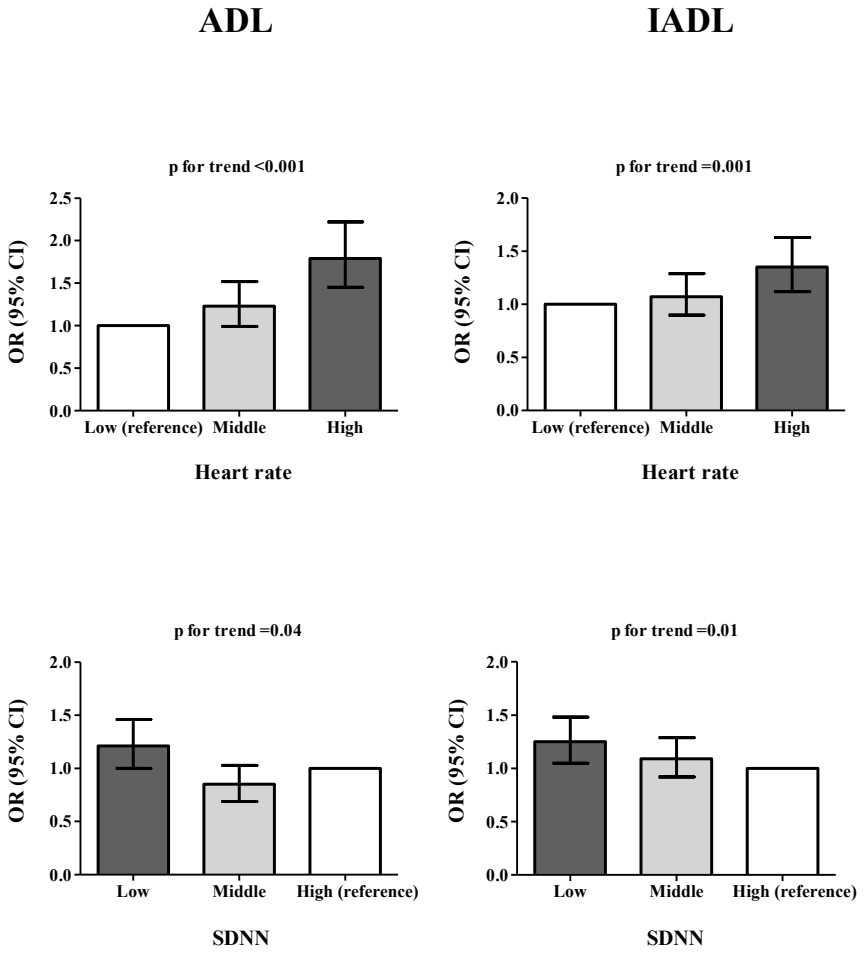
ADL and IADL scores are presented as means (standard errors). Abbreviations: HR: Heart Rate; SDNN: Standard Deviation of the Normal to Normal R-R intervals; n: Number; beats/min: beats/minute; ms: milliseconds; ADL: basic Activities of Daily Living; IADL: Instrumental Activities of Daily Living. Model 1: adjusted for country, age, sex, education. Model 2: adjusted for country, age, sex, education, smoking, body mass index, history of hypertension, history of diabetes mellitus, history of claudication, history of myocardial infarction, history of stroke/transient ischemic attack, statin treatment, diuretics, angiotensin converting enzyme inhibitors, angiotensin receptor blockers, beta-blockers, calcium-channel blockers, nitrates, aspirin and anticoagulants.

Table 3. Risk of decline in functional status in tertiles of resting heart rate and SDNN after exclusion of participants taking beta-blockers

	Tertiles of HR/SDNN			
	Low	Middle	High	**P for trend**
Heart Rate	n=863	n=1379	n=1480	
ADL, OR (95% CI)				
Model 1	1 (ref)	1.27 [0.97;1.67]	1.95 [1.50;2.53]	<0.001
Model 2	1 (ref)	1.25 [0.95;1.65]	1.86 [1.43;2.42]	<0.001
IADL, OR (95% CI)				
Model 1	1 (ref)	1.09 [0.87;1.37]	1.46 [1.17;1.81]	<0.001
Model 2	1 (ref)	1.07 [0.85;1.35]	1.39 [1.11;1.74]	0.002
SDNN	n=1312	n=1192	n=1218	
ADL, OR (95% CI)				
Model 1	1.31 [1.06;1.63]	0.85 [0.67;1.08]	1 (ref)	0.009
Model 2	1.25 [1.00;1.55]	0.82 [0.65;1.04]	1 (ref)	0.03
IADL, OR (95% CI)				
Model 1	1.30 [1.07;1.58]	1.09 [0.89;1.34]	1 (ref)	0.008
Model 2	1.26 [1.03;1.53]	1.07 [0.87;1.31]	1 (ref)	0.02

Abbreviations: HR: Heart Rate; SDNN: Standard Deviation of the Normal to Normal R-R intervals; n: Number; msec: milliseconds; ADL: basic Activities of Daily Living; IADL: Instrumental Activities of Daily Living; OR: Odds Ratio; CI: Confidence Interval. Model 1: adjusted for country, age, sex, education. Model 2: adjusted for country, age, sex, education, ADL/IADL at baseline, smoking, body mass index, history of hypertension, history of diabetes mellitus, history of claudication, history of myocardial infarction, history of stroke/transient ischemic attack, statin treatment, diuretics, angiotensin converting enzyme inhibitors, angiotensin receptor blockers, calcium-channel blockers, nitrates, aspirin and anticoagulants.

Figure 1. Risk of decline in functional status in tertiles of resting heart rate and SDNN



Bars represent odds ratios with 95% confidence intervals. All analyses are adjusted for country, age, sex, education, ADL/IADL at baseline, smoking, body mass index, history of hypertension, history of diabetes mellitus, history of claudication, history of myocardial infarction, history of stroke/transient ischemic attack, statin treatment, diuretics, angiotensin converting enzyme inhibitors, angiotensin receptor blockers, beta-blockers, calcium-channel blockers, nitrates, aspirin and anticoagulants. Range of heart rate (number of participants) in heart rate tertiles: low 34-60 beats/minute (n=1649); middle 61-70 beats/minute (n=1742); high 71-117 beats/minute (n=1651). Range of SDNN (number of participants) in SDNN tertiles: low 1.70-13.30 msec (n=1689); middle 13.40-26.50 msec (n=1670); high 26.60-422.60 msec (n=1683). Abbreviations: ADL: basic Activities of Daily Living; IADL: Instrumental Activities of Daily Living; OR: Odds Ratios; CI: Confidence Interval; SDNN: Standard Deviation of the Normal to Normal R-R intervals.

Supplementary Table 1. Characteristics of study population in tertiles of resting heart rate

Characteristics	Tertiles of HR (beats/minute)			p-value
	Low (34-60) n=1649	Middle (61-70) n=1742	High (71-117) n=1651	
Heart rate, beats/minute, mean (SD)	54.3 (4.7)	65.3 (2.8)	79.3 (7.6)	
Socio-demographics				
Age, years, mean (SD)	75.1 (3.3)	75.3 (3.3)	75.4 (3.4)	0.01
Female, n (%)	706 (42.8)	928 (53.3)	985 (59.7)	<0.001
Age left school, years, mean (SD)	15.2 (2.1)	15.2 (2.1)	15.1 (1.9)	0.37
Cardiovascular risk factors				
History of hypertension, n (%)	1046 (63.4)	1056 (60.6)	1025 (62.1)	0.24
History of stroke or TIA, n (%)	184 (11.2)	187 (10.7)	181 (11.0)	0.93
History of MI, n (%)	273 (16.6)	189 (10.8)	200 (12.1)	<0.001
History of claudication, n (%)	94 (5.7)	124 (7.1)	118 (7.1)	0.16
History of diabetes mellitus, n (%)	115 (7.0)	180 (10.3)	222 (13.4)	<0.001
Current smoking, n (%)	361 (21.9)	499 (28.6)	474 (28.7)	<0.001
BMI, kg/m^2, mean (SD)	26.8 (3.9)	26.7 (4.1)	27.1 (4.5)	0.02
Medications				
Beta-blockers, n (%)	786 (47.7)	363 (20.8)	171 (10.4)	<0.001
Calcium channel blockers, n (%)	432 (26.2)	415 (23.8)	428 (25.9)	0.22
Statins, n (%)	843 (51.1)	867 (49.8)	794 (48.1)	0.22

Abbreviations: HR: Heart Rate; SD: Standard Deviation; n: Number; TIA: Transient Ischemic Attack; MI: Myocardial Infarction; BMI: Body Mass Index.

Supplementary Table 2. Characteristics of study population in tertiles of SDNN

Characteristics	Tertiles of SDNN (msec)			p-value
	Low (1.70-13.30) n=1689	Medium (13.40-26.50) n=1670	High (26.60-422.60) n=1683	
Socio-demographics				
Age, years, mean (SD)	75.36 (3.39)	75.12 (3.30)	75.32 (3.32)	0.08
Female, n (%)	879 (52.0)	907 (54.3)	833 (49.5)	0.02
Age left school, years, mean (SD)	15.16 (2.00)	15.17 (2.08)	15.10 (2.08)	0.59
Cardiovascular factors:				
History of hypertension, n (%)	1079 (63.9)	1028 (61.6)	1020 (60.6)	0.13
History of stroke or TIA, n (%)	202 (12.0)	188 (11.3)	162 (9.6)	0.08
History of MI, n (%)	220 (13.0)	212 (12.7)	230 (13.7)	0.61
History of claudication, n (%)	129 (7.4)	113 (6.8)	94 (5.6)	0.06
History of diabetes mellitus, n (%)	210 (12.4)	160 (9.2)	147 (8.7)	0.001
Current smoking, n (%)	423 (25.0)	452 (27.1)	459 (27.3)	0.27
BMI, kg/m ^{2} mean (SD)	27.12 (4.25)	26.78 (4.13)	26.64 (4.12)	0.003
Medications:				
Beta-blockers, n (%)	377 (22.3)	478 (28.6)	465 (27.6)	<0.001
Calcium channel blockers, n (%)	424 (25.1)	424 (25.4)	427 (25.4)	0.98
Statins, n (%)	846 (50.1)	811 (48.6)	847 (50.3)	0.54

Abbreviations: SD: standard deviation; n: Number; TIA: Transient Ischemic Attack; MI: Myocardial Infarction; BMI: Body Mass Index.

Supplementary Table 3. Risk of decline in functional status in tertiles of resting heart rate and SDNN in the minimally adjusted models

	Tertiles of HR/SDNN			
	Low	Middle	High	**P for trend**
Heart Rate	n=1649	n=1742	n=1651	
HR range, beats/min	34-60	61-70	71-117	
ADL, OR (95% CI)				
Model 1	1 (ref)	1.16 [0.95;1.42]	1.70 [1.40;2.06]	<0.001
IADL, OR (95% CI)				
Model 1	1 (ref)	1.06 [0.90;1.27]	1.37 [1.15;1.63]	<0.001
SDNN	n=1689	n=1670	n=1683	
SDNN range, ms	1.70-13.30	13.40-26.50	26.60-422.60	
ADL, OR (95% CI)				
Model 1	1.28 [1.06;1.54]	0.88 [0.72;1.07]	1 (ref)	0.008
IADL, OR (95% CI)				
Model 1	1.30 [1.10;1.53]	1.11 [0.94;1.32]	1 (ref)	0.003

Abbreviations: HR: Heart Rate; SDNN: Standard Deviation of the Normal to Normal R-R intervals; n: Number; msec: milliseconds; ADL: basic Activities of Daily Living; IADL: Instrumental Activities of Daily Living; OR: Odds Ratio; CI: Confidence Interval. Model 1: adjusted for country, age, sex and education.

Supplementary Table 4. Risk of decline in functional status in tertiles of resting heart rate and SDNN after exclusion of participants with incident non-fatal stroke during follow-up

	Tertiles of HR/SDNN			
	Low	Middle	High	**P for trend**
Heart Rate	n=1580	n=1685	n=1577	
ADL, OR (95% CI)				
Model 1	1 (ref)	1.16 [0.94;1.44]	1.74 [1.41;2.13]	<0.001
Model 2	1 (ref)	1.24 [0.99;1.55]	1.84 [1.47;2.30]	<0.001
IADL, OR (95% CI)				
Model 1	1 (ref)	1.08 [0.90;1.29]	1.41 [1.17;1.68]	<0.001
Model 2	1 (ref)	1.07 [0.89;1.30]	1.37 [1.12;1.66]	0.001
SDNN	n=1626	n=1603	n=1613	
ADL, OR (95% CI)				
Model 1	1.34 [1.11;1.63]	0.92 [0.75;1.13]	1 (ref)	0.002
Model 2	1.27 [1.04;1.55]	0.88 [0.72;1.09]	1 (ref)	0.02
IADL, OR (95% CI)				
Model 1	1.35 [1.13;1.61]	1.14 [0.95;1.36]	1 (ref)	0.001
Model 2	1.29 [1.08;1.54]	1.11 [0.93;1.33]	1 (ref)	0.005

Abbreviations: HR: Heart Rate; SDNN: Standard Deviation of the Normal to Normal R-R intervals; n: Number; msec: milliseconds; ADL: basic Activities of Daily Living; IADL: Instrumental Activities of Daily Living; OR: Odds Ratio; CI: Confidence Interval. Model 1: adjusted for country, age, sex, education. Model 2: adjusted for country, age, sex, education, ADL/IADL at baseline, smoking, body mass index, history of hypertension, history of diabetes mellitus, history of claudication, history of myocardial infarction, history of stroke/transient ischemic attack, statin treatment, diuretics, angiotensin converting enzyme inhibitors, angiotensin receptor blockers, beta-blockers, calcium-channel blockers, nitrates, aspirin and anticoagulants.

Supplementary Table 5. Risk of decline in functional status in tertiles of resting heart rate and SDNN after exclusion of participants with incident coronary events during follow-up

	Tertiles of HR/SDNN			
	Low	Middle	High	**P for trend**
Heart Rate	n=1459	n=1564	n=1478	
ADL, OR (95% CI)				
Model 1	1 (ref)	1.06 [0.85;1.31]	1.58 [1.28;1.95]	<0.001
Model 2	1 (ref)	1.13 [0.90;1.42]	1.69 [1.34;2.12]	<0.001
IADL, OR (95% CI)				
Model 1	1 (ref)	0.97 [0.81;1.17]	1.31 [1.09;1.57]	0.003
Model 2	1 (ref)	0.97 [0.80;1.18]	1.27 [1.04;1.55]	0.02
SDNN	n=1489	n=1507	n=1505	
ADL, OR (95% CI)				
Model 1	1.33 [1.09;1.63]	0.89 [0.71;1.08]	1 (ref)	0.004
Model 2	1.27 [1.04;1.55]	0.85 [0.68;1.05]	1 (ref)	0.02
IADL, OR (95% CI)				
Model 1	1.31 [1.10;1.57]	1.10 [0.91;1.32]	1 (ref)	0.003
Model 2	1.27 [1.06;1.53]	1.08 [0.90;1.30]	1 (ref)	0.01

Abbreviations: HR: Heart Rate; SDNN: Standard Deviation of the Normal to Normal R-R intervals; n: Number; msec: milliseconds; ADL: basic Activities of Daily Living; IADL: Instrumental Activities of Daily Living; OR: Odds Ratio; CI: Confidence Interval. Model 1: adjusted for country, age, sex, education. Model 2: adjusted for country, age, sex, education, ADL/IADL at baseline, smoking, body mass index, history of hypertension, history of diabetes mellitus, history of claudication, history of myocardial infarction, history of stroke/transient ischemic attack, statin treatment, diuretics, angiotensin converting enzyme inhibitors, angiotensin receptor blockers, beta-blockers, calcium-channel blockers, nitrates, aspirin and anticoagulants.

Supplementary Table 6. Risk of decline in functional status in tertiles of resting heart rate and SDNN after exclusion of participants with incident heart failure hospitalization during follow-up

	Tertiles of HR/SDNN			
	Low	Middle	High	**P for trend**
Heart Rate	n=1599	n=1672	n=1575	
ADL, OR (95% CI)				
Model 1	1 (ref)	1.12 [0.91;1.37]	1.63 [1.33;1.99]	<0.001
Model 2	1 (ref)	1.18 [0.95;1.47]	1.72 [1.38;2.14]	<0.001
IADL, OR (95% CI)				
Model 1	1 (ref)	1.00 [0.84;1.19]	1.30 [1.09;1.55]	0.003
Model 2	1 (ref)	1.00 [0.83;1.20]	1.27 [1.05;1.54]	0.01
SDNN	n=1606	n=1620	n=1620	
ADL, OR (95% CI)				
Model 1	1.33 [1.10;1.61]	0.89 [0.73;1.10]	1 (ref)	0.003
Model 2	1.27 [1.04;1.54]	0.87 [0.70;1.06]	1 (ref)	0.02
IADL, OR (95% CI)				
Model 1	1.30 [1.09;1.55]	1.15 [0.96;1.37]	1 (ref)	0.003
Model 2	1.25 [1.05;1.50]	1.13 [0.94;1.35]	1 (ref)	0.01

Abbreviations: HR: Heart Rate; SDNN: Standard Deviation of the Normal to Normal R-R intervals; n: Number; msec: milliseconds; ADL: basic Activities of Daily Living; IADL: Instrumental Activities of Daily Living; OR: Odds Ratio; CI: Confidence Interval. Model 1: adjusted for country, age, sex, education. Model 2: adjusted for country, age, sex, education, ADL/IADL at baseline, smoking, body mass index, history of hypertension, history of diabetes mellitus, history of claudication, history of myocardial infarction, history of stroke/transient ischemic attack, statin treatment, diuretics, angiotensin converting enzyme inhibitors, angiotensin receptor blockers, beta-blockers, calcium-channel blockers, nitrates, aspirin and anticoagulants.

Supplementary Table 7. Risk of decline in functional status in tertiles of resting heart rate and SDNN after inclusion of participants with maximum functional status at baseline

	Tertiles of HR/SDNN			
	Low	Middle	High	**P for trend**
Heart Rate				
ADL, OR (95% CI)	n=1465	n=1491	n=1387	
Model 1	1 (ref)	1.25 [1.00;1.57]	1.75 [1.41;2.18]	<0.001
Model 2	1 (ref)	1.34 [1.06;1.70]	1.88 [1.48;2.40]	<0.001
IADL, OR (95% CI)	n=1397	n=1433	n=1299	
Model 1	1 (ref)	1.14 [0.94;1.39]	1.35 [1.10;1.65]	0.003
Model 2	1 (ref)	1.19 [0.97;1.46]	1.42 [1.14;1.76]	0.002
SDNN				
ADL, OR (95% CI)	n=1441	n= 1428	n= 1474	
Model 1	1.19 [0.97;1.46]	0.78 [0.63;0.98]	1 (ref)	0.09
Model 2	1.13 [0.92;1.40]	0.77 [0.61;0.96]	1 (ref)	0.23
IADL, OR (95% CI)	n=1350	n=1383	n=1396	
Model 1	1.29 [1.06;1.56]	1.12 [0.92;1.36]	1 (ref)	0.01
Model 2	1.26 [1.03;1.53]	1.10 [0.90;1.34]	1 (ref)	0.02

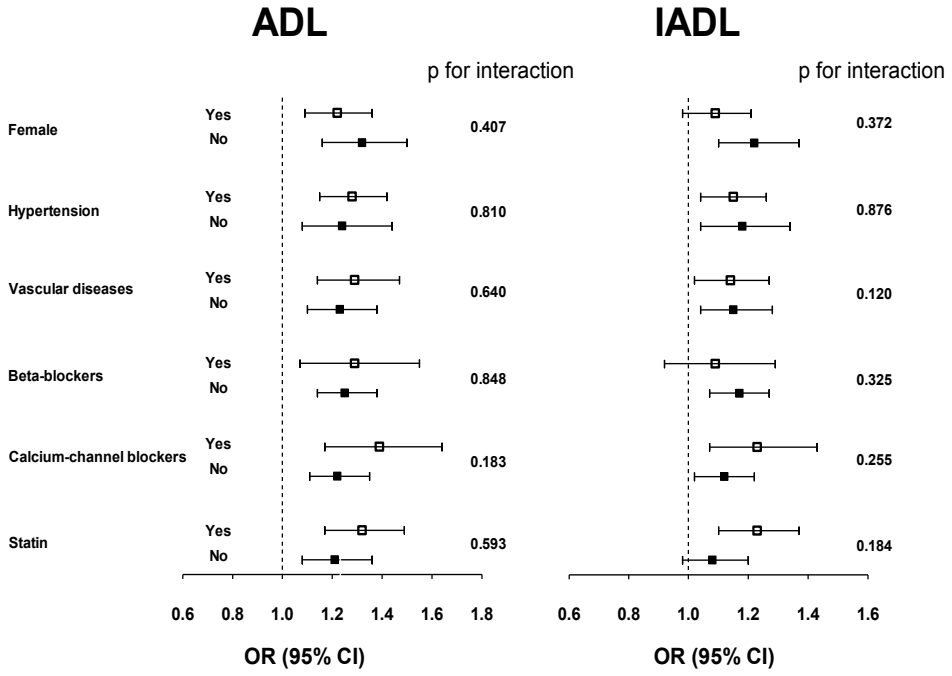
Abbreviations: HR: Heart Rate; SDNN: Standard Deviation of the Normal to Normal R-R intervals; n: Number; msec: milliseconds; ADL: basic Activities of Daily Living; IADL: Instrumental Activities of Daily Living; OR: Odds Ratio; CI: Confidence Interval. Model 1: adjusted for country, age, sex, education. Model 2: adjusted for country, age, sex, education, smoking, body mass index, history of hypertension, history of diabetes mellitus, history of claudication, history of myocardial infarction, history of stroke/transient ischemic attack, statin treatment, diuretics, angiotensin converting enzyme inhibitors, angiotensin receptor blockers, beta-blockers, calcium-channel blockers, nitrates, aspirin and anticoagulants.

Supplementary Table 8. Risk of decline in functional status in tertiles of resting heart rate and SDNN after inclusion of participants who completed 36 months of follow-up

	Tertiles of HR/SDNN			
	Low	Middle	High	**P for trend**
Heart Rate	n=1513	n=1576	n=1463	
ADL, OR (95% CI)				
Model 1	1 (ref)	1.15 [0.93;1.43]	1.72 [1.40;2.12]	<0.001
Model 2	1 (ref)	1.23 [0.98;1.54]	1.86 [1.48;2.33]	<0.001
IADL, OR (95% CI)				
Model 1	1 (ref)	1.04 [0.86;1.25]	1.30 [1.08;1.56]	0.005
Model 2	1 (ref)	1.06 [0.87;1.28]	1.30 [1.06;1.59]	0.009
SDNN	n=1491	n=1530	n=1531	
ADL, OR (95% CI)				
Model 1	1.30 [1.07;1.58]	0.91 [0.74;1.12]	1 (ref)	0.008
Model 2	1.23 [1.01;1.50]	0.87 [0.71;1.08]	1 (ref)	0.04
IADL, OR (95% CI)				
Model 1	1.34 [1.12;1.61]	1.11 [0.93;1.34]	1 (ref)	0.001
Model 2	1.28 [1.07;1.54]	1.08 [0.90;1.30]	1 (ref)	0.008

Abbreviations: HR: Heart Rate; SDNN: Standard Deviation of the Normal to Normal R-R intervals; n: Number; msec: milliseconds; ADL: basic Activities of Daily Living; IADL: Instrumental Activities of Daily Living; OR: Odds Ratio; CI: Confidence Interval. Model 1: adjusted for country, age, sex, education. Model 2: adjusted for country, age, sex, education, ADL/IADL at baseline, smoking, body mass index, history of hypertension, history of diabetes mellitus, history of claudication, history of myocardial infarction, history of stroke/transient ischemic attack, statin treatment, diuretics, angiotensin converting enzyme inhibitors, angiotensin receptor blockers, beta-blockers, calcium-channel blockers, nitrates, aspirin and anticoagulants.

Supplementary Figure 1. Risk of functional decline in relation to heart rate in stratified analyses



Bars represent odds ratios (95% confidence interval) per each standard deviation increase in resting heart rate in stratified analyses. Adjusted for country, age, sex, education, ADL/IADL at baseline, smoking, body mass index, history of hypertension, history of diabetes mellitus, history of claudication, history of myocardial infarction, history of stroke/transient ischemic attack, statin treatment, diuretics, angiotensin converting enzyme inhibitors, angiotensin receptor blockers, calcium-channel blockers, nitrates, aspirin and anticoagulants. P- values show p for interaction. Abbreviations: OR: odds ratio; CI: confidence interval; ADL: activities of daily living; IADL: instrumental activities of daily living.

Supplementary Table 9. Baseline functional status in tertiles of standardized SDNN

	Tertiles of standardized SDNN			
	Low n=1680	Middle n=1681	High n=1681	**P for trend**
SDNN range, ms	0.02-0.20	0.20-0.43	0.43-7.85	
ADL score				
Model 1	19.73 (0.02)	19.77 (0.02)	19.79 (0.02)	0.021
Model 2	19.22 (0.25)	19.25 (0.25)	19.26 (0.25)	0.180
IADL score				
Model 1	13.54 (0.03)	13.60 (0.02)	13.66 (0.02)	0.001
Model 2	12.83 (0.33)	12.88 (0.34)	12.93 (0.34)	0.008

ADL and IADL scores are presented as means (standard errors). Abbreviations: SDNN: Standard Deviation of the Normal to Normal R-R intervals; n: number; ms: milliseconds; ADL: basic Activities of Daily Living; IADL: Instrumental Activities of Daily Living. Model 1: adjusted for country, age, sex, education. Model 2: adjusted for country, age, sex, education, smoking, body mass index, history of hypertension, history of diabetes mellitus, history of claudication, history of myocardial infarction, history of stroke/transient ischemic attack, statin treatment, diuretics, Angiotensin Converting Enzyme inhibitors, angiotensin receptor blockers, beta-blockers, calcium-channel blockers, nitrates, aspirin, anticoagulants.

Supplementary Table 10. Risk of decline in functional status in tertiles of standardized SDNN

	Tertiles of standardized SDNN			
	Low n=1680	Middle n=1681	High n=1681	**P for trend**
SDNN range, ms	0.02-0.20	0.20-0.43	0.43-7.85	
ADL, OR (95% CI)				
Model 1	1.32 [1.10;1.59]	0.98 [0.80;1.19]	1 (ref)	0.003
Model 2	1.26 [1.04;1.53]	0.94 [0.77;1.15]	1 (ref)	0.015
IADL, OR (95% CI)				
Model 1	1.31 [1.11;1.55]	1.17 [0.99;1.39]	1 (ref)	0.002
Model 2	1.26 [1.06;1.49]	1.14 [0.96;1.35]	1 (ref)	0.010

Abbreviations: SDNN: Standard Deviation of the Normal to Normal R-R intervals; n: number; ms: milliseconds; ADL: basic Activities of Daily Living; IADL: Instrumental Activities of Daily Living; OR: Odds Ratio; CI: Confidence Interval. Model 1: adjusted for country, age, sex, education. Model 2: adjusted for country, age, sex, education, ADL/IADL at baseline, smoking, body mass index, history of hypertension, history of diabetes mellitus, history of claudication, history of myocardial infarction, history of stroke/transient ischemic attack, statin treatment, diuretics, Angiotensin Converting Enzyme inhibitors, angiotensin receptor blockers, beta-blockers, calcium-channel blockers, nitrates, aspirin, anticoagulants.

Supplementary Table 11. Risk of decline in functional status in groups of resting heart rate

	Groups of HR (beats/minute)				
	< 50 n = 284	50-60 n = 1365	61-70 n = 1742	71-117 n = 1651	**P for trend**
ADL, OR (95% CI)	0.76 [0.49;1.19]	1 (ref)	1.19 [0.95;1.48]	1.73 [1.39;2.15]	0.002
IADL, OR (95% CI)	1.32 [0.95;1.84]	1 (ref)	1.12 [0.93;1.36]	1.41 [1.16;1.72]	<0.001

Abbreviations: HR: Heart Rate; n: Number; ADL: basic Activities of Daily Living; IADL: Instrumental Activities of Daily Living; OR: Odds Ratio; CI: Confidence Interval. Adjusted for country, age, sex, education, ADL/IADL at baseline, smoking, body mass index, history of hypertension, history of diabetes mellitus, history of claudication, history of myocardial infarction, history of stroke/transient ischemic attack, statin treatment, diuretics, angiotensin converting enzyme inhibitors, angiotensin receptor blockers, calcium-channel blockers, nitrates, aspirin and anticoagulants.

Chapter 6

Blood pressure variability and functional decline

Manuscript based on this chapter has been submitted as:

Ogliari G, Smit RA, Westendorp RG, Jukema JW, de Craen AJ, Sabayan B. Visit-to-visit blood pressure variability and future functional decline in old age.

ABSTRACT

Background: Higher blood pressure variability (BPV), independent of mean blood pressure, has been associated with adverse health outcomes.

Objective: To determine the association between visit-to-visit BPV and functional decline in older adults at high cardiovascular risk.

Design: Prospective cohort study.

Setting: PROSPER (PROspective Study of Pravastatin in the Elderly at Risk) study.

Participants: 4745 participants with mean age of 75.2 years and high cardiovascular risk were followed for a mean of 3.2 years. Blood pressure was measured every three months during the first 18 months. BPV was defined as the intra-individual standard deviation (SD) of measurements across these visits.

Measurements: Functional status in basic and instrumental activities of daily living was measured using the Barthel (ADL) and Lawton (IADL) scales, at 18 months and during follow-up. Functional decline between 18 months and the end of follow-up was calculated.

Results: BPV was not associated with functional status at 18 months. Higher systolic BPV was associated with steeper functional decline, whereas diastolic BPV was not. Each 10 mmHg higher SD of SBP was associated with a 0.064 (95% CI 0.016-0.112, p = 0.009) annual decline in ADL score and with a 0.078 decline (95% CI 0.020-0.136, p = 0.008) in IADL score. These associations were not modified by sex, hypertension or use of antihypertensives. These findings were independent of mean blood pressure, cardiovascular risk factors and morbidities and cognition.

Conclusion: Higher visit-to-visit systolic but not diastolic BPV was associated with steeper functional decline in older adults at high cardiovascular risk. Higher systolic BPV is a novel risk factor for functional decline.

INTRODUCTION

Visit-to-visit blood pressure variability (BPV) is the intra-individual variation in blood pressure measures over different clinical visits[1]. BPV is a reproducible phenomenon, with a possible genetic basis, and may be increased or decreased by antihypertensive drugs[2-4]. Higher visit-to-visit BPV, independent of mean blood pressure, has been associated with clinical events, such as stroke, myocardial infarction, cardiovascular and all-cause mortality[5,6]. Furthermore, it has been associated with subclinical end-organ damage, including cerebral small vessel disease, cardiac diastolic dysfunction, micro- and macro-albuminuria[7,8]. All these associations may be mediated by vascular damage, as suggested by the link of BPV with endothelial injury, arterial stiffness and atherosclerosis[7-10].

Vascular damage is a determinant of functional decline in older adults[11,12]. With the rapid ageing of populations worldwide, it is crucial to identify potential etiologic factors underlying vascular damage and functional decline. Although evidence shows the association between BPV and vascular damage, the relationship between BPV and functional decline has not been explored.

Therefore, we investigated whether visit-to-visit BPV is associated with functional decline in older adults at high risk of cardiovascular disease, independent of mean blood pressure, cardiovascular risk factors, and co-morbidities.

METHODS

Study design and participants

The data in this study were obtained from the PROspective Study of Pravastatin in the Elderly at Risk (PROSPER), a randomized, double-blind, placebo-controlled trial on the effect of pravastatin on subsequent risk of vascular events in men and women aged 70-82 years with pre-existing vascular disease or risk factors thereof. We recruited 5804 participants from three centres in Ireland, Scotland and the Netherlands. Details of study design have been previously reported on[13,14]. Physical inability to attend clinic visits and impaired cognition at baseline (Mini Mental State Examination (MMSE) < 24 points) were exclusion criteria[15]. Blood

pressure levels at baseline were no exclusion criteria. Participants were followed up for a mean of 3.2 years.

From the original study population, we first excluded 820 participants with missing data on BPV, covariates or functional decline and subsequently excluded 239 participants in whom cardiovascular events (stroke, myocardial infarction and hospitalization for heart failure) occurred in the period of assessment of BPV, thus potentially affecting the variability. We included participants from both the pravastatin and the placebo arm as we previously showed that pravastatin did not affect functional status during follow-up[14]. Hence, we included 4745 participants in the present study. The trial was approved by the Medical Ethics Committees of the three centres. All participants provided written informed consent.

Blood pressure measurements

We measured systolic (SBP) and diastolic (DBP) blood pressure at rest, in the sitting position, using a fully automatic electronic sphygmomanometer (Omron M4®). In our estimate of BPV, we took into account blood pressure measurements at baseline and at 3, 6, 9, 12, 15 and 18 months. First, we calculated the intra-individual mean SBP and DBP across these measurements. Subsequently, we computed the intra-individual standard deviation (SD) and the coefficient of variation (CV; CV= intra-individual SD / intra-individual mean x 100) of SBP and DBP as indices of BPV.

Functional status

We assessed functional status using two questionnaires: the Barthel Index for basic Activities of Daily Living (ADL)[16] and Lawton's Instrumental Activities of Daily Living (IADL)[17]. ADL assesses 10 basic items: faecal continence, urinary continence, grooming, toilet use, feeding, transfers (for example, from chair to bed), walking, dressing, climbing stairs and bathing. IADL evaluates 7 more complex items: doing housework, taking medications, managing money, shopping, using a telephone, using technology and transportation within the community. ADL and IADL scores range from 0 to 20 and from 0 to 14, respectively, with higher scores indicating more preserved functional status.

We measured functional status at 18 months, after the assessment of BPV, and subsequently at 30, 36, 39, 42, 45 or 48 months. Functional decline in ADL and IADL was defined as the difference between the last measured score and the score at 18 months, divided by the time interval.

Demographic and clinical characteristics

At baseline, a detailed medical history was taken and blood tests performed. Education was defined as age at leaving school. Smoking was dichotomized as a current smoker versus a non-current smoker. Body mass index (BMI) was calculated as weight divided by squared height (Kg/m^2). Data on history of vascular diseases were provided by each participant's general practitioner. Diabetes mellitus was defined as self-reported diagnosis, use of anti-diabetic drug or as fasting blood glucose of ≥ 7 mmol/L. Glomerular filtration rate (GFR), which is an index of kidney function, was calculated using the Modification of Diet in Renal Disease Study Group formula[18].

Statistical analysis

We used IBM SPSS Statistics (version 20) for all analyses. Baseline characteristics of participants were reported as number (percentage) for categorical variables and as mean (standard deviation) for continuous variables. We assessed differences in baseline characteristics across tertiles of BPV using Pearson's chi-squared test for categorical variables and ANOVA for continuous variables.

We analysed the associations of systolic and diastolic BPV with functional status at 18 months, and with functional decline during follow-up, using linear regression models. All analyses were performed in three steps. First, we adjusted for age, sex, country of enrolment and education (Model 1). Second, we further adjusted for cardiovascular risk factors (smoking, BMI, hypertension, diabetes mellitus), cardiovascular morbidities (myocardial infarction, stroke / transient ischemic attack, claudication, GFR), use of antihypertensives (diuretics, angiotensin converting enzyme inhibitors (ACE-inhibitors), angiotensin receptor blockers, beta-blockers, calcium-channel blockers), statin treatment group, intra-individual mean SBP or DBP and

number of measurements of SBP or DBP and, in the longitudinal associations, for ADL or IADL scores at 18 months (Model 2). Finally, we additionally adjusted for baseline MMSE scores (Model 3). We calculated adjusted means of decline in ADL and IADL scores across tertiles of BPV.

Furthermore, we repeated all analyses after stratifying the participants by 1) sex, 2) history of hypertension and 3) use of antihypertensives or pravastatin, in order to explore the influence of these parameters on the relationship between BPV and functional status. Interaction terms were computed by multiplying measures of BPV by these parameters.

In addition, we performed a series of sensitivity analyses. First, we restricted the analyses to participants with maximum functional status at 18 months, to assess whether poor functional status may affect subsequent decline. Second, we restricted the analyses to participants with maximum MMSE score. Third, we repeated the analyses in participants who did not experience major incident cardiovascular events (stroke and / or myocardial infarction) during follow-up, to test whether the relationship between BPV and functional decline is independent of these incident events.

RESULTS

In the period between baseline and 18 months, the intra-individual mean SBP and DBP were 153.7 and 83.4 mmHg and the intra-individual mean SD of SBP and DBP were 16.7 and 7.8 mmHg, respectively. Table 1 shows the characteristics of participants in the whole study population and in tertiles of systolic BPV. Subjects with higher systolic BPV were older and more likely to be females, to have a history of hypertension, to be treated with beta-blockers and ACE-inhibitors at baseline, and to have lower MMSE scores and higher mean SBP (all p-values<0.05). Similarly, subjects with higher diastolic BPV were older, more frequently had a history of hypertension, were on treatment with diuretics or ACE-inhibitors and had lower MMSE scores (all p-values<0.05, data not shown).

Neither systolic nor diastolic BPV was associated with functional status at 18 months (data not shown). Figure 1 presents the longitudinal associations of systolic and diastolic BPV with functional decline. Higher systolic BPV was associated with a steeper decline in both ADL and IADL scores. After full adjustment, each 10 mmHg higher systolic BPV, as defined by SD,

was associated with a 0.064 (95% confidence interval (CI) 0.016-0.112) annual decline in ADL score and with a 0.078 decline (95% CI 0.020-0.136) in IADL score (p-values 0.009 and 0.008, respectively). In contrast, diastolic BPV, as defined by SD, was not associated with functional decline (all p-values>0.05, for all models). Likewise, higher CV of SBP was associated with a steeper functional decline (all p-values<0.05), while no association was observed between CV of DBP and functional decline (all p-values>0.05, data not shown).

The longitudinal associations of systolic and diastolic BPV with functional decline were not modified by 1) sex, 2) history of hypertension or 3) use of antihypertensives or pravastatin (all p-values for interaction > 0.05, Figure 2 for systolic BPV, data not shown for diastolic BPV).

The relationship between higher systolic BPV and steeper functional decline remained statistically significant when restricting the analyses to participants with maximum functional status at 18 months (Supplementary Table 1). Likewise, it remained unchanged when restricting the analyses to those with maximum MMSE score (Supplementary Table 2).

The association between higher systolic BPV and steeper functional decline tended to remain essentially similar in analyses restricted to participants who did not experience cardiovascular events during follow-up (data not shown).

DISCUSSION

In older adults at high cardiovascular risk, higher systolic BPV was associated with steeper functional decline, independent of intra-individual mean blood pressure, cardiovascular risk factors and morbidities, cognition and use of antihypertensives.

Consistent with earlier reports, we found that higher BPV was associated with older age, female sex, use of ACE-inhibitors, and poor cognition[19,20]. Our novel finding is the association between systolic BPV and functional decline, independent of cognition. The lack of association between diastolic BPV and clinical outcomes in our study is in line with previous research, showing no association of diastolic BPV with subclinical brain damage, cognition and mortality[19, 21-24]. This may result from a lower magnitude of diastolic compared to systolic BPV.

Different interpretations may explain our findings. First, both higher BPV and functional decline may share common causes such as atherosclerosis. Atherosclerosis may lead to higher BPV through increased arterial stiffness and reduced damping of blood pressure wave within vessels[10]. Moreover, the link between atherosclerosis and functional decline is well established[11,12]. However, adjustment for history of cardiovascular morbidities, which can be seen as a marker of atherosclerotic burden, did not change our findings. Another shared cause may be neurodegenerative diseases. Neurodegenerative diseases, including Alzheimer's, have been linked to central autonomic dysregulation, in particular impaired baroreflex function[25]. Impaired baroreflex function could explain higher BPV and is present in the early stages of Alzheimer's, when cognition is quite preserved[25]. Furthermore, it is debated whether neurodegenerative diseases may be linked to unbalanced blood pressure regulation via impaired parasympathetic cholinergic neurotransmission[25-27]. However, our results remained unchanged after adjusting for cognition and after restricting the analyses to participants with maximum MMSE score.

Alternatively, higher BPV may contribute to functional decline by causing oscillations in cerebral perfusion[28]. Inadequate cerebral perfusion could lead to neuronal injury and degeneration[29]. Indeed, hypotension has been linked to lesions in brain frontal-subcortical circuits involved in motor and bladder control[30]. Cerebral white matter is particularly vulnerable to abrupt drops in blood pressure and transient ischemia[31]. Of note, higher systolic but not diastolic BPV has been linked to cerebral white matter lesions, which, in turn, have been associated with gait and balance instability, falls, executive dysfunction and depression in older adults[22-24,32,33]. Furthermore, recent studies suggest that inadequate cerebral perfusion may reduce the paravascular clearance of amyloid-β_{42} from the brain, thus favouring the development of neurodegenerative diseases[34,35].

Higher BPV may be linked to functional decline through incident major cardiovascular events. A trend of association between higher systolic BPV and steeper functional decline was observed also in participants who did not experience incident major vascular events, thus suggesting that subclinical vascular damage may also play a role. In particular, higher BPV can potentially lead to subclinical damage of crucial organs such as the heart and the kidneys, by inducing frequent episodes of hypo- and hypertension. In turn, subclinical cardiac and kidney damage may lead to functional decline in older adults.

Observational studies have provided evidence for the association of higher BPV with subclinical cardiac and kidney damage. A correlation between higher BPV and subclinical left ventricular diastolic dysfunction, but not hypertrophy, has been found[36,37]; in turn, left ventricular diastolic dysfunction predicts reduced likelihood of independent living in older adults[38]. Furthermore, higher BPV has been longitudinally associated with deterioration of kidney function[39-41]. Diminished kidney function, even within the normal range, has been associated with muscle atrophy, reduced walking speed, and steeper decline in lower-extremity strength in older adults[42]. Nonetheless, evidence from observational studies cannot prove a causal relationship between BPV and subclinical cardiac and kidney damage.

In our study, the association between systolic BPV and functional decline did not differ between hypertensive and non-hypertensive individuals. This finding may highlight the significance of current BPV, independently of previous history of hypertension.

The major strength of our study is the longitudinal design, showing that higher blood pressure variability precedes the decline in functional status. Further strengths are the large sample size, the lack of restriction on blood pressure at baseline, the consistency of our findings using two different indexes of BPV (SD and CV) and the adjustment for several potential confounders. Our participants were older adults at high cardiovascular risk, generally with high functional status. Therefore, our findings may not be generalizable to populations with different age, risk profile or functional status. However, our study population combines a high risk for functional decline together with a high potential for benefit from interventions aimed at preventing or delaying it.

In conclusion, higher visit-to-visit systolic BPV was longitudinally associated with steeper functional decline in older adults at high cardiovascular risk. We suggest that higher systolic BPV is a risk factor for functional decline and should be included in the clinical assessment of older adults at high cardiovascular risk. Further research should explore whether interventions aimed at reducing systolic BPV may preserve functional status.

REFERENCES

1. Rothwell PM. Limitations of the usual blood-pressure hypothesis and importance of variability, instability, and episodic hypertension. Lancet 2010;375:938-48.

2. Howard SC, Rothwell PM. Reproducibility of measures of visit-to-visit variability in blood pressure after transient ischaemic attack or minor stroke. Cerebrovasc Dis 2009;28:331-40.
3. Yadav S, Cotlarciuc I, Munroe PB, et al; International Stroke Genetics Consortium. Genome-wide analysis of blood pressure variability and ischemic stroke. Stroke 2013;44:2703-9.
4. Rothwell PM, Howard SC, Dolan E, et al; ASCOT-BPLA and MRC Trial Investigators. Effects of beta blockers and calcium-channel blockers on within-individual variability in blood pressure and risk of stroke. Lancet Neurol 2010;9:469-80.
5. Rothwell PM, Howard SC, Dolan E, et al. Prognostic significance of visit-to-visit variability, maximum systolic blood pressure, and episodic hypertension. Lancet 2010;375:895-905.
6. Tai C, Sun Y, Dai N, et al. Prognostic significance of visit-to-visit systolic blood pressure variability: a meta-analysis of 77,299 patients. J Clin Hypertens (Greenwich) 2015;17:107-15.
7. Brickman AM, Reitz C, Luchsinger JA, et al. Long-term blood pressure fluctuation and cerebrovascular disease in an elderly cohort. Arch Neurol 2010;67:564-9.
8. Parati G, Ochoa JE, Lombardi C, et al. Blood pressure variability: assessment, predictive value, and potential as a therapeutic target. Curr Hypertens Rep 2015;17:537.
9. Nagai M, Hoshide S, Ishikawa J, et al. Visit-to-visit blood pressure variations: new independent determinants for carotid artery measures in the elderly at high risk of cardiovascular disease. J Am Soc Hypertens 2011;5:184-92.
10. Shimbo D, Shea S, McClelland RL, et al. Associations of aortic distensibility and arterial elasticity with long-term visit-to-visit blood pressure variability: the Multi-Ethnic Study of Atherosclerosis (MESA). Am J Hypertens 2013;26:896-902.
11. Lawes CMM, Hoorn SV, Rodgers A, for the International Society of Hypertension. Global burden of blood-pressure-related disease, 2001. Lancet 2008;371:1513–18.
12. Mathers C, Fat DM, Boerma J. The global burden of disease: 2004 update. World Health Organization, 2008.
13. Shepherd J, Blauw GJ, Murphy MB, et al. The design of a prospective study of Pravastatin in the Elderly at Risk (PROSPER). PROSPER Study Group. PROspective Study of Pravastatin in the Elderly at Risk. Am J Cardiol 1999;84:1192-1197.
14. Shepherd J, Blauw GJ, Murphy MB, et al. Pravastatin in elderly individuals at risk of vascular disease (PROSPER): a randomised controlled trial. Lancet 2002;360:1623-1630.

15. Folstein MF, Folstein SE, McHugh PR. "Mini-mental state". A practical method for grading the cognitive state of patients for the clinician. J Psychiatr Res 1975;12:189-98.
16. Mahoney FI, Barthel DW. Functional Evaluation: The Barthel Index. Md State Med J 1965;14:61-65.
17. Lawton MP. The functional assessment of elderly people. J Am Geriatr Soc 1971;19:465-481.
18. Levey AS, Bosch JP, Lewis JB et al. A more accurate method to estimate glomerular filtration rate from serum creatinine: A new prediction equation. Modification of Diet in Renal Disease Study Group. Ann Intern Med 1999;130:461–470.
19. Muntner P, Shimbo D, Tonelli M, et al. The relationship between visit-to-visit variability in systolic blood pressure and all-cause mortality in the general population: findings from NHANES III, 1988 to 1994. Hypertension 2011;57:160-6.
20. Böhm M, Schumacher H, Leong D, et al. Systolic blood pressure variation and mean heart rate is associated with cognitive dysfunction in patients with high cardiovascular risk. Hypertension 2015;65:651-61.
21. Grove JS, Reed DM, Yano K, et al. Variability in systolic blood pressure--a risk factor for coronary heart disease? Am J Epidemiol 1997;145:771-6.
22. Goldstein IB, Bartzokis G, Hance DB, et al. Relationship between blood pressure and subcortical lesions in healthy elderly people. Stroke 1998;29:765-72.
23. Gunstad J, Cohen RA, Tate DF, et al. Blood pressure variability and white matter hyperintensities in older adults with cardiovascular disease. Blood Press 2005;14:353-8.
24. Puisieux F, Monaca P, Deplanque D, et al. Relationship between leuko-araiosis and blood pressure variability in the elderly. Eur Neurol 2001;46:115-20.
25. Meel-van den Abeelen AS, Lagro J, Gommer ED, et al. Baroreflex function is reduced in Alzheimer's disease: a candidate biomarker? Neurobiol Aging 2013;34:1170-6.
26. Milutinović S, Murphy D, Japundzić-Zigon N. Central cholinergic modulation of blood pressure short-term variability. Neuropharmacology 2006;50:874-83.
27. Van Beek AH, Claassen JA. The cerebrovascular role of the cholinergic neural system in Alzheimer's disease. Behav Brain Res 2011;221:537-42.
28. van Beek AH, Claassen JA, Rikkert MG, et al. Cerebral autoregulation: an overview of current concepts and methodology with special focus on the elderly. J Cereb Blood Flow Metab 2008;28:1071-85.

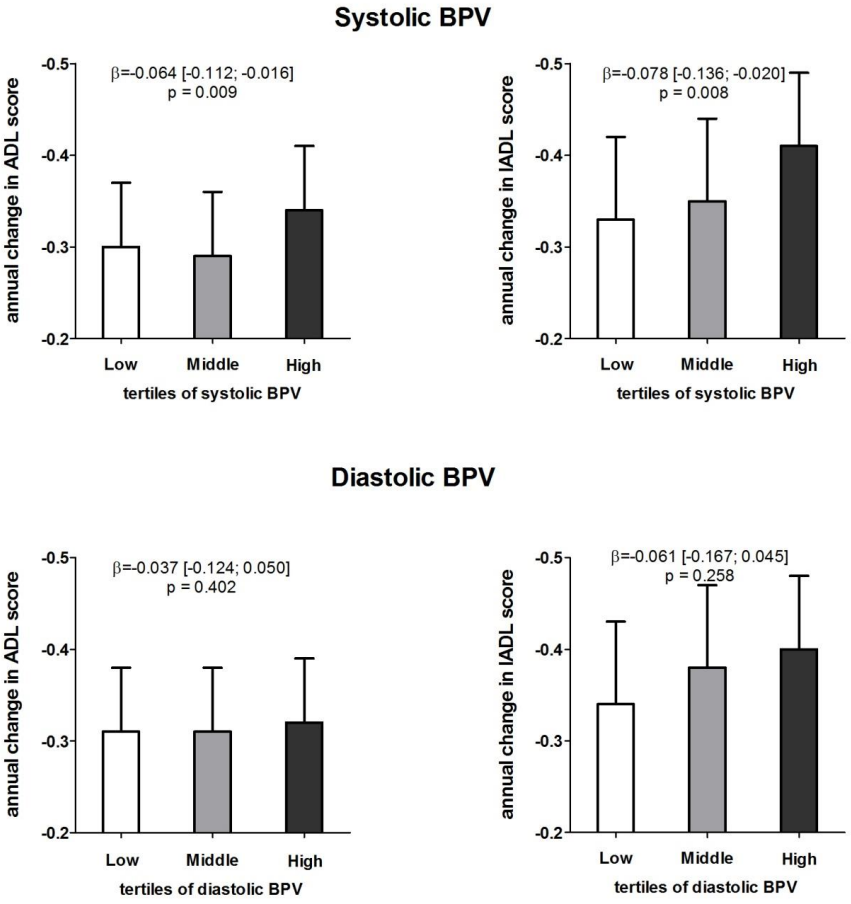
29. de la Torre JC. Pathophysiology of neuronal energy crisis in Alzheimer's disease. Neurodegener Dis 2008;5:126-32.
30. Pugh KG, Lipsitz LA. The microvascular frontal-subcortical syndrome of aging. Neurobiol Aging 2002;23:421-31.
31. Pantoni L, Garcia JH. Pathogenesis of leukoaraiosis: a review. Stroke 1997;28:652-9.
32. Zheng JJ, Delbaere K, Close JC, et al. Impact of white matter lesions on physical functioning and fall risk in older people: a systematic review. Stroke 2011;42:2086-90.
33. Taylor WD, Aizenstein HJ, Alexopoulos GS. The vascular depression hypothesis: mechanisms linking vascular disease with depression. Mol Psychiatry 2013;18:963-74.
34. Kress BT, Iliff JJ, Xia M, et al. Impairment of paravascular clearance pathways in the aging brain. Ann Neurol 2014;76:845-61.
35. Roberts KF, Elbert DL, Kasten TP, et al. Amyloid-β efflux from the central nervous system into the plasma. Ann Neurol 2014;76:837-44.
36. Masugata H, Senda S, Murao K, et al. Visit-to-visit variability in blood pressure over a 1-year period is a marker of left ventricular diastolic dysfunction in treated hypertensive patients. Hypertens Res Official J Japanes Soc Hyperten 2011;34:846–50.
37. Vishram JK, Dahlöf B, Devereux RB, et al. Blood pressure variability predicts cardiovascular events independently of traditional cardiovascular risk factors and target organ damage: a LIFE substudy. J Hypertens 2015;33:2422-30.
38. Alshawabkeh LI, Yee LM, Gardin JM, et al. Years of able life in older persons-the role of cardiovascular imaging and biomarkers: the Cardiovascular Health Study. J Am Heart Assoc 2015;4.
39. Kilpatrick ES, Rigby AS, Atkin SL. The role of blood pressure variability in the development of nephropathy in type 1 diabetes. Diabetes Care 2010;33:2442–7.
40. Okada H, Fukui M, Tanaka M, et al. Visit-to-visit blood pressure variability is a novel risk factor for the development and progression of diabetic nephropathy in patients with type 2 diabetes. Diabetes Care 2013;36:1908-12.
41. Kawai T, Ohishi M, Kamide K, et al. The impact of visit-to-visit variability in blood pressure on renal function. Hypertens Res Official J Japanes Soc Hyperten 2012;35:239–43.
42. Roshanravan B, Patel KV, Robinson-Cohen C, et al. Creatinine clearance, walking speed, and muscle atrophy: a cohort study. Am J Kidney Dis 2015;65:737-47.

Table 1. Characteristics of study population in tertiles of systolic blood pressure variability

Characteristics	Whole cohort	Tertiles of SD of SBP (mmHg)			p-value
	n=4745	Low (1.73-11.21) n=1581	Middle (11.22-15.63) n=1582	High (15.64-42.71) n=1582	
Socio-demographics					
Age, years, mean (SD)	75.2 (3.3)	74.8 (3.3)	75.2 (3.3)	75.6 (3.4)	<0.001
Female, n (%)	2494 (52.6)	797 (50.4)	824 (52.1)	873 (55.2)	0.024
Age left school, years, mean (SD)	15.2 (2.1)	15.2 (2.1)	15.1 (2.1)	15.2 (2.0)	0.152
Vascular risk factors / morbidities					
Hypertension, n (%)	2970 (62.6)	920 (58.2)	985 (62.3)	1065 (67.3)	<0.001
Stroke or TIA, n (%)	515 (10.9)	161 (10.2)	162 (10.2)	192 (12.1)	0.133
Myocardial infarction, n (%)	593 (12.5)	215 (13.6)	192 (12.1)	186 (11.8)	0.255
Claudication, n (%)	282 (5.9)	86 (5.4)	90 (5.7)	106 (6.7)	0.283
Diabetes mellitus, n (%)	483 (10.2)	160 (10.1)	168 (10.6)	155 (9.8)	0.743
Current smoking, n (%)	1212 (25.5)	433 (27.4)	396 (25.0)	383 (24.2)	0.104
BMI, kg/m ^{2} , mean (SD)	26.9 (4.2)	26.9 (4.2)	26.8 (4.2)	26.9 (4.1)	0.947
GFR, mL/min, mean (SD)	60.2 (14.3)	60.3 (13.6)	60.7 (14.4)	59.7 (14.8)	0.115
Antihypertensive drugs					
Beta-blockers, n (%)	1238 (26.1)	350 (22.1)	418 (26.4)	470 (29.7)	<0.001
Diuretics, n (%)	1909 (40.2)	629 (39.8)	634 (40.1)	646 (40.8)	0.824
ACE-inhibitors, n (%)	765 (16.1)	184 (11.6)	255 (16.1)	326 (20.6)	<0.001
Angiotensin-antagonists, n (%)	93 (2.0)	29 (1.8)	31 (2.0)	33 (2.1)	0.878
Calcium-channel blockers, n (%)	1175 (24.8)	410 (25.9)	374 (23.6)	391 (24.7)	0.328
Treatment group					
Pravastatin, n (%)	2370 (49.9)	810 (51.2)	794 (50.2)	766 (48.4)	0.278
Cognition					
MMSE, score, mean (SD)	28.1 (1.5)	28.2 (1.5)	28.1 (1.5)	28.0 (1.5)	0.022
Blood pressure (0-18 months)					
Average SBP, mmHg, mean (SD)	153.7 (16.7)	150.3 (16.4)	153.3 (16.3)	157.5 (16.7)	<0.001
N of measurements, mean (SD)	6.9 (0.5)	6.9 (0.6)	6.9 (0.4)	6.9 (0.5)	0.321

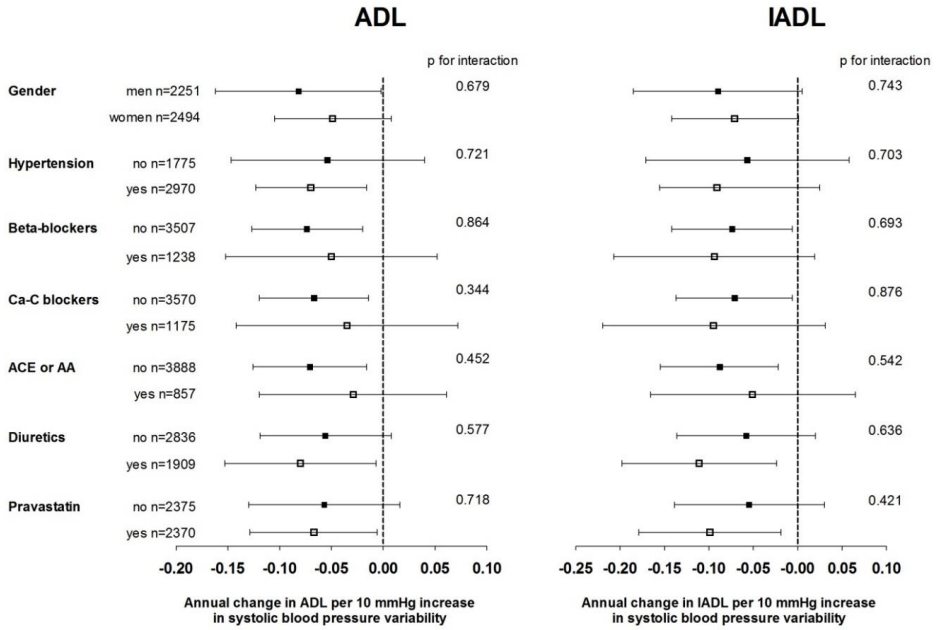
Abbreviations: SD: Standard Deviation, SBP: systolic blood pressure, n: number, TIA: Transient Ischemic Attack, BMI: Body Mass Index, GFR: glomerular filtration rate. P-values were computed using Pearson's chi-square test or Kruskal-Wallis test as appropriate.

Figure 1. Decline in ADL and IADL scores in relation to systolic and diastolic blood pressure variability



Bars represent means and standard error. β are beta-coefficients (95% confidence intervals) for annual change in ADL/IADL score for each 10 mmHg increase in blood pressure variability. P-values were calculated using continuous measures of systolic and diastolic blood pressure variability. Abbreviations: BPV: blood pressure variability, ADL: activities of daily living, IADL: instrumental activities of daily living. Analyses were adjusted for country, age, sex, education, smoking, body mass index, history of hypertension, history of diabetes mellitus, history of claudication, history of myocardial infarction, history of stroke/transient ischemic attack, glomerular filtration rate, statin treatment, diuretics, Angiotensin Converting Enzyme inhibitors, angiotensin receptor blockers, beta-blockers, calcium-channel blockers, ADL/IADL at 18 months, mean systolic blood pressure, number of measurements of systolic blood pressure and baseline Mini Mental State Examination score.

Figure 2. Decline in ADL and IADL scores in relation to systolic blood pressure variability by sex, history of hypertension, use of antihypertensives or statin



Bars represent means and 95% confidence interval. Analyses were adjusted for country, age, sex, education, smoking, body mass index, history of hypertension, history of diabetes mellitus, history of claudication, history of myocardial infarction, history of stroke/transient ischemic attack, glomerular filtration rate, statin treatment, diuretics, Angiotensin Converting Enzyme inhibitors, angiotensin receptor blockers, beta-blockers, calcium-channel blockers, ADL/IADL score at 18 months, mean systolic blood pressure, number of measurements of systolic blood pressure and baseline Mini Mental State Examination score. Abbreviations: n: number, ADL: activities of daily living, IADL: instrumental activities of daily living, Ca-C: calcium-channel, ACE: Angiotensin Converting Enzyme inhibitors, AA: angiotensin receptor blockers.

Supplementary Table 1. Decline in ADL and IADL scores in relation to systolic and diastolic BPV in participants with maximum ADL/IADL score at 18 months

	Annual change in functional status per 10 mmHg increase in BPV [95% CI]	**p-value**
Systolic BPV		
ADL (score/year, n=4069)		
Model 1	-0.084 [-0.127; -0.042]	<0.001
Model 2	-0.085 [-0.129; -0.041]	<0.001
Model 3	-0.083 [-0.127; -0.039]	<0.001
IADL (score/year, n=3776)		
Model 1	-0.121 [-0.179; -0.062]	<0.001
Model 2	-0.116 [-0.176; -0.056]	<0.001
Model 3	-0.111 [-0.170; -0.051]	<0.001
Diastolic BPV		
ADL (score/year, n=4069)		
Model 1	-0.060 [-0.139; 0.020]	0.140
Model 2	-0.057 [-0.137; 0.022]	0.159
Model 3	-0.053 [-0.133; 0.027]	0.192
IADL (score/year, n=3776)		
Model 1	-0.064 [-0.171; 0.042]	0.237
Model 2	-0.053 [-0.161; 0.054]	0.332
Model 3	-0.040 [-0.147; 0.067]	0.465

Annual change is calculated as the difference between the last ADL/IADL score and ADL/IADL score at 18 months divided by time interval. BPV was defined as standard deviation of systolic/diastolic blood pressure across visits. Abbreviations: BPV: blood pressure variability; ADL: Activities of Daily Living; IADL: Instrumental Activities of Daily Living. Model 1: adjusted for country, age, sex, education. Model 2: adjusted for country, age, sex, education, smoking, body mass index, history of hypertension, history of diabetes mellitus, history of claudication, history of myocardial infarction, history of stroke/transient ischemic attack, glomerular filtration rate, statin treatment, diuretics, Angiotensin Converting Enzyme inhibitors, angiotensin receptor blockers, beta-blockers, calcium-channel blockers, ADL/IADL at 18 months, mean systolic/diastolic blood pressure and number of measurements of systolic/diastolic blood pressure. Model 3: model 2 is additionally adjusted for baseline Mini Mental State Examination score.

Supplementary Table 2. Decline in ADL and IADL scores in relation to systolic and diastolic blood pressure variability in participants with maximum MMSE score

	Annual change in functional status per 10 mmHg increase in BPV [95% CI]	**p-value**
Systolic BPV		
ADL (score/year, n=881)		
Model 1	-0.105 [-0.182; -0.027]	0.008
Model 2	-0.088 [-0.168; -0.007]	0.032
IADL (score/year, n=881)		
Model 1	-0.132 [-0.229; -0.035]	0.008
Model 2	-0.120 [-0.221; -0.019]	0.020
Diastolic BPV		
ADL (score/year, n=881)		
Model 1	-0.045 [-0.186; 0.096]	0.527
Model 2	-0.028 [-0.168; 0.112]	0.697
IADL (score/year, n=881)		
Model 1	-0.070 [-0.247; 0.106]	0.435
Model 2	-0.052 [-0.228; 0.125]	0.565

Annual change is calculated as the difference between the last ADL/IADL score and ADL/IADL score at 18 months divided by time interval. BPV was defined as standard deviation of systolic/diastolic blood pressure across visits. Abbreviations: BPV: blood pressure variability; ADL: Activities of Daily Living; IADL: Instrumental Activities of Daily Living, MMSE: Mini Mental State Examination score. Model 1: adjusted for country, age, sex, education. Model 2: adjusted for country, age, sex, education, smoking, body mass index, history of hypertension, history of diabetes mellitus, history of claudication, history of myocardial infarction, history of stroke/transient ischemic attack, glomerular filtration rate, statin treatment, diuretics, Angiotensin Converting Enzyme inhibitors, angiotensin receptor blockers, beta-blockers, calcium-channel blockers, ADL/IADL at 18 months, mean systolic/diastolic blood pressure and number of measurements of systolic/diastolic blood pressure.

Chapter 7

General discussion

General discussion

Key findings

This thesis aimed to explore the determinants of healthy ageing and longevity in older populations, with particular focus on the homeostasis of the cardiovascular system. This thesis examined both a cohort of geriatric outpatients, a population in whom homeostasis may be affected by a complex interplay with comorbidities and frailty, and a trial cohort of old adults with preserved functional status but high cardiovascular risk, in whom the early stages of functional decline could be depicted.

In geriatric outpatients, we showed that the relationship between blood pressure and health outcomes was modified by chronological and biological age as defined by functional and cognitive status. In particular, higher blood pressure was associated with better cognitive function in those outpatients aged 85 years and older and in those with impaired functional status. Moreover, higher systolic blood pressure (below 180 mmHg) was associated with decreased all-cause mortality risk in outpatients with impaired functional and cognitive status, but not in those with preserved functional and/or cognitive status. In the same cohort, we reported that optimal thyroid status differed by sex, possibly as a consequence of higher cardiovascular burden in men compared to women. Among euthyroid older outpatients, low normal thyroid status, as characterised by high TSH and low fT4, was associated with decreased mortality risk in men whereas not in women.

In the PROSPER cohort, we examined the cross-sectional and longitudinal associations between markers of cardiac autonomic function and functional status. Cardiac autonomic function has been consistently associated with cardiovascular risk factors and comorbidities, which in turn have been related to functional decline[1-6]. Here, we showed the direct link between cardiac autonomic function and functional decline, independent of cardiovascular risk factors and comorbidities. Even among adults with preserved functional status, higher heart rate, lower heart rate variability and higher blood pressure variability could identify those at a higher risk of future functional decline.

Generalizability of findings

In our geriatric outpatients, chronological and biological age, as defined by impairments in functional and cognitive status, modulated the relationship between blood pressure and health outcomes. In the oldest and in the frailest adults, higher blood pressure was associated with better cognition and decreased mortality risk. Our findings are in line with those from population-based studies, whereas they conflict with those from clinical trials[7-14]. Looking into the demographic and clinical characteristic of our study population, we could hypothesize a continuum of frailty that places older adults of clinical trials among the fittest, those of population-based studies in an intermediate position and those of the Milan Geriatrics 75+ Cohort Study among the frailest (Figure 1). This hypothesis may explain why our findings are closer to those of population-based studies rather than those from clinical trials. It further highlights that findings from trials may not be applicable to everyday clinical practice due to the higher complexity of patient populations. Therefore, it is of high clinical relevance to perform research in patient populations.

Figure 1. Continuum fitness-frailty in different types of epidemiologic studies

Type of study	Randomized clinical trials	Population-based cohort studies	Outpatients cohort study
Limit to generalizability	Exclusion criteria: frailer are excluded	Response bias: frailer are less likely to participate	*Heterogeneity*
Removal of limit	Targeting the frailer	Removal of barriers (home-interviews, involving caregivers)	*Stratified analyses*

Clinical implications: towards tailored treatment in old age

This thesis adds to previous literature showing that the homeostasis of frail older adults may differ from that of younger or fitter adults. In particular, our findings may indicate that optimal blood pressure targets may shift towards higher values with advancing age and frailty, possibly to guarantee adequate perfusion of vital organs[15-17]. Therefore, we advocate that blood pressure management in old age should be tailored taking into account both chronological and biological age. Although a consensus definition of biological age or frailty is lacking[18], by defining it according to functional and cognitive status, we were able to effectively distinguish subpopulations of older adults in whom blood pressure goals may differ.

Furthermore, we showed that optimal thyroid status may vary according to sex and age, with high normal TSH being associated with reduced mortality risk in the oldest men. In middle-age, high normal TSH may be indicative of occult thyroid disease, which has been linked to increased morbidity and mortality[19]. Conversely, high normal TSH in old age may be a heritable trait linked to longevity, as it has been has been found in exceptionally long-lived adults and their offspring[20-21]. Therefore, in line with previous research, we advocate that TSH reference limits should be age- and sex-specific[22]. In particular, we advocate that TSH upper normal reference limit should be higher in older compared to younger men.

Novel cardiovascular risk factors for functional decline

This thesis' novel finding is that heart rate, heart rate variability and blood pressure variability, markers of cardiovascular homeostasis, are associated with risk of future functional decline, independent of comorbidities. Suboptimal cardiovascular homeostasis may be linked to functional decline through reduced resilience to stressors. While previous research focussed on specific cardiovascular diseases as outcomes, our outcome was functional decline, a global measure of disability of high relevance for individuals and society. As functional decline may considerably differ among individuals with the same diseases, it is worth shifting the focus on functional decline itself and gain insight into the mechanisms underlying it, independent of diseases.

Future perspectives

This thesis explored a complex and heterogeneous outpatient population, highlighting its diversity from those of clinical trials and population-based studies, and suggesting that associations between cardiovascular parameters and health outcomes may be reversed by age and frailty. Frailer older adults are generally excluded or less likely to be included in clinical trials and population-based studies, whereas they are referred to geriatric outpatient practices. The geriatric outpatients of the Milan Geriatrics 75+ Cohort Study comprised both the fitter and the frailer older adults. In this population, cognitive status ranged from preserved to severely impaired, functional status from complete independence to complete dependence, comorbidities from absent to numerous, and medications from none to polypharmacy. This heterogeneity represents a limit when not taken into account, whereas a strength when stratified analyses are performed. Our stratified analyses allowed us to show that the relationship between blood pressure and health outcomes may be reversed by impaired functional and cognitive status.

It is crucial to further investigate this outpatient population, as it is representative of patients for whom clinicians have to make treatment decisions in everyday practice.

This thesis reports also on findings from more classic observational studies, here based on a trial cohort. This cohort is skewed towards better health as frail individuals were by design not enrolled. Because of this selection, and given the observational design, we cannot infer whether our reported associations are causal. Further research is needed to establish whether cardiovascular parameters and thyroid status are merely risk markers or potentially modifiable risk factors for adverse health outcomes. Identifying modifiable risk factors is the first step towards intervention to delay functional and cognitive decline and mortality. Any intervention should be tested in the context of randomized controlled trials. However, trials may be long, costly or unfeasible due to ethical considerations in vulnerable older populations.

Most trials selectively include older adults with few comorbidities and no dementia; indeed, a recent community-based study found that only 9% of the oldest adults with hypertension were eligible for inclusion in The Hypertension in the Very Elderly Trial[23,24]. Recently, the randomised controlled trial Discontinuation of Antihypertensive Treatment in Elderly People (DANTE) Study Leiden investigated the effects of withdrawing antihypertensive treatment on cognition in older adults with mild cognitive impairment, showing no effect at the 16-week

follow-up[25]. The generalizability and the power of this trial were limited by the strict inclusion criteria (only old adults without serious cardiovascular diseases were included) and by the short duration of follow-up due to safety reasons. Thus, the DANTE Study Leiden illustrates the constraints encountered by researchers when dealing with older populations.

Nonetheless, we advocate that future trials may be performed in older populations in hospital-based outpatients setting, thus combining the evidence on the intervention with the high generalizability to clinical practice. These hospital-based trials may investigate the safety and efficacy of either introducing or withdrawing medications. Moreover, they may test non-pharmacological treatments such as diet and physical exercise. Choosing functional and cognitive decline as outcomes may favour both the comprehension and the willingness to participate of patients and caregivers. Including both the frailer and the fitter older adults may favour insight on how the benefits and harms of intervention may be modified by frailty. The threshold of frailty at which the harms may equal or outweigh the benefits should be assessed.

At present, given the limits of trials, valuable evidence may be derived from observational studies, coupled with deeper understanding of the pathophysiological mechanisms underlying homeostasis and homeostasis loss.

References

40. Fox K, Borer JS, Camm AJ, et al. Resting heart rate in cardiovascular disease. J Am Coll Cardiol 2007;50:823-30.
41. Palatini P, Benetos A, Julius S. Impact of increased heart rate on clinical outcomes in hypertension: implications for antihypertensive drug therapy. Drugs 2006;66:133-44.
42. Custodis F, Schirmer SH, Baumhakel M, et al. Vascular pathophysiology in response to increased heart rate. J Am Coll Cardiol 2010;56:1973-83.
43. Heart rate variability. Standards of measurement, physiological interpretation, and clinical use. Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology. Eur Heart J 1996;17:354-81.
44. Sajadieh A, Nielsen OW, Rasmussen V, et al. Increased heart rate and reduced heart-rate variability are associated with subclinical inflammation in middle-aged and elderly subjects with no apparent heart disease. Eur Heart J 2004;25:363-70.

45. Mathers C, Fat DM, Boerma J. The global burden of disease: 2004 update. Geneva: World Health Organization; 2008.
46. Sabayan B, Oleksik AM, Maier AB et al. High blood pressure and resilience to physical and cognitive decline in the oldest old: The Leiden 85-plus Study. J Am Geriatr Soc 2012;60:2014–2019.
47. Muller M, Smulders YM, de Leeuw PW et al. Treatment of hypertension in the oldest old: A critical role for frailty? Hypertension 2014;63:433–441.
48. Lewington S, Clarke R, Qizilbash N, et al; Prospective Studies Collaboration. Age-specific relevance of usual blood pressure to vascular mortality: a meta-analysis of individual data for one million adults in 61 prospective studies. Lancet 2002; 360: 1903–13.
49. Boshuizen HC, Izaks GJ, van Buuren S, et al. Blood pressure and mortality in elderly people aged 85 and older: community based study. BMJ 1998; 316: 1780–4.
50. Odden MC, Peralta CA, Haan MN, et al. Rethinking the association of high blood pressure with mortality in elderly adults: the impact of frailty. Arch Intern Med 2012; 172: 1162–8.
51. Post Hospers G, Smulders YM, Maier AB, et al. Relation between blood pressure and mortality risk in an older population: role of chronological and biological age. J Intern Med 2015; 277: 488–97.
52. Bejan-Angoulvant T, Saadatian-Elahi M, Wright JM et al. Treatment of hypertension in patients 80 years and older: the lower the better? A meta-analysis of randomized controlled trials. J Hypertens 2010; 28: 1366–72.
53. Warwick J, Falaschetti E, Rockwood K, et al. No evidence that frailty modifies the positive impact of antihypertensive treatment in very elderly people: an investigation of the impact of frailty upon treatment effect in the HYpertension in the Very Elderly Trial (HYVET) study, a double-blind, placebo-controlled study of antihypertensives in people with hypertension aged 80 and over. BMC Med 2015;13:78.
54. Marshall RS, Lazar RM. Pumps, aqueducts, and drought management: vascular physiology in vascular cognitive impairment. Stroke 2011;42:221-6.
55. Birns J, Markus H, Kalra L. Blood pressure reduction for vascular risk: is there a price to be paid? Stroke 2005;36:1308-13.
56. de la Torre JC. Pathophysiology of neuronal energy crisis in Alzheimer's disease. Neurodegener Dis 2008;5:126-32.
57. Cesari M, Gambassi G, van Kan GA, et al. The frailty phenotype and the frailty index: different instruments for different purposes. Age Ageing 2014;43:10-2.

58. Åsvold BO, Vatten LJ, Midthjell K, et al. Serum TSH within the reference range as a predictor of future hypothyroidism and hyperthyroidism: 11-year follow-up of the HUNT Study in Norway. J Clin Endocrinol Metab 2012;97:93-99.
59. Atzmon G, Barzilai N, Hollowell JG, et al. Extreme longevity is associated with increased serum thyrotropin. J Clin Endocrinol Metab 2009;94:1251-4.
60. Atzmon G, Barzilai N, Surks MI, et al. Genetic predisposition to elevated serum thyrotropin is associated with exceptional longevity. J Clin Endocrinol Metab 2009;94:4768-75.
61. Boucai L, Hollowell JG, Surks MI. An approach for development of age-, gender-, and ethnicity-specific thyrotropin reference limits. Thyroid 2011;21:5-11.
62. Peters R, Beckett N, Forette F et al. Incident dementia and blood pressure lowering in the Hypertension in the Very Elderly Trial cognitive function assessment (HYVET-COG): A double-blind, placebo controlled trial. Lancet Neurol 2008;7:683–689.
63. Jacobs JM, Stessman J, Ein-Mor E et al. Hypertension and 5-year mortality among 85-year-olds: The Jerusalem Longitudinal Study. J Am Med Dir Assoc 2012;13:759e1–759e6.
64. Moonen JE, Foster-Dingley JC, de Ruijter W, et al. Effect of Discontinuation of Antihypertensive Treatment in Elderly People on Cognitive Functioning-the DANTE Study Leiden: A Randomized Clinical Trial. JAMA Intern Med 2015;175:1622-30.

Chapter 8

Summary in English

Summary in English

Chapter 1 provides a background on the demography of population ageing in Europe and Italy and the consequent emergence of age-related diseases as causes of disability and mortality. It illustrates how preserving homeostasis is crucial to delay functional and cognitive decline as well as mortality, and suggests that homeostasis may vary in old compared to young age. Furthermore, it highlights the controversies in scientific literature regarding optimal blood pressure and thyroid status in old age and the pitfalls of evidence derived from clinical trials and population-based studies. Finally, it introduces the Milan Geriatrics 75+ Cohort Study, which was conceived to provide novel evidence on older outpatients' populations, whom clinicians encounter in everyday clinical practice. It also describes PROSPER, a randomized controlled trial on statin use in a population of old adults at high cardiovascular risk, the second cohort on which the research work for this thesis has been done.

Chapter 2 presents findings on the association between blood pressure and cognition in the Milan Geriatrics 75+ Cohort Study, indicating that higher blood pressure was associated with better cognition especially in the oldest old and in those with impaired functional status. Both chronological age and biological age as defined as impaired functional status significantly modified the relationship between blood pressure and cognition.

In **Chapter 3**, we investigated the relationship between blood pressure and mortality risk in the Milan Geriatrics 75+ Cohort Study. We showed that the relationships of systolic and diastolic blood pressure with mortality risk were U-shaped; systolic blood pressure of 165 mmHg and diastolic blood pressure of 85 mmHg were associated with the lowest mortality risk. When focussing on older adults with systolic blood pressure below 180 mmHg, higher systolic blood pressure was associated with lower mortality risk in older adults with impaired functional and cognitive status but not in those with preserved functional and/or cognitive status.

Chapter 4 explores the association between thyroid status and mortality risk in a sample of euthyroid older adults of the Milan Geriatrics 75+ Cohort Study. It shows that higher TSH and lower fT4 were associated with decreased mortality risk in men, but not in women. Sex significantly modified the relationships of TSH and fT4 with mortality risk. In addition, the inverse relationship between TSH and mortality risk was most pronounced in men aged 85 years and over.

Chapter 5 presents novel findings on the association between resting heart rate, heart rate variability and functional decline in older adults at high cardiovascular risk. Higher resting heart rate and lower heart rate variability were associated with worse functional status and with higher risk of future functional decline. It also explores the pathophysiological mechanisms that may link these markers of cardiac autonomic function to functional decline.

Chapter 6 examines the relationship between visit-to-visit blood pressure variability and functional decline in older adults at high cardiovascular risk. It expands our current knowledge on the associations between blood pressure variability and adverse health outcomes, by showing that higher systolic blood pressure variability was associated with steeper functional decline. This association was independent of mean blood pressure, cardiovascular risk factors and comorbidities and cognition.

Chapter 7 summarises the key findings of this thesis, highlighting their novelty in the context of scientific literature. It was concluded that there is a need for hospital-based cohorts in which to explore whether the relationships between risk factors and health outcomes may vary across the wide spectrum of biological and chronological age. Moreover, findings from these cohorts should guide appropriate clinical trials that are needed to assess the benefits and harms of either introducing or withdrawing medications in older adults.

Chapter 9

Summary in Italian

Riassunto in italiano

Il Capitolo 1 fornisce una panoramica sugli aspetti demografici dell'invecchiamento della popolazione in Europa e in Italia e sul consequente emergere delle patologie età-correlate quali cause di disabilità e mortalità. Illustra come preservare l'omeostasi sia cruciale al fine di posporre sia il declino funzionale e cognitivo sia la mortalità, e suggerisce come l'omeostasi possa variare nell'età avanzata rispetto all'età giovanile. Inoltre, evidenzia le controversie esistenti nella letteratura scientifica riguardo ai valori ottimali di pressione arteriosa e funzionalità tiroidea nell'età avanzata e le problematiche nell'estrapolare l'evidenza ottenuta da trial clinici e studi di popolazione. Infine, introduce lo studio Milan Geriatrics 75+ Cohort Study, che fu disegnato al fine di fornire nuova evidenza su popolazioni di pazienti anziani ambulatoriali, che i clinici incontrano nella pratica clinica quotidiana. Il **Capitolo 1** descrive inoltre lo studio PROSPER, un trial randomizzato controllato sull'uso di statina in una popolazione di anziani ad alto rischio cardiovascolare, la seconda coorte in cui il lavoro di ricerca per questa tesi è stato svolto.

Il **Capitolo 2** presenta i risultati sulla associazione tra pressione arteriosa e cognizione nello studio Milan Geriatrics 75+ Cohort Study, che mostrano come maggiori valori di pressione arteriosa siano associati a migliori performance cognitive soprattutto nei grandi anziani ''oldest old'' e negli anziani con compromissione dello stato funzionale. Sia l'età cronologica sia l'età biologica, definita quale compromissione dello stato funzionale, modificarono significativamente la relazione tra pressione arteriosa e stato cognitivo.

Nel **Capitolo 3**, abbiamo investigato la relazione tra pressione arteriosa e rischio di mortalità nello studio Milan Geriatrics 75+ Cohort Study. Abbiamo dimostrato che le associazioni di pressione arteriosa sistolica e diastolica con il rischio di mortalità avevano un andamento ad U; la pressione arteriosa sistolica di 165 mmHg e la pressione arteriosa diastolica di 85 mmHg erano associate con il minore rischio di mortalità. Quando ci siamo soffermati sugli anziani con pressione arteriosa sistolica inferiore a 180 mmHg, maggiori valori di pressione arteriosa sistolica erano associati a minor rischio di mortalità negli anziani con compromesso stato funzionale e cognitivo ma non in quelli con preservato stato funzionale e/o cognitivo.

Il **Capitolo 4** esplora l'associazione tra funzionalità tiroidea e rischio di mortalità in un campione di anziani eutiroidei dello studio Milan Geriatrics 75+ Cohort Study. Dimostra che maggiori valori di TSH e minori valori di fT4 erano associati ad un minore rischio di mortalità

negli uomini, ma non nelle donne. Il sesso significativamente modificava le associazioni di TSH e fT4 con il rischio di mortalità. Inoltre, l'associazione inversa tra TSH e rischio di mortalità era più accentuata negli uomini di età pari o superiore a 85 anni.

Il **Capitolo 5** presenta nuovi riscontri sull'associazione di frequenza cardiaca e variabilità nella frequenza cardiaca a riposo con il declino funzionale in anziani ad alto rischio cardiovascolare. Maggiori valori di frequenza cardiaca a riposo e minori valori di variabilità nella frequenza cardiaca a riposo erano associati ad un peggiore stato funzionale e ad un maggiore rischio di futuro declino funzionale. Il **Capitolo 5** inoltre esplora i meccanismi patofisiologici che possono legare tali marcatori della funzione autonomica cardiaca al declino funzionale.

Il **Capitolo 6** esamina l'associazione tra variabilità della pressione arteriosa, da visita a visita, con il declino funzionale in anziani ad alto rischio cardiovascolare. Il **Capitolo 6** espande la nostra attuale conoscenza sulle associazioni tra variabilità della pressione arteriosa e eventi avversi alla salute, dimostrando che una maggiore variabilità della pressione arteriosa sistolica era associata ad un più rapido declino funzionale. Tale associazione era indipendente dai valori medi di pressione arteriosa, dai fattori di rischio cardiovascolare, dalle comorbidità cardiovascolari e dallo stato cognitivo.

Il **Capitolo 7** riassume i risultati principali di tale tesi, sottolineando la loro originalità nel contesto della letteratura scientifica. Si conclude che vi è la necessità di coorti basate negli ospedali in cui esplorare se le associazioni tra fattori di rischio e obiettivi di salute possa variare nell'ampio spettro dell'età biologica e cronologica. Inoltre, i riscontri osservati in tali coorti devono guidare appropriati trial clinici che sono necessari al fine di stabilire i benefici e i danni dell'introduzione o della sospensione di terapie farmacologiche negli anziani.

Chapter 10

Summary in Dutch

Nederlandse samenvatting

Hoofdstuk 1 introduceert de demografie van de veroudering in Europa en Italië en de daaruit voortvloeiende opkomst van aan leeftijd gerelateerde ziekten als oorzaken van functionele beperkingen en mortaliteit. Het illustreert hoe het behoud van homeostase cruciaal is om het optreden van zowel cognitieve en functionele achteruitgang als mortaliteit uit te stellen, en stelt dat de homeostase op oudere en jongere leeftijd uiteen kan lopen. Bovendien wordt gewezen op de tegenstrijdige literatuur wat betreft optimale bloeddruk en schildklierfunctie op oudere leeftijd, en de tekortkomingen van wetenschappelijk bewijs verkregen uit klinische *trials* en bevolkingsonderzoek. Ten slotte wordt de Milan Geriatrics 75+ Cohort Study geïntroduceerd, die werd uitgevoerd om nieuw inzichten te verwerven over poliklinische populaties van oudere patiënten, waar clinici dagelijks mee te maken krijgen. Tevens wordt PROSPER beschreven, een gerandomiseerd placebo-gecontroleerd onderzoek naar statinegebruik bij oudere individuen met een hoog cardiovasculair risico, het tweede cohort waarop het onderzoek beschreven in dit proefschrift is gebaseerd.

Hoofdstuk 2 presenteert de bevindingen over het verband tussen bloeddruk en cognitie uit de Milan Geriatrics 75+ Cohort Study. Een hogere bloeddruk was geassocieerd met betere cognitie, met name bij de oudste ouderen en bij diegenen met een verminderde functionele status. Zowel chronologische leeftijd als biologische leeftijd, gedefinieerd als een verminderde functionele status, modificeerde de relatie tussen bloeddruk en cognitie.

Hoofdstuk 3 toont ons onderzoek naar de relatie tussen bloeddruk en sterfterisico in de Milan Geriatrics 75+ Cohort Study. Wij lieten zien dat de relatie tussen bloeddruk en sterfterisico U-vormig is. Hierbij waren een systolische bloeddruk van 165 mmHg en een diastolische bloeddruk van 85 mmHg geassocieerd met het laagste sterfterisico. Bij ouderen met een systolische bloeddruk onder de 180 mmHg was een hogere systolische bloeddruk geassocieerd met een lager risico op sterfte bij diegenen met een verminderde functionele en cognitieve status, echter niet bij diegenen met een behouden functionele en/of cognitieve status.

Hoofdstuk 4 beschrijft de onderzochte relatie tussen schildklierfunctie en het sterfterisico in de euthyroïde deelnemers van de Milan Geriatrics 75+ Cohort Study. Het toont hoe zowel hogere TSH als lagere fT4 geassocieerd waren met een lagere kans op overlijden bij mannen,

maar niet bij vrouwen. Geslacht modificeerde de associatie van TSH en fT4 met sterfterisico. Bovendien was de omgekeerde relatie tussen TSH en sterfterisico het meest uitgesproken bij mannen van 85 jaar en ouder.

Hoofdstuk 5 presenteert nieuwe inzichten in het verband tussen de hartslag in rust, hartslagvariabiliteit en functionele achteruitgang bij oudere volwassenen met een hoog cardiovasculair risico. Hogere hartslag in rust en lagere hartslagvariabiliteit waren geassocieerd met zowel slechtere functionele status als met een hoger risico op functionele achteruitgang in de toekomst. Tevens werden de pathofysiologische mechanismen onderzocht die deze markers van cardiale autonome functie kunnen koppelen aan functionele achteruitgang.

Hoofdstuk 6 gaat in op de relatie tussen de 'bezoek-tot-bezoek' bloeddrukvariabiliteit en functionele achteruitgang bij oudere volwassenen met een hoog cardiovasculair risico. Het breidt onze huidige kennis over de relatie tussen bloeddrukvariabiliteit en negatieve gezondheidseffecten uit, door te laten zien dat een hogere systolische bloeddrukvariabiliteit associeert met snellere functionele achteruitgang. Deze bevinding was onafhankelijk van de gemiddelde bloeddruk, cardiovasculaire risicofactoren, cardiovasculaire co-morbiditeit en cognitie.

Hoofdstuk 7 geeft een overzicht van de belangrijkste bevindingen van dit proefschrift en benadrukt de nieuw verworven inzichten voor de wetenschappelijke literatuur. Er werd geconcludeerd dat ziekenhuis-gebaseerde cohorten nodig zijn om te onderzoeken of de samenhang tussen risicofactoren en gezondheidseffecten varieert over het brede spectrum aan biologische en chronologische leeftijd. Bovendien dienen de bevindingen van deze cohorten leidend te zijn voor klinische *trials*, welke noodzakelijk zijn om de voor- en nadelen van zowel het starten als stoppen van medicatie bij oudere volwassenen te evalueren.

List of publications

1. **Ogliari G**, Westendorp RG, Muller M, Mari D, Torresani E, Felicetta I, Lucchi T, Rossi PD, Sabayan B, de Craen AJ. Blood pressure and 10-year mortality risk in the Milan Geriatrics 75+ Cohort Study: role of functional and cognitive status. *Age and Ageing*. 2015 Nov;44(6):932-7.

2. **Ogliari G**, Mahinrad S, Stott DJ, Jukema JW, Mooijaart SP, Macfarlane PW, Clark EN, Kearney PM, Westendorp RG, de Craen AJ, Sabayan B. Resting heart rate, heart rate variability and functional decline in old age. *CMAJ*. 2015 Oct 20;187(15):E442-9.

3. **Ogliari G**, Sabayan B, Mari D, Rossi PD, Lucchi TA, de Craen AJ, Westendorp RG. Age- and Functional Status-Dependent Association Between Blood Pressure and Cognition: The Milan Geriatrics 75+ Cohort Study. *J Am Geriatr Soc*. 2015 Sep;63(9):1741-8.

4. Tedone E, Arosio B, Gussago C, Casati M, Ferri E, **Ogliari G**, Ronchetti F, Porta A, Massariello F, Nicolini P, Mari D. Leukocyte telomere length and prevalence of age-related diseases in semisupercentenarians, centenarians and centenarians' offspring. *Exp Gerontol*. 2014 Oct;58:90-5.

5. Bucci L, Ostan R, Giampieri E, Cevenini E, Pini E, Scurti M, Vescovini R, Sansoni P, Caruso C, Mari D, Ronchetti F, Borghi MO, **Ogliari G**, Grossi C, Capri M, Salvioli S, Castellani G, Franceschi C, Monti D. Immune parameters identify Italian centenarians with a longer five-year survival independent of their health and functional status. *Exp Gerontol*. 2014 Jun;54:14-20.

6. Vitale G, Brugts MP, **Ogliari G**, Castaldi D, Fatti LM, Varewijck AJ, Lamberts SW, Monti D, Bucci L, Cevenini E, Cavagnini F, Franceschi C, Hofland LJ, Mari D, Janssen J. Low circulating IGF-I bioactivity is associated with human longevity: findings in centenarians' offspring. *Aging* (Albany NY). 2012 Sep;4(9):580-9.

7. Gentilini D, Mari D, Castaldi D, Remondini D, **Ogliari G**, Ostan R, Bucci L, Sirchia SM, Tabano S, Cavagnini F, Monti D, Franceschi C, Di Blasio AM, Vitale G. Role of epigenetics in human aging and longevity: genome-wide DNA methylation profile in centenarians and centenarians' offspring. *Age* (Dordr). 2013 Oct;35(5):1961-73.

8. Benigni A, Orisio S, Noris M, Iatropoulos P, Castaldi D, Kamide K, Rakugi H, Arai Y, Todeschini M, **Ogliari G**, Imai E, Gondo Y, Hirose N, Mari D, Remuzzi G. Variations of the angiotensin II type 1 receptor gene are associated with extreme human longevity. *Age* (Dordr). 2013 Jun;35(3):993-1005.

9. Meazza C, Vitale G, Pagani S, Castaldi D, **Ogliari G**, Mari D, Laarej K, Tinelli C, Bozzola M. Common adipokine features of neonates and centenarians. *J Pediatr Endocrinol Metab*. 2011;24(11-12):953-7.

10. Spazzafumo L, Olivieri F, Abbatecola AM, Castellani G, Monti D, Lisa R, Galeazzi R, Sirolla C, Testa R, Ostan R, Scurti M, Caruso C, Vasto S, Vescovini R, **Ogliari G**, Mari D, Lattanzio F, Franceschi C. Remodelling of biological parameters during human ageing: evidence for complex regulation in longevity and in type 2 diabetes. *Age* (Dordr). 2013 Apr;35(2):419-29.

11. Marengoni A, Corrao S, Nobili A, Tettamanti M, Pasina L, Salerno F, Iorio A, Marcucci M, Bonometti F, Mannucci PM; SIMI Investigators. Collaborators (77) **Ogliari G**, at al. In-hospital death according to dementia diagnosis in acutely ill elderly patients: the REPOSI study. *Int J Geriatr Psychiatry*. 2011 Sep;26(9):930-6.

12. Mari D, **Ogliari G**, Castaldi D, Vitale G, Bollini EM, Lio D. Hemostasis and ageing. *Immun Ageing*. 2008 Oct 23;5:12.

Curriculum vitae

Giulia Ogliari was born in Treviglio (BG), Italy on 11 December 1980. She studied Medicine and Surgery at the "Università degli Studi di Milano" in Milan, Italy, and graduated *cum laude* in 2006. After qualifying as a physician in 2007, she obtained a scholarship within the Italian Research Project of National Interest (PRIN) ''*Does parental longevity impact on the healthy ageing of their offspring?*" in a collaboration between the "Università degli Studi di Milano" and four other Italian universities. She carried out fieldwork in the city of Milan and neighbouring areas, actively recruiting centenarians.

In 2008, she was admitted to the School of Geriatrics at the University of Milan. She was mainly trained at the Geriatric Unit of I.R.C.C.S. Ca' Granda Maggiore Policlinico Hospital Foundation. In 2011, she participated in an exchange programme and attended classes at the Leyden Academy on Vitality and Ageing in Leiden, the Netherlands. In this context, she developed particular interest into epidemiological studies. Together with Professor Westendorp and Doctor de Craen, she conceived the first idea of an outpatients' cohort study. She graduated *cum laude* as a geriatrician in April 2012, and worked as a consultant geriatrician for a few months.

In November 2012, she started her joint PhD between the University of Milan and the Leiden University Medical Center (LUMC) in Leiden, the Netherlands.

Acknowledgements

I wish to express my admiration and gratitude to Professor Carlo Vergani, founder of the Geriatric Unit of I.R.C.C.S. Ca' Granda and of the School of Geriatrics of the Università degli Studi di Milano. By dedicating this thesis to Professor Carlo Vergani, I wish to acknowledge the silent, essential, unwitnessed, daily work of all the professionals in these two institutions in Milan, over a period of decades. I just wish to remember my former supervisor Doctor Tiziano Lucchi for his professional balance and integrity.

To the determination, resourcefulness and generosity of Professor Daniela Mari, I owe the opportunity of exploring new fields.

To the scientific creativity of Professor Westendorp and Doctor de Craen, I owe the conception of this thesis.

My warm thanks to Doctor Behnam Sabayan, as a source of scientific inspiration and support.

I would like to express my appreciation for the spirit of cooperation within the Department of Gerontology and Geriatrics of Leiden University Medical Center. I thank all my colleagues, especially my co-authors Evie and Simin. My warm regards to Majon Muller and Saskia le Cessie for several helpful suggestions.

I would like to acknowledge my friends for their support. I will mention but a few. Paola, as we shared a lot throughout the years at the Geriatric Unit as well as recently. Priscilla, for her honesty. Ruifang, for her curiosity and positive outlook.

Roelof, dearest, my greatest thanks and best wishes are to you.