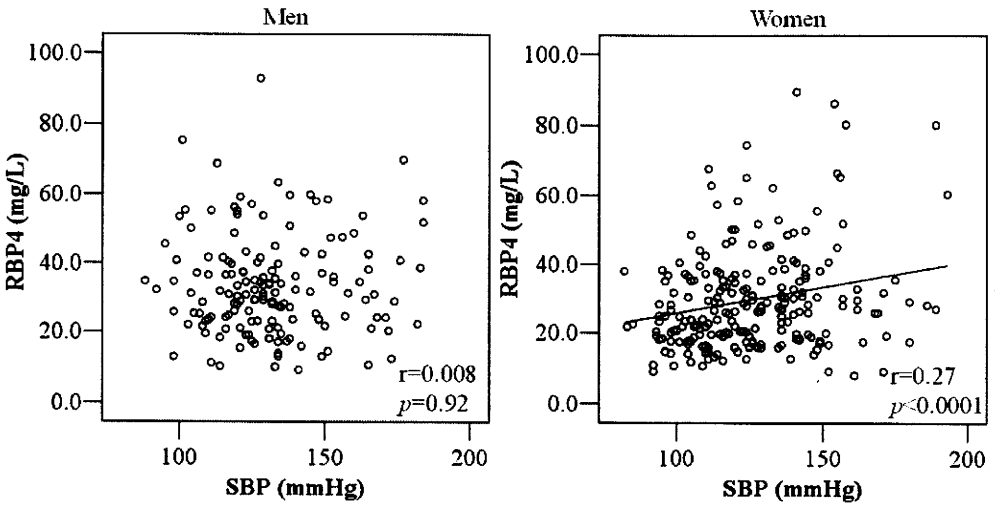

Fig. 1 There was a significant positive correlation between RBP4 and systolic BP in women (r=0.27, $p<0.0001$), but not in men (r=0.008, $p=0.92$).

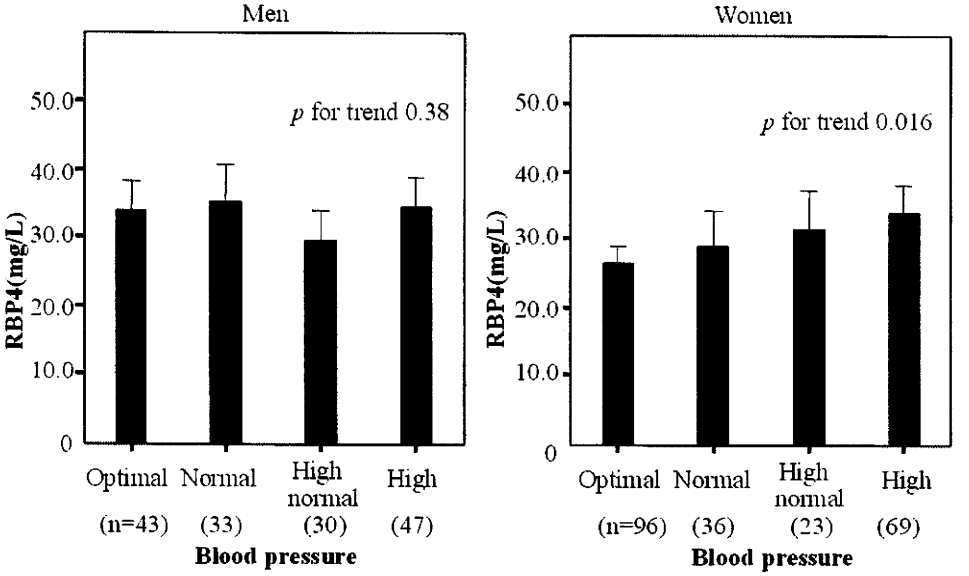

Fig. 2 There was a significant positive correlation between serum RBP4 and blood pressure (BP) categories in women. This correlation was not observed in men. The blood pressure categories are as follows: Optimal, systolic BP <120 mmHg and diastolic BP <80 mmHg; Normal, systolic BP <130 mmHg and diastolic BP <85 mmHg; High normal, systolic BP 130 to 139 mmHg and/or diastolic BP 85 to 89 mmHg; High, systolic BP ≥140 mmHg and/or diastolic BP ≥90 mmHg.

Table 4 Multiple regression analysis related to SBP

	Standardized β	*p*
Age	0.44	<0.0001
Sex	-0.053	0.24
Body mass index	0.19	<0.0001
Total cholesterol	0.095	0.046
Estimated GFR	0.041	0.42
Retinol-binding protein 4 (mg/L)	0.12	0.011

R^2=0.30, n=377 SBP, systolic blood pressure; GFR, glomerular filtration rate

Discussion

To the researchers' knowledge, there has been no epidemiologic study in Japan on the correlations between metabolic factors and RBP4. The Tanno and Sobetsu study has been performed by our department since 1976. This study is a prospective epidemiologic study aimed at clarifying the causes of and factors related to cardiovascular diseases and hypertension. Public health checkups for all residents aged 30 years or more have been conducted once a year in both of the towns and we have supported the health checkup every year. The two towns are rural areas located in the island of Hokkaido in the north part of Japan and the major industry of both towns is agriculture. Most of the participants in this cohort are middle-aged and elderly people, and their life-style, prevalence of obesity, blood pressure, blood glucose and lipid levels are similar to the results of national surveys in Japan. Therefore, this cohort is considered to be a general representation of the Japanese population.

Some prospective epidemiologic studies have shown that hyperinsulinemia and/or insulin resistance play an important role in the development of hypertension [7, 8]. Increased renal sodium retention, rennin-angiotensin system, endothelial dysfunction and microvascular remodeling have been proposed as possible mechanisms of elevation of blood pressure by insulin resistance [9-17]. Also, cross-sectional evidence suggests higher c-reactive protein levels in subjects with increased blood pressure [18-22].

Initially, reports in mice revealed that RBP4 was strongly correlated with insulin resistance. However, some subsequent studies in human subjects failed to identify a relationship between RBP4 and insulin sensitivity [4, 23]. In our cross-sectional study, a relationship between RBP4 and insulin resistance was not found, one possible reason being the characteristics of the subjects in this study. Our subjects were relatively old and healthy and had a small range of insulin sensitivities. This result was consistent with the report of Solini *et al.* [5]. We speculate that detection of RBP4-mediated changes in insulin sensitivity may require subjects with a wider range of insulin sensitivities than the range of sensitivities in our subjects [4, 5]. Another possible reason is that our subjects had relatively normal eGFR. It is known that RBP4 levels are elevated in states of impaired kidney function [24]. A previous study also showed that RBP4 levels are significantly increased in end-stage renal disease and that renal excretion is a primary pathway for RBP4 clearance [25]. Previous studies with positive results for a relationship between RBP4 and insulin resistance were conducted for human subjects with impaired kidney function and/or type 2 diabetes mellitus [3, 24]. On the other hand, studies conducted for human subjects with normal kidney function did not have positive results for a relationship between RBP4 and insulin resistance [5]. This study circumvents the difficulties previously described regarding the correlations of RBP4 with eGFR and diabetic nephropathy [26].

RBP4 levels were found to be sexually dimorphic, which had previously been reported for adipocytokines, such as leptin and adiponectin, and explained on the basis of different fat amounts and the influence of sex hormones [27-30]. Although data regarding menopausal status was unavailable in our female subjects, RBP4 levels in women over the age of 50 years were found to be significantly higher than those of women under the age of 50 years (data not shown) while levels in men were higher than both. Thus, it appears that differences in sex hormone status might affect RBP4 levels.

It has been reported that men have increased blood pressure compared to women of the same age group. In the present study group, the effect of age on men's level of blood pressure might have weakened the relationship between RBP4 and elevation of blood pressure in men.

As mentioned above, there are some pathways involving the role of insulin resistance in the elevation of blood pressure. On the other hand, there are other pathways of blood pressure elevation that are not mediated by insulin resistance.

Previous studies that support a positive relationship between RBP4 and atherosclerosis reported the mechanism as follows: their findings that higher fat content in the vessel wall and in atherosclerotic plaques might reflect lipid-modulation activities of retinoids and retinol-binding proteins, such as expression of several genes involved in triglyceride metabolism [5, 31]. In addition, based on the results of the present study regarding the relationship between RBP4 and systolic blood pressure, the following mechanism can be speculated. Peroxisome proliferator-activated receptors (PPARs) belong to a subfamily of the nuclear receptors superfamily and are ligand-activated transcription factors that heterodimerize with the retinoid

X receptor (RXR) and recognize PPAR response elements (PPRE) [32]. PPAR/RXR heterodimer directly binds to the PPRE and increases the promoter transcriptional activity in cells. Inflammation markers in macrophages such as CD36 have functional PPRE in their promoters [33]. From these findings, we speculate that cellular uptake of retinol by RBP4 activates this inflammation and induces arteriosclerosis, resulting in elevated blood pressure.

Our study has several limitations. First, causality cannot be assessed due to the cross-sectional nature of our study. Therefore, a longitudinal study is needed to better explain the relationship between RBP4 and blood pressure. Second, because the subjects of our study were only Japanese people, the generalizability to other ethnicities is unknown. Third, our findings are based on an epidemiological study, and the pathophysiological mechanism is therefore a speculation. Further studies are needed to evaluate clinical implications of our findings. Measurements of IMT, pulse wave velocity (PWV) and other cardiovascular markers such as monocyte chemotactic protein 1 (MCP-1) and interleukin-6 (IL-6) may strengthen our findings.

In conclusion, in addition to correlations between blood pressure and both HOMA-R and hs-CRP, increased levels of RBP4 were significantly associated with increased systolic blood pressure.

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〈原　著〉

地域一般住民高齢者・非高齢者における腹部肥満の糖尿病発症リスクに関する検討—端野・壮瞥町研究—

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要　約　**目的**：地域一般住民における腹部肥満と糖尿病発症との関連について高齢者と非高齢者での影響の違いを，端野・壮瞥町住民健診受診者のデータから検討した．**方法**：1994年に住民健診を受診し，かつ2003年または2004年にも健診を受診した1,023名中，データ欠損者，1994年の時点での糖尿病者（空腹時血糖値≧126 mg/dlまたは糖尿病治療中の者）を除いた827名を対象とした．1994年のデータに基づいて65歳以上の高齢者群，65歳未満の非高齢者群に分け，さらに日本のメタボリックシンドローム診断基準に基づいて腹部肥満群と非腹部肥満群に分けた．上記4群において，2003・04年の受診時点での糖尿病発症者の頻度を比較検討した．**結果**：非高齢者群においては非腹部肥満群に対し腹部肥満群からの糖尿病発症が有意に高率であったが，高齢者群において統計学的有意差は認められなかった．非高齢者・高齢者で糖尿病発症を従属変数とし，年齢，性別，総コレステロール，収縮期血圧値，喫煙の有無，糖尿病発症家族歴有無，空腹時血糖110 mg/dlの有無で調整したロジスティック回帰分析では，高齢者群において腹部肥満は関連要因とはならず，非高齢者群ではオッズ比2.68で糖尿病発症の有意なリスクとなった．腹部肥満の有無と血圧高値，血糖高値，脂質異常症の危険因子集積の有無を同時にモデルにいれたロジスティック回帰分析では，非高齢者群で腹部肥満が3.10，危険因子集積が3.00とそれぞれ独立して新規糖尿病発症の有意なリスクとなったが，高齢者群では，危険因子集積のみが3.70と新規糖尿病発症の有意なリスクとなった．**結論**：65歳未満の非高齢者において青壮年期からの腹部肥満への介入が重要であるのはもちろんのこと，高齢者においては腹部肥満なしと判定される者の中でも危険因子集積者は糖尿病のハイリスクであるため，生活習慣見直し等の介入が必要となる可能性が示唆された．

Key words：高齢者，腹部肥満，糖尿病，インスリン抵抗性，端野・壮瞥町研究

（日老医誌　2011；48：71–77）

緒　言

糖尿病患者は世界的に増加傾向にあり，2000年の1億7,100万人から2030年には3億6,600万人に増加すると推計されており，特に65歳以上の高齢糖尿病者の増加が懸念されている[1]．またアジア地域では肥満者の増加と共に糖尿病患者が急増しており[2]，日本でも生活習慣の欧米化により糖尿病患者は増加傾向で，厚生労働省の調査では2002年の740万人から2007年では890万人へと増加している．特に高齢者において糖尿病とその予備群の増加が顕著であり，高齢者における糖尿病予防対策は極めて重要であると考えられる．

近年，内臓脂肪の蓄積から動脈硬化危険因子が個人に集積する病態としてメタボリックシンドロームが注目され，心血管疾患イベントのみならず糖尿病発症と密接に関係するという報告がされている[3)4)]．わが国において2008年度より開始された特定健診・特定保健指導においてはメタボリックシンドロームが重要な骨子として採用されており[5)]，心血管疾患や糖尿病も含めた種々の生活習慣病の予防対策としてメタボリックシンドロームに該当する者やその予備群に積極的に介入を行うことになっている．その中でも腹囲径によって判定される腹部肥満は保健指導対象者の階層化の最初のステップであり，現在の腹部肥満の基準が将来の生活習慣病罹患に与える影響を検討することは重要である．

高齢者の糖尿病発症は，耐糖能異常からの発症が多く[6)]，その原因としては加齢によるインスリン抵抗性の増大や内臓脂肪の蓄積といった体組成の変化などが知ら

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れているが，腹囲径によって判定される腹部肥満が糖尿病発症に与える影響に関する報告は少ない．特に，特定健診・特定保健指導では生活習慣改善による予防効果は若年の方が高いという理由から，65歳以上の高齢者に対しては，積極的支援の対象になった場合でも動機付け支援に留めることとなっている．よって階層化の最初のステップとなる腹部肥満が将来の糖尿病の罹患に与える影響について高齢者と非高齢者での影響の違いを検討することは非常に重要である．

今回我々は端野・壮瞥町住民健診受診者を対象に地域一般住民における高齢者と非高齢者における腹部肥満の糖尿病罹患に与える影響について検討を行った．

方　法

端野・壮瞥町研究は1976年に札幌医大第二内科で開始された疫学研究である[6,7]．今回は1994年の受診者1,934名から2003年あるいは2004年にも受診した1,023名中，体重，腹囲，血圧などのデータ欠損者および1994年の時点での糖尿病者（空腹時血糖値（FPG）≧126 mg/d *l* または糖尿病治療中の者）を除いた827名（男性347名；平均年齢59.6±9.0歳，女性480名；平均年齢58.3±8.5歳）が解析対象となった．

早朝空腹時に身長，体重，臍周囲腹囲径，安静坐位にて血圧値（収縮期血圧（SBP），拡張期血圧（DBP））を測定し，採血にて血糖値，総コレステロール値(T.chol)，中性脂肪値（TG），HDLコレステロール値（HDL-C）を測定した．服薬状況や喫煙状況，糖尿病家族歴に関しては保健師の問診により確認した．

1994年の健診データに基づいて65歳未満の非高齢者群と65歳以上の高齢者群の2群に分け，さらにわが国のメタボリックシンドローム診断基準[8]に基づいて腹部肥満（AO）群と非腹部肥満（Non-AO）群の2群に分け，高齢者・非高齢者別にAO，Non-AOからの2003・04年の健診時の新規糖尿病者（FPG≧126 mg/d *l* または1994年から2003・04年までの間に糖尿病治療が開始となった者）の割合を比較した．また血圧高値（SBP≧130 mmHg かつ/または DBP≧85 mmHg かつ/または高血圧治療中），血糖高値（110 mg/d *l* ≦FPG＜126 mg/d *l*），脂質代謝異常（TG≧150 mg/d *l* かつ/または HDL-C＜40 mg/d *l* かつ/または脂質異常症治療中）の危険因子のうち二つ以上をもつ者を集積群，二つ未満を非集積群とし，初年度の危険因子の集積の有無が糖尿病罹患に与える影響についても高齢者・非高齢者別に検討を行った．

また2003・04年の健診では空腹時血漿インスリン値（FIRI）も測定しているため，断面成績の結果から腹部肥満の有無とHOMA-R（＝FPG×FIRI/405）を指標としたインスリン抵抗性との関連を高齢者・非高齢者別に検討した．なおこの解析においては治療中の者も含めた糖尿病者を除外して検討を行っている．インスリン抵抗性の評価としては教室既報の結果より[9]HOMA-R≧1.73をインスリン抵抗性あり，HOMA-R＜1.73をインスリン抵抗性なしと判定し，高齢者・非高齢者群，AO・Non-AO群でのインスリン抵抗性者の頻度を比較検討した．

統計解析はIBMSPSS ver. 17.0を使用した．統計学的有意水準は$p<0.05$とし，値は平均値±標準偏差で表した．連続変数の差の検定にはunpaired t-testを，頻度の差の検定にはχ^2検定を用いた．また2003・04年の健診時の新規糖尿病の有無を従属変数としたロジスティック回帰分析を用いて，腹部肥満や危険因子集積の有無のオッズ比を算出した．交絡要因としては年齢，性別，喫煙，糖尿病家族歴，T.chol，SBPを考慮した．

本研究は札幌医科大学倫理委員会の承認を得ており，また健診受診者全員に研究内容を説明の上，文書による同意が得られた者のみを対象とした．

結　果

Non-AO群とAO群別の高齢者・非高齢者の背景を示す（表1）．Non-AO群では年齢以外にSBP，DBP，FPGが高齢者において非高齢者に比べて有意に高値であった．AO群では高齢者・非高齢者間で年齢以外の項目に有意な差はみられなかった．

高齢者・非高齢者別に初年度の腹部肥満の有無からの糖尿病発症頻度を比較したところ，非高齢者群においてはNon-AO群に対しAO群からの糖尿病発症頻度が有意に高率であった（Non-AO群5.4% vs. AO群16.9%，$p<0.0001$）が，高齢者群においてはNon-AO群に対してAO群からの糖尿病発症頻度は高い傾向にはあったが統計学的有意差は認められなかった（Non-AO群7.1% vs.AO群12.7%）（図1）．

また初年度の腹囲径と新規糖尿病発症との関連を検討すると，非高齢者では腹囲径の増大とともに糖尿病発症頻度が増加し（男性：p for trend＝0.008，女性：p for trend＝0.021），80 cm以上になると増加する傾向にあるが，高齢者では特に女性において80 cm未満の低い腹囲径からすでに糖尿病発症者の増加がみられ，統計的には有意な傾向は認められなかった（図2）．さらに新規糖尿病発症を従属変数としたロジスティック回帰分析において，年齢，性別，T. chol，SBP，喫煙の有無，糖尿病家族歴の有無，初年度の血糖高値の有無で調整すると，

表1　対象背景（1994年）
Non-AO群では年齢以外にSBP，DBP，FPGが高齢者において非高齢者に比べて有意に高値であり，AO群では高齢者・非高齢者間では年齢以外の項目に有意差を認めなかった．

	Non-AO (n=654)		AO (n=173)	
	非高齢者 n=484	高齢者 n=170	非高齢者 n=118	高齢者 n=55
Age	54.9±7.0	68.6±3.2**	56.5±7.2	69.2±4.1**
Male/Female	136/348	65/105	103/15	43/12
WC (cm)	74.5±6.8	76.3±6.5**	91.8±5.6	91.8±4.9
SBP (mmHg)	128.7±17.3	140.9±19.1**	135.9±16.8	139.6±18.9
DBP (mmHg)	76.5±9.6	79.0±8.7**	82.0±8.7	79.9±9.3
T.chol (mg/dl)	194.0±33.1	192.1±30.1	192.1±30.5	189.0±25.2
TG (mg/dl)	117.8±81.3	125.4±79.0	167.3±130.7	153.4±83.1
HDL-C (mg/dl)	57.8±13.4	56.0±4.0	50.3±12.1	49.5±13.8
FPG (mg/dl)	92.2±9.0	93.8±9.5*	98.2±9.4	97.5±10.5

*p<0.05，**p<0.01 vs. 非高齢者
WC：腹囲，SBP：収縮期血圧，DBP：拡張期血圧，T.chol：総コレステロール，TG：トリグリセリド，HDL-C：HDLコレステロール，FPG：空腹時血糖値

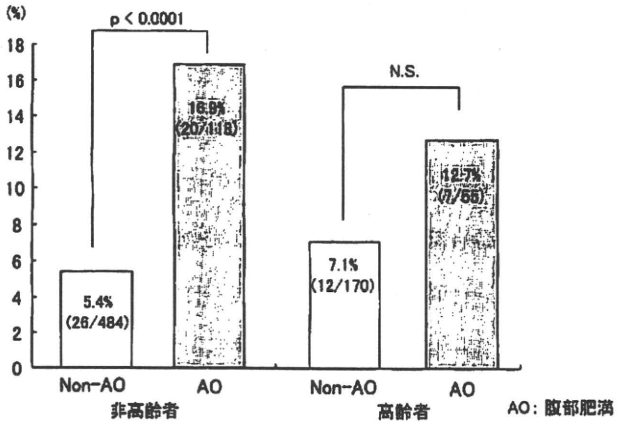

図1　高齢者・非高齢別の腹部肥満の有無での糖尿病発症頻度の比較
非高齢者群においてはNon-AO群に対しAO群からの糖尿病発症頻度が有意に高率であったが，高齢者群においてはNon-AO群に対してAO群からの糖尿病発症頻度は高い傾向にはあったが有意差は認められなかった

高齢者群において腹部肥満は有意なリスクとはならず，非高齢者群ではオッズ比2.68（95%CI：1.05～6.90）と新規糖尿病発症リスクとなった（表2）．

また腹部肥満の有無と危険因子集積の有無を同時にモデルに入れた場合，年齢，性別，T.chol，喫煙の有無，糖尿病家族歴の有無で調整したオッズ比は，非高齢者群においては腹部肥満で3.10（95%CI：1.26～7.63），危険因子集積が3.00（95%CI：1.89～4.76）とそれぞれ独立して有意な糖尿病発症のリスクとなったが，高齢者群において腹部肥満は有意なリスクとはならず，危険因子集積のみが3.70（95%CI：1.65～8.72）と糖尿病発症の有意なリスクとなった（表3）．

高齢者と非高齢者での腹部肥満の糖尿病罹患に対する影響の違いの原因を検討する目的で，2003・04年の断面調査からNon-AO・AO別に高齢者・非高齢者群におけるインスリン抵抗性者の頻度を比較検討すると，Non-AO群では非高齢者に比べ高齢者でのインスリン抵抗性者の頻度が有意に高値を示したが（高齢者8.6%vs非高齢者2.8%，p=0.015），AO群では高齢者において非高齢者よりも頻度は高い傾向にあるものの，統計学的に有意な差は認められなかった（高齢者23.8% vs. 非高齢者19.4%）（図3）．

考　察

今回の検討において，非高齢者では日本基準の腹部肥満が糖尿病のリスクとなるが，高齢者では有意なリスクとはならなかった．これまで腹部肥満を糖尿病のリスクとして多民族・人種間で検討した先行研究では，BMI25以下・25～30・30以上のいずれとも糖尿病は関係があったと報告[10]，またアジアでの糖尿病疫学研究では，2型糖尿病は短期間にその割合が増加し，欧米よりも比較的若年層からの発症や低いBMIからの発症が特徴であると報告されている[2]．両者とも年齢は直接的な要因ではなく，交絡要因として解析で調整されているため，高齢者と非高齢者で層別化して腹部肥満の影響度の違いを検討した報告はほとんどみられない．

また今回の解析において非高齢者では腹囲径の増大と

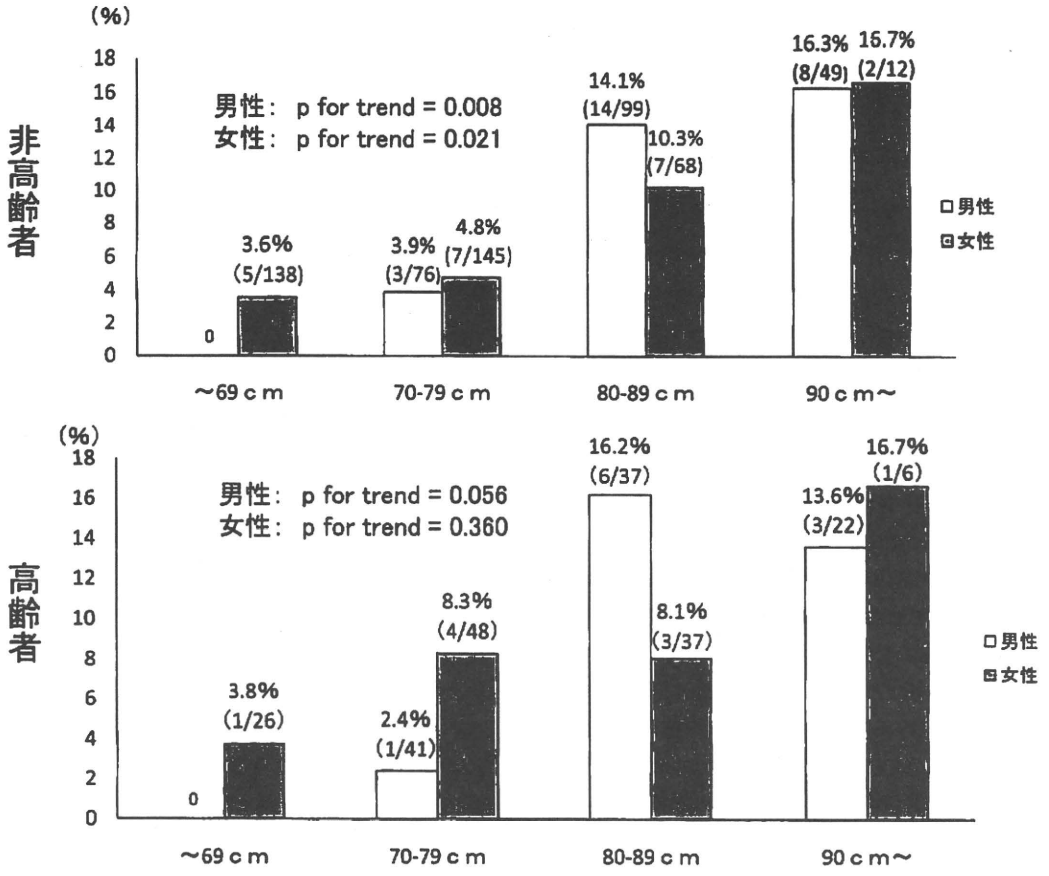

図2　高齢者・非高齢者別の各腹囲区分からの糖尿病発症頻度
初年度の腹囲径からの新規糖尿病発症の関連を検討すると，非高齢者では腹囲径の増大とともに糖尿病発症頻度が増加し，80 cm 以上になると増加する傾向にあるが，高齢者では特に女性において 80 cm 未満の低い腹囲径からすでに糖尿病発症者の増加がみられた

表2　ロジスティック回帰分析による高齢者・非高齢者別の腹部肥満の糖尿病発症に対するオッズ比
新規糖尿病発症を従属変数としたロジスティック回帰分析において，年齢，性別，T. chol. SBP，喫煙の有無，糖尿病家族歴の有無，初年度の血糖高値の有無で調整すると，高齢者群において腹部肥満は有意なリスクとはならず，非高齢者群では有意な新規糖尿病発症リスクとなった．

	Model 1	Model 2	Model 3
非高齢者	3.67** (1.67 ～ 8.27)	3.72** (1.59 ～ 8.67)	2.68* (1.05 ～ 6.90)
高齢者	1.47 (0.47 ～ 4.59)	1.60 (0.49 ～ 5.19)	0.67 (0.16 ～ 2.84)

*p<0.05，**p<0.01．（ ）は95% 信頼区間
T. chol：総コレステロール，SBP：収縮期血圧，FPG：空腹時血糖値
Model 1：年齢，性別で調整
Model 2：Model 1＋T. chol. SBP，喫煙，糖尿病家族歴で調整
Model 3：Model 2＋FPG≧110 mg/dl の有無で調整

ともに糖尿病発症頻度が増加するが，高齢者では特に女性においてはより低い腹囲径から糖尿病発症者の増加が認められた．すなわち高齢者では現在の腹囲基準で腹部肥満と判定されない群からも糖尿病発症が非高齢者より

表3　ロジスティック回帰分析による高齢者・非高齢者の腹部肥満と危険因子集積の糖尿病発症に対するオッズ比
腹部肥満の有無と危険因子集積の有無を同時にモデルに入れたロジスティック回帰分析では年齢，性別，T. chol，喫煙の有無，糖尿病家族歴の有無で調整したオッズ比は，非高齢者群においては腹部肥満，危険因子集積でそれぞれ独立して有意な糖尿病発症のリスクとなったが，高齢者群において腹部肥満は有意なリスクとはならず，危険因子集積のみが新規糖尿病発症の有意なリスクとなった．

		Model 1	Model 2
非高齢者	腹部肥満	2.54* (1.07 ～ 6.02)	3.10* (1.26 ～ 7.63)
	危険集積	2.68** (1.72 ～ 4.17)	3.00** (1.89 ～ 4.76)
高齢者	腹部肥満	0.84 (0.23 ～ 3.11)	0.81 (0.22 ～ 3.08)
	危険集積	3.72** (1.69 ～ 8.19)	3.70** (1.65 ～ 8.72)

*p<0.05，**p<0.01，（ ）は95%信頼区間
T. chol：総コレステロール
Model 1：年齢，性別で調整
Model 2：Model 1＋T. chol，喫煙，糖尿病家族歴で調整

も多く認められる結果であった．

日本人糖尿病患者を対象とした先行研究では，高齢者では非肥満者においてもインスリン抵抗性者の頻度や危険因子集積者の頻度の増加が報告されている[11]．この中で腹部肥満とインスリン抵抗性の増大について年齢に伴う増加がみられ，腹囲とインスリン抵抗性に明確な関係があった．ただしインスリン抵抗性の増大は腹囲のみならず，メタボリックシンドロームを構成する危険因子集積があり腹部肥満がない事例でもみられたと報告している．本研究においても，腹部肥満群においては非高齢者も高齢者も確かにインスリン抵抗性が増大しているが，非腹部肥満群であっても高齢者では加齢による影響もありインスリン抵抗性を背景として危険因子が集積している者が非高齢者よりも多く含まれる結果であったことから，前述の報告を支持するものと考えられた．

またメタボリックシンドロームは内臓脂肪蓄積を上流にインスリン抵抗性を一つの背景として動脈硬化危険因子が集積する病態であり，心血管疾患イベントのリスクのみならず糖尿病発症のリスクとなることも報告されている[3]．久山町研究では，メタボリックシンドロームが空腹時血糖高値とは独立した糖尿病発症の予測因子として有用であると報告している[4]．この報告で使用されたメタボリックシンドロームの診断基準はATP-IIIであり腹囲基準は必須ではなくリスクの1つとして定義されている．本研究において危険因子集積が糖尿病発症の予測因子となったことは，この結果に矛盾しないものと考えられる．

非高齢者においては腹部肥満が有意な糖尿病発症予測因子であったのに対し，高齢者においては有意な予測因子とはならなかった理由として，一つには高齢者においては腹部肥満なしと判定される者にもインスリン抵抗性を背景とした危険因子集積者が非高齢者に比べて多く含まれており，腹部肥満の有無で2群に分けた場合，非腹部肥満群でも腹部肥満群と同程度に糖尿病リスクが高くなるために2群の差が薄まって有意な差として認められなかった可能性が考えられる．また，もう一つの可能性として，今回の検討において初年度にすでに糖尿病のため除外した者のうち約6割が65歳以上の高齢者であり，さらにその約半数が腹部肥満者であったことから，腹部肥満者は青壮年期から肥満傾向を認めておりそのような糖尿病発症のハイリスク者は65歳までには多くが糖尿病を発症しているため，65歳の時点で糖尿病でない腹部肥満者のその後の糖尿病発症リスクはそれほど高くならなかった可能性も考えられる．

三つ目の理由として，高齢者の体組成の変化として，筋肉等の除脂肪体重の減少に加え，脂肪組織でも皮下脂肪が相対的に減少して内臓脂肪が増加するという変化を示す[12)~14)]ことから，今回の検討での高齢者の非腹部肥満群の中には，腹囲径としては基準に該当しないものの相対的に内臓脂肪蓄積量が多い者が含まれるために，非腹部肥満群と腹部肥満群との間に有意な差が認められなかった可能性も考えられる．

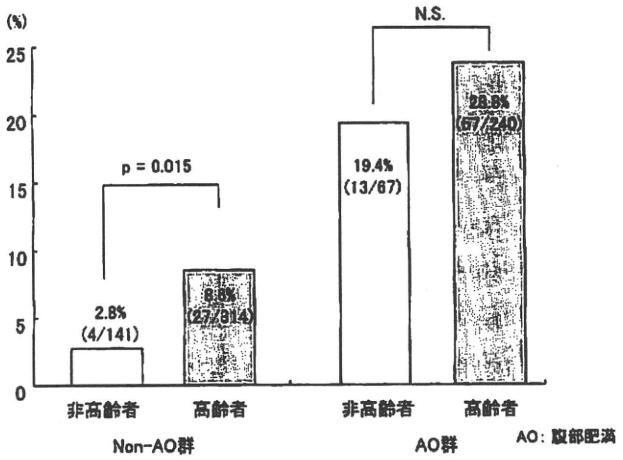

図3　HOMA-R≧1.73で判定したインスリン抵抗性の頻度
2003・04年の断面調査からNon-AO・AO別に高齢者・非高齢者群におけるインスリン抵抗性者の頻度を比較検討すると，Non-AO群では非高齢者に比べ高齢者でのインスリン抵抗性者の頻度が有意に高値を示したが，AO群では高齢者において非高齢者よりも頻度は高い傾向にあるものの，統計学的に有意な差は認められなかった

本研究の限界について述べる．今回は1994年と2003・04年の両年受診者を解析対象としたため，解析対象とならなかった2003・04年受診なし群と今回の解析対象との間で初年度特性の比較をしたところ，2003・04年受診なし群において男性の比率が高く（2003・04受診あり群男/女：412/621，なし群男/女：427/484），平均年齢（2003・04受診あり群58.1±9.5，なし群63.0±13.2，$p<0.01$），SBP（2003・04受診あり群132.8±19.7，なし群140.0±21.9，$p<0.01$），DBP（2003・04受診あり群77.8±9.7，なし群79.6±10.1，$p<0.01$），FPG（2003・04受診あり群95.3±14.7，なし群101.4±39.7，$p<0.01$）が有意に高い傾向を示していた．よって，より高齢あるいは動脈硬化危険因子が重症な者ほど，死亡や入院あるいは医療機関での厳格な管理が必要となって10年後の健診を受診できなかった可能性が考えられ，今回の結果は10年後も健診を受診できる比較的健康状態がよく，また定期的に検診を受ける健康意識の高い者が中心となる対象での解析であり，結果を過小評価している可能性が考えられた．

また，今回は初年度の腹囲径と10年後の糖尿病罹患との関連を示したが，途中の時点での腹囲径は測定されておらず，腹囲径の変化がその後の糖尿病の罹患に与える影響については検討できなかった．本研究は観察研究であるが，腹囲径の変化がその後の糖尿病罹患に与える影響を検討することは重要であり，今後の検討課題であると考えられる．

結論として，非高齢者においては腹部肥満が強い糖尿病発症リスクとなることから青壮年期からの腹部肥満への介入が重要である．また特定健診・特定保健指導では65歳以上の前期高齢者においてはメタボリックシンドローム該当者にも動機付け支援を行うこととなっているが，今回の検討においては腹部肥満に該当しない高齢者にも将来の糖尿病のハイリスク者が非高齢者と比較して多く含まれていたことから，糖尿病予防の観点からは，高齢者の危険因子集積者においては例え腹部肥満の基準を満たさなくとも糖尿病予防のためのライフスタイル改善の指導が重要となる可能性が示唆された．

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Gender-specific risk stratification with plasma B-type natriuretic peptide for future onset of congestive heart failure and mortality in the Japanese general population

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Abstract

Background: Elevated plasma B-type natriuretic peptide (BNP) levels suggest a high risk for future onset of cardiovascular events including congestive heart failure (CHF) and mortality. In the general population, although median plasma BNP levels have been reported to be higher in women than in men, the incidence of CHF and mortality are lower in women. However, no studies have examined gender-specific risk stratification of plasma BNP levels for future onset of CHF and mortality.
Methods: Subjects of this study were recruited from our general population. Baseline data including plasma BNP were determined in 13,466 subjects (men 4527, women 8939; median age = 64 yrs). A multivariate Cox regression analysis was performed to examine the predictive ability of plasma BNP for new onset of CHF and mortality.
Results: The mean follow-up duration was 2.9 years. After adjustment for traditional cardiovascular risk factors including atrial fibrillation, hazard ratios for CHF development for values above the 75th percentile of BNP were 13.4 ($p<0.001$) in men and 8.5 ($p<0.001$) in women. Similarly, each increment of 1SD in log BNP levels increased the hazard ratio by 8.8 ($p<0.001$) in men, and 6.7 ($p<0.001$) in women. The area under the receiver operating characteristic curve was significant for prediction of the onset of CHF (men; 0.853, women; 0.838). In addition, increased plasma BNP levels implied high risk of any-cause mortality in men (above the 75th percentile; hazard ratio = 1.8, $p=0.005$: increment of 1SD; hazard ratio = 1.4, $p=0.024$), but this relationship was suboptimal in women.
Conclusion: Measurements of plasma BNP provides strong predictive information about future onset of CHF in both sexes, with predictive ability for death being effective especially in men.

Keywords: Mortality; Community; Non-white; Japanese

B-type natriuretic peptide (BNP) has been recognized as a hormone released with its biologically inactive N-terminal peptide (NT-proBNP) from the heart [1,2]. It has been suggested that cardiac secretion of BNP is increased by elevated left ventricular end-diastolic pressure, decreased

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cardiac systolic/diastolic function, hypertensive heart disease, atrial fibrillation, and myocardial ischemia [3–7]. It is therefore expected that measurement of plasma BNP levels would provide useful information for identification of subjects at high risk of CHF due to various phenotypes of structural heart disease [8,9].

In fact, Wang et al. have shown for the first time that a 1standard deviation increment in plasma BNP as well as elevated plasma BNP above the 80th percentile was associated with a significant increase in the risk of new onset of CHF and any-cause mortality in the Framingham general population [10]. Similar positive associations between plasma NT-proBNP levels and risk of cardiovascular events including onset of CHF and mortality have been reported in the Danish general population [11]. These previous studies have suggested that plasma levels of BNP or NT-proBNP may serve as a possible screening tool for subjects at high risk of CHF and death within the general population. However, few studies have examined the utility of plasma BNP measurement as a predictor of future onset of CHF by a sex-stratified analysis, because women have a relatively lower incidence of CHF and higher median plasma BNP levels than men [12–14].

Moreover, subjects of previous studies were mainly drawn from white populations [10,11]. No studies have confirmed the relationship between plasma BNP levels and cardiovascular events and mortality in community-based cohorts taken from non-white populations. The incidence and prevalence of cardiovascular events including CHF has been reported to differ among ethnic groups [15]. In the general population, several reports have shown that plasma BNP levels are affected by anthropometric factors such as body mass index [16] and genetic features [17], and this may alter the utility of plasma BNP measurement as a predictor of cardiovascular events.

These suggest that the relationship between plasma BNP levels and cardiovascular outcomes should be evaluated separately in men and women, and it may also be important to examine whether the relationship is applicable in other ethnic populations. The present study has therefore sought to determine whether 1) plasma BNP levels are associated with an increased risk of CHF and any-cause death in both sexes in the general population; and 2) the relationship between plasma BNP and cardiovascular outcome observed in the white population is valid in other ethnic groups, specifically the Japanese population.

1. Methods

1.1. Study population

The original cohort of the Iwate-KENCO study was recruited from the community-dwelling population living in the three districts (Ninohe, Kuji, and Miyako) of the northern Iwate prefecture, Japan. This region has a resident population of over 144,000 adults over the age of 40 years based on census data from October 2005. The cohort was recruited from subjects of a government-regulated multiphasic health checkup for the general population. Invitations to participate in the multiphasic health screening program were issued by government offices in each municipality.

The total number of participants who agreed to join the Iwate-KENCO study in the three districts was 26,469 (original cohort). The acceptance rate for participation from government-regulated health screening was 84.5%. Of the original cohort living in Ninohe and Kuji districts (n=15,927), 15,394 subjects (97%) had BNP measurements (BNP cohort: men 5288; women 10,106). Subjects were excluded from this cohort for the following reasons: age under 40 (575), history of cardiovascular events such as myocardial infarction, stroke or heart failure (507), and missing covariates (846). The final statistical analysis was therefore performed on 13,466 subjects (men 4527; women 8939: Table 1). This study protocol was approved by our university ethics committee and local institutional review committees. All participants gave written informed consent.

1.2. BNP assay

Non-fasting blood samples were drawn from the antecubital vein while participants were seated. Blood samples were collected into vacuum tubes. While blood samples for routine blood testing were being taken, an additional 2 ml sample of venous blood was collected into a test tube containing EDTA-2Na for plasma BNP measurement. Tubes were stored immediately after sampling in an icebox and transported to the Iwate Health Service Association

Table 1
Baseline characteristics of study population, the Iwate KENCO study.

	Men	Women
Number	4527	8939
Age (years)		
Mean (±SD)	64.1±10.3	62.0±10.0
Median	66.0	63.0
Plasma BNP (pg/ml)		
1st quartile	<6.5	<8.9
2nd quartile	6.5–14.8	8.9–17.1
3rd quartile	14.8–30.0	17.1–30.4
4th quartile	≥30.0	≥30.4
Systolic blood pressure (mmHg)		
Mean (±SD)	130.1±19.4	125.5±19.7
Median	128.5	123.5
Diastolic blood pressure (mmHg)		
Mean (±SD)	77.8±10.8	73.7±10.8
Median	77.5	73.0
Hypertension (%)	44.4	38.8
Antihypertensive drugs (%)	23.6	24.3
Body mass index (kg/m2)		
Mean (±SD)	23.9±2.9	24.1±3.4
Median	23.8	23.9
Atrial fibrillation (%)	3.0	0.6
HbA1c (%)		
Mean (±SD)	5.2±0.8	5.1±0.6
Median	5.0	5.0
Diabetes (%)	8.0	4.3
Antidiabetic medication (%)	4.6	2.4
Total cholesterol (mg/dl)		
Mean (±SD)	193.2±32.6	206.7±31.8
Median	191.0	206.0
Hypercholestrolemia (%)	10.3	20.3
Antihypercholestrolemic drugs (%)	2.8	7.3
HDL-cholesterol (mg/dL)		
Mean (±SD)	56.4±15.4	61.8±14.5
Median	54.0	60.0
Current smoking (%)	33.4	2.5
Regular alcohol intake (%)	61.7	11.1
Regular exercise (%)	17.0	10.5

laboratory each afternoon. They were then centrifuged at 1500 g for 10 min. After separation, plasma samples were stored frozen at −20 °C until transportation to the Shionogi central laboratory for the assay. Plasma BNP levels were measured by direct radioimmunoassay using monoclonal antibodies specific for human BNP (Shiono RIA BNP kit, Shionogi, Japan). Cross-reactivity of the antibody was 100% for human BNP and 0.001% for human atrial natriuretic peptide. Intra- and inter-assay coefficients of variation were 5% and 6%, respectively.

1.3. Risk factor definitions

Subjects used a self-reported questionnaire to document medical history including status (yes or no) of prescribed drugs for hypertension, diabetes, hypercholesterolemia, stroke, angina, CHF and myocardial infarction. Smoking habits (current or non-smoker), regular alcohol intake (yes or no), and regular exercise (≥60 min of exercise and ≥8 times per month) were also assessed by a questionnaire developed by the study committee. Systolic and diastolic blood pressures were determined with an automated sphygmomanometer with subjects seated for at least 5 min before measurement. Measurement was performed twice, with the mean value used for statistical analysis. Hypertension was defined as systolic blood pressure ≥140 mm Hg and/or diastolic blood pressure ≥90 mm Hg, and/or the use of antihypertensive medication. Body height was measured with participants in stocking feet and weight was measured wearing light clothing. Body mass index was calculated as weight (kg) divided by the square of height (m^2). Diabetes was ascertained by detection of a non-fasting glucose concentration ≥200 mg/dl and/or HbA1c value ≥6.5% [18] and/or a use of anti-diabetic agents including insulin. Hypercholesterolemia was defined as a serum concentration ≥240 mg/dl and/or the use of anti-lipidemic medications.

1.4. Outcome

A follow-up survey assessing mortality, migration, and the incidence of CHF was carried out after the baseline study. Admission cases of CHF in the cohort were checked by the regional registration survey data for heart diseases, which records primary hospital discharge diagnoses in the study area. Details of this register have been described previously [15]. The cases of CHF were objectively defined by Framingham criteria [19]. All deaths and migration were confirmed by the official resident registration data issued by the local government offices.

1.5. Statistical analysis

Continuous variables are shown as median or mean. Owing to the purpose of the present study, the following analyses were performed separately in men and women. Participants were divided into quartiles according to their baseline plasma BNP levels. Survival from entry into the study was estimated using the Kaplan–Meier method, followed by a trend test (Log rank). We evaluated the association between baseline plasma BNP levels and two clinical endpoints (new onset of CHF and death from any cause). Using a Cox proportional hazards regression model, hazard ratios (HRs) for plasma BNP with each outcome were assessed. In these analyses, plasma BNP levels were used as a categorical variable and as continuous variables after natural logarithmic transformation. For the categorical analyses, we used prespecified thresholds corresponding to the 75th percentile values. In this multivariable proportional-hazards regression model, we adjusted analyses for age, body mass index, and the presence or absence of hypertension, diabetes, hypercholesterolemia, current smoking, regular exercise and atrial fibrillation. Receiver-operating-characteristic (ROC) curves were constructed to assess the sensitivity and specificity of plasma BNP throughout the range of concentrations as an indicator of CHF or all cause of mortality. The area under the curve and 95% confident interval (CI) of each ROC curve were calculated to provide a measure of the overall diagnostic accuracy of the test. All statistical analysis was performed using SPSS software. A significant difference was defined as $p<0.05$.

2. Results

2.1. Baseline characteristics of the cohort

The median age of male and female cohorts was 66 in men, and 63 in women, respectively (Table 1). The number of women participants was approximately twice the number of men. The median plasma BNP level was higher in women than men (17.1 pg/ml versus 14.8 pg/ml; $p<0.001$). The prevalence of hypertension, atrial fibrillation, diabetes, current smoking, regular alcohol intake tended to be higher in men than in women.

2.2. Congestive heart failure and all-cause death

During the 2.9 year follow-up period, there were 44 cases of new onset CHF (men=23; women=21). The crude incidence rate of CHF was 1.75/1000 person-years in men and 0.82/1000 person-years in women. In addition, a total of 182 participants died from any cause (men=106; women=76). The crude death rate was 8.07/1000 person-years in men and 2.98/1000 person-years in women. The event free probabilities for CHF according to BNP quartiles are shown in Fig. 1. In both sexes, the highest quartile showed the lowest event free rate for onset of CHF (p for trends: men, $p<0.0001$; women, $p<0.0001$). Similarly, mortality rate increased with increasing quartile levels of plasma BNP (p for trends: men, $p<0.0001$; women, $p=0.0014$) (Fig. 2).

2.3. Multivariate analysis of outcomes

Increased plasma BNP levels predicted new onset of CHF even after adjusting for cardiovascular risk factors such as age, atrial fibrillation, BMI, current smoking, diabetes, hypercholesterolemia, hypertension, regular alcohol intake, and exercise habit in both sexes. As shown in Table 2, male participants with plasma BNP values above the 75th percentile had 13.4-fold increased risk of onset of CHF (95% CI, 4.1 to 43.6; $p<0.001$) relative to those with values equal to or below. In women, the association between the risk of CHF and plasma BNP above the 75th percentile was also significant (hazard ratio=8.5; 95% CI, 2.9 to 25.3: $p<0.001$). When plasma BNP was analyzed as a continuous variable, increasing plasma BNP for each 1SD increment in log BNP was associated with an increased risk of onset of CHF, with adjusting hazard ratios of 8.8 (95% CI, 3.9 to 20.1; $p<0.001$) in men and 6.7 (95% CI, 2.9 to 15.3; $p<0.001$) in women.

Median plasma BNP levels were higher in subjects complicated with atrial fibrillation than in subjects in sinus rhythm (men, 106.0 pg/ml versus 14.3 pg/ml; $p<0.001$: women, 118.0 pg/ml versus 17.0 pg/ml; $p<0.001$). To eliminate possible confounding effects of atrial fibrillation on the onset of CHF and plasma BNP, participants with atrial fibrillation at baseline were excluded from the analysis. The relationship between plasma BNP levels above the 75th percentile (categorical variable) and risk of CHF onset remained robust in both men (hazard ratio=15.5; 95% CI, 4.5 to 53.9: $p<0.0001$) and women (hazard ratio=7.9; 95% CI, 2.6 to 23.9: $p<0.001$) (Table 2). The risk of CHF for each 1SD increment in log BNP increased 12.8-fold in men (95% CI, 5.4 to 30.5; $p<0.0001$), and 7.5-fold in women (95% CI, 3.2 to 17.5; $p<0.001$) after exclusion of subjects with atrial fibrillation at baseline.

In men, the hazard ratio for mortality according to plasma BNP as a categorical variable (above the 75th percentile) was 1.8 (95% CI, 1.2 to 2.7; $p=0.005$). Also, a 1SD increment in plasma BNP as a log transformed value was associated with a significant (1.4-fold) increase in the hazard ratio for death (95% CI, 1.0 to 1.8; $p=0.024$) in men (Table 2). However, in women, the association between plasma BNP and mortality was not significant (above the 75th percentile, $p=0.41$: each 1SD increment, $p=0.23$).

2.4. ROC analysis

As shown in Fig. 3, the overall power of plasma BNP for prediction of CHF was significant. The optimal threshold of BNP for prediction of CHF was 32 pg/ml (sensitivity; 83%, specificity; 77%) in men, and 62 pg/ml

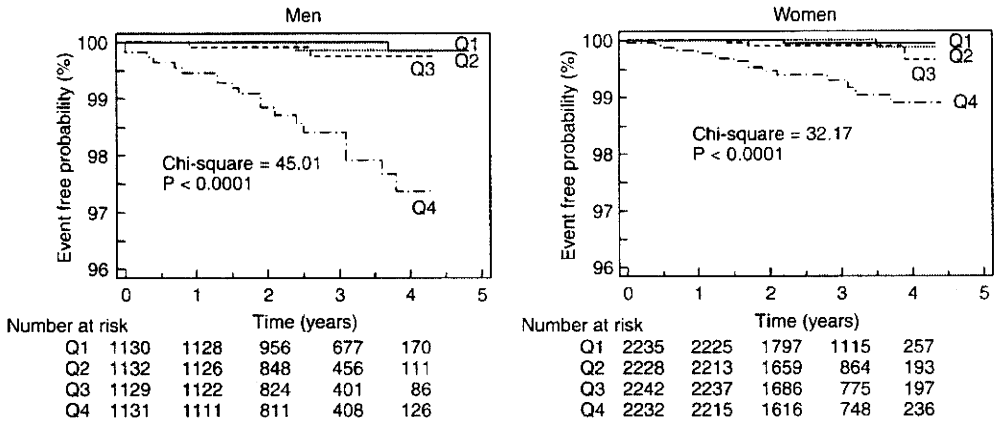

Fig. 1. Kaplan–Meier curves of unadjusted cumulative CHF free probabilities according to quartiles of plasma BNP at baseline in the general population. Q = quartile.

(sensitivity; 67%, specificity; 94%) in women, respectively. The area under the ROC curve was 0.853 (95% CI, 0.842 to 0.863) in men, and 0.838 (95% CI, 0.830 to 0.845) in women, respectively. The predictive ability of plasma BNP for all-cause of death as represented by the area under the curve was lower than that for CHF (men; 0.665, women; 0.615).

3. Discussion

The present study has found that elevated plasma BNP in the middle aged and elderly general population serves as a significant indicator of high risk for future onset of CHF in both men and women. This relationship remains statistically robust even after adjustment for clinical risk factors for CHF and after exclusion of subjects having atrial fibrillation at baseline. In addition, increased plasma BNP is a useful biomarker for prediction of high risk for any cause mortality in men, whereas this relationship was obscure in women.

Wang et al. have reported that elevated levels of plasma BNP were a useful predictor of the risk of death and cardiovascular events including CHF in a mainly white US population living in Framingham [10]. The incidence of CHF is well known to be lower in women than in men. Conversely, median plasma BNP levels are reported to be higher in women than in men in the general population [12–14]. This appears to contradict the epidemiological fact of a lower prevalence of cardiovascular disorders in women. However, little information is available to show whether the relationship between plasma BNP and the risk of onset of CHF remains significant in both sexes. Subject numbers in these previous studies may have been insufficient for separate analysis of predictive values for CHF in each gender group. The present study has suggested for the first time that plasma BNP may be a feasible screening tool for identification of individuals at high risk of future development of CHF within an apparently healthy population without gender bias.

Moreover, there have been no reports about the predictive abilities of plasma BNP for any cardiovascular events in non-white populations having a different incidence of cardiovascular disease. The incidence of cardiovascular events including CHF differs among races, with the Japanese having a relatively lower rate [15,20–22]. Plasma BNP levels have been demonstrated to be affected by anthropometric factors such as body mass index [16], and to be modified by heritability and genetic factors in a community based sample [17]. These findings suggest that the distribution of plasma BNP in apparently healthy populations may differ among

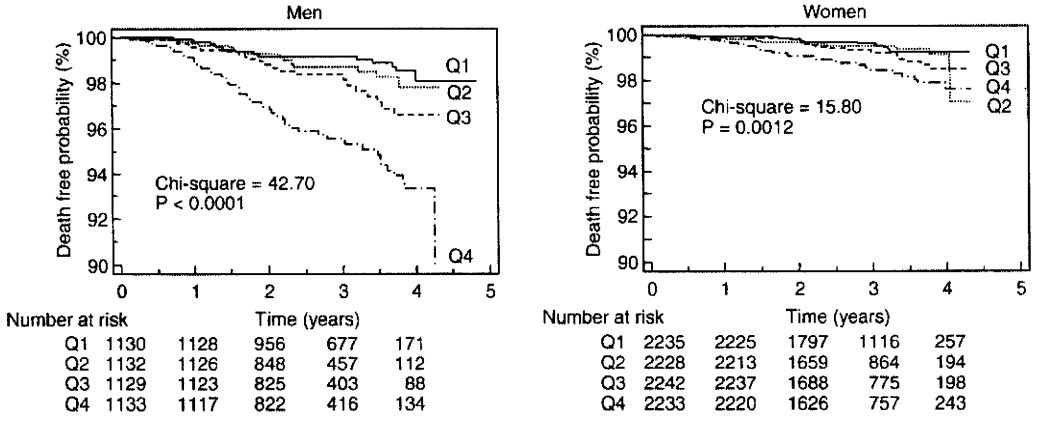

Fig. 2. Kaplan–Meier curves of unadjusted cumulative survival according to quartiles of plasma BNP at baseline in the general population. Q = quartile.

Table 2
Multivariate analysis of the association of plasma BNP levels, congestive heart failure and death.

	Adjusted hazard ratio for BNP values above 75th percentile			Adjusted hazard ratio per 1SD increment in Log BNP		
	HR	95%CI	*P* value	HR	95%CI	*P* value
Men						
Heart failure	13.45	4.15 to 43.56	0.001	8.84	3.88 to 20.13	0.001
Heart failure (ex. Afib)	15.50	4.46 to 53.88	0.001	12.85	5.41 to 30.51	0.001
All cause death	1.81	1.20 to 2.75	0.005	1.38	1.04 to 1.82	0.024
Women						
Heart failure	8.54	2.88 to 25.31	0.001	6.68	2.93 to 15.26	0.001
Heart failure (ex. Afib)	7.88	2.60 to 23.91	0.001	7.53	3.24 to 17.53	0.001
All cause death	1.22	0.76 to 1.98	0.408	1.17	0.90 to 1.53	0.231

The hazard ratios were adjusted for age, atrial fibrillation, BMI, current smoking, diabetes, hypercholesterolemia, hypertension, regular alcohol intake, and regular exercise: ex. Afib = analysis after exclusion of atrial fibrillation.

ethnicities and communities. It would therefore be important to confirm that the predictive ability of BNP for CHF reported in US and European populations could be extrapolated to other ethnic populations. The present study has established the relationship between plasma BNP levels and risk of CHF in a non-white, specifically Japanese, population. Thus the present study suggests that BNP testing may be useful even in a low-risk population.

In contrast to the value of plasma BNP for predicting future onset of CHF in either sex, the association between plasma BNP and all cause mortality was less robust after adjustment for cardiovascular risk factors especially in the female cohort. In several previous studies without gender-specific analysis, elevated plasma BNP or plasma NT-proBNP levels were associated with an increased risk of death in the general population [10,11,23,24]. The present study has confirmed this association in men only, with the reasons for the lack of predictive ability of BNP testing for all causes of death in women remained unknown. Although person-years among female subjects in the present study may not have been insufficient (>=25,000 person-years), the follow-up may have been shorter than those of earlier studies (>5 years). In addition, it seems that cardiovascular death rate among the present female cohort may have been lower due to the lower incidence of cardiovascular risk factors compared to the male cohort. These biases may account for the possible dissociation between mortality and plasma BNP in women.

3.1. Limitations

This study has several limitations. The capture of CHF during the follow-up period was restricted to hospitalized cases so that CHF patients treated at an outpatient clinic only may be missing from the follow-up data. As the Framingham criteria for CHF employed in this study tend to capture relatively advanced CHF, the observed predictive value of plasma BNP is assured in cases with clinically overt CHF. As echocardiographic parameters were not included in the baseline data, the reasons for the elevation of plasma BNP are not known. However, according to our previous cross-sectional cohort study of the general population, elevated plasma BNP concentrations has a significant sensitivity and specificity for screening several phenotypes of structural heart disease [8]. The predictive abilities of high plasma BNP levels may be due to the capability for selection of subjects who have underlying cardiac disorders which are prone to progress to overt CHF. More than 20% of our cohort was receiving antihypertensive agents at baseline. Several types of angiotensin converting enzyme inhibitors or angiotensin II receptor blockers have been reported to reduce cardiovascular events and mortality in high risk subjects [25,26]. This type of drug also has been reported to reduce plasma levels of BNP [27]. In view of these findings, the present study did not assess the effects of these drugs on the incidence of outcomes or on plasma BNP levels. However, when the use of antihypertensive medication (yes or no) was entered into the multivariate regression model, the significance of the predictive ability of plasma BNP did not weaken for CHF (hazard ratios >5.0, $p<0.0001$; both above the 75th percentile level and each 1SD increment).

In conclusion, measurement of plasma BNP provides strong predictive information about future onset of CHF in both sexes, with the predictive ability for death effective especially in men.

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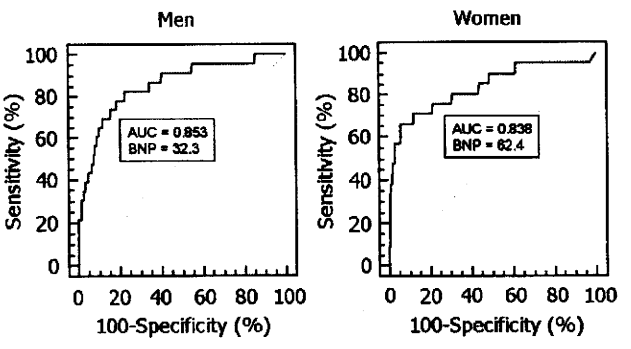

Fig. 3. Receiver-operating-characteristic curves of plasma BNP concentration to predict future onset of CHF.

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The authors of this manuscript have certified that they comply with the Principles of Ethical Publishing in the International Journal of Cardiology [28].

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ORIGINAL ARTICLE
Renal Disease

Plasma B-Type Natriuretic Peptide Level and Cardiovascular Events in Chronic Kidney Disease in a Community-Based Population

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Background: Plasma B-type natriuretic peptide (BNP) levels are confounded by renal dysfunction, so this study examined whether plasma BNP might be a reliable biomarker of the onset of cardiovascular (CV) events in a population-based cohort with impaired renal function.

Methods and Results: Baseline data, including plasma BNP, serum creatinine, and urinary protein levels, were determined in participants from a community-based population. Estimated glomerular filtration rate (eGFR) was calculated, and chronic kidney disease (CKD) was defined as either: eGFR <60 ml·min^{-1}·1.73 m^{-2} and/or proteinuria (CKD definition-1) or GFR <60 ml·min^{-1}·1.73 m^{-2} (CKD definition-2). The CV endpoint was surveyed prospectively. The cohorts were followed for 5,275 person-years for CKD definition-1, and for 4,350 person-years for CKD definition-2. The CV event-free survival rate in the highest BNP quartile in either CKD definition was the lowest among the quartile groups (P<0.001). In multivariate Cox regression models adjusted by traditional CV risk factors and atrial fibrillation, relative risk (RR) for CV events was significantly higher in the highest BNP quartile compared with the lowest BNP quartile (CKD definition-1, RR 3.51, P<0.01: CKD definition-2, RR 4.67, both P<0.01).

Conclusions: Plasma BNP level provides strong predictive information about the future onset of CV events in CKD subjects selected from the general population. (*Circ J* 2010; **74**: 792–797)

Key Words: General population; Heart failure; Renal failure; Stroke

Chronic kidney disease (CKD), defined as reduced glomerular filtration rate (GFR) and/or proteinuria, increases the risk of cardiovascular (CV) disease and endstage renal disease.[1] In population-based studies, the prevalence of CKD has been shown to be 7% in persons aged more than 30 years and to be increased 23–36% in persons aged more than 65 years.[2] The trend in the prevalence of CKD has been speculated to increase over time in line with the recent increasing prevalence of diabetes, obesity, and hypertension.[3] Several reports have emphasized that early identification and treatment of CKD are necessary to prevent serious outcomes in this disorder.[1,4] However, considering the large number of persons with CKD in the general population, it may not be easy to provide pharmacological and non-pharmacological interventions for all stages of CKD. In view of these limitations, it may be practical to select CKD subjects at relatively high risk for CV diseases from the general population, and then provide treatment to prevent their onset. However, there are no established markers to stratify CV risk in CKD subjects with mild renal dysfunction, such as stage 3 CKD, in the mass screening setting.

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Natriuretic peptide family proteins, including B-type natriuretic peptide (BNP) and its N-terminal fragment (NT-pro BNP), are released from the heart in response to increased intracardiac pressure, cardiac pump dysfunction, hypertensive ventricular hypertrophy, and myocardial ischemia. In community-based studies, increased circulating levels of BNP and NT-pro BNP have been reported to relate to a high risk of CV events and mortality.[5,6] The high prevalence of CV events in the group with elevated plasma levels of BNP and NT-proBNP is believed related to the high prevalence of subclinical heart disease. However, plasma concentrations of BNP and NT-proBNP increase as GFR declines in patients

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with and without apparent cardiac disorders.[7,8] In view of these facts, it is unclear whether plasma BNP levels would be a reliable biomarker for predicting CV events in the cohort of CKD selected from a community-based population.

CKD is usually defined by 2 biomarkers of renal function: urinary protein and reduced GFR. Several community-based studies have applied only the latter definition.[9–11] However, it is uncertain whether these biomarkers (GFR and urinary protein) provide complementary or overlapping information for CV risk. Cirillo et al reported that the use of only 1 of the biomarkers underscores the potential to misclassify patients as having or lacking CKD, thus misinterpreting the CV risk.[12] Therefore, the present study used 2 definitions of CKD to examine whether plasma BNP might be a reliable biomarker for predicting onset of CV diseases in a CKD cohort selected from a community-based general population.

Methods

Study Population

The original cohort of the Iwate-KENCO study was recruited from a community-based population living in Ninohe, Kuji, and Miyako districts of northern Iwate prefecture, Japan. The details of the recruitment and measurements of the cohort were shown in previous reports.[13,14] The total number of participants who agreed to join the Iwate-KENCO study in the 3 districts was 26,469 (original cohort). Of the original cohort living in Ninohe and Kuji districts (n=15,927), 15,394 subjects (97%) had BNP measurements (BNP cohort: men 5,288; women 10,106).

Subjects were excluded from the present analysis for the following reasons: age under 40 (n=575); history of CV events, such as myocardial infarction, stroke or heart failure (n=507); missing data of serum creatinine level (n=28), body mass index (n=47), ECG tracing (n=717), or blood pressure (n=4); estimated GFR <30 ml·min^{-1}·1.73 m^{-2} (n=28). The final statistical analysis included 13,526 subjects (men 4,542; women 8,984). The study protocol was approved by the university ethics committee and local institutional review committees. All participants gave written informed consent.

Definition of CKD

The eGFR was calculated using an equation from the Modification Diet in Renal Disease Study (MDRD) for the Japanese population.[15] A urine sample was obtained during a multiphase health examination and urinary protein was semi-quantitatively determined using a dipstick test (Uropaper alpha II, Eiken): proteinuria was defined as trace or more. CKD was defined in the present study in 2 ways: (1) eGFR <60 ml·min^{-1}·1.73 m^{-2} and/or proteinuria (CKD definition-1); (2) eGFR <60 ml·min^{-1}·1.73 m^{-2} (CKD definition-2).

Measurements

Blood samples were drawn from a peripheral vein while the subject was seated. When blood samples for routine blood testing were being taken, an additional 2 ml was collected into a test tube containing EDTA-2Na for plasma BNP measurement. Tubes were stored immediately in an icebox after sampling and transported to the laboratory each afternoon where they were centrifuged at 1,500 g for 10 min. After separation, plasma samples were stored frozen at −20°C until transportation to the Shionogi central laboratory for assaying (Osaka, Japan). Plasma BNP levels were measured by direct radioimmunoassay using monoclonal antibodies specific for human BNP (Shiono RIA BNP kit, Shionogi). Cross-reactivity of the antibody was 100% for human BNP and 0.001% for human atrial natriuretic peptide. Intra- and interassay coefficients of variation were 5% and 6%, respectively. Serum creatinine level was determined by an enzymatic method using an auto-analyzer (Hitachi 7700).

All subjects used a self-reported questionnaire to confirm their medical history, including status (yes/no) of prescribed drugs for hypertension, diabetes, hypercholesterolemia, stroke, angina, heart failure and myocardial infarction. Smoking status (current, past smoker or non-smoker) was also assessed by questionnaire.

Risk Factor Definitions

Systemic blood pressure was measured by experienced technicians. All subjects were seated for at least 5 min before measurement using an automatic device (BP-103i II, model 513000, Nippon Colin). Measurement was performed twice, with the mean value used for statistical analysis. Hypertension was defined as systolic blood pressure ≥140 mmHg and/or diastolic blood pressure ≥90 mmHg and/or the use of antihypertensive medication. Body mass index was calculated as weight (kg) divided by the square of height (m^2). Diabetes was ascertained by non-fasting glucose concentration ≥200 mg/dl and/or hemoglobin A_{1c} value ≥6.5% and/or use of antidiabetic agents including insulin. Hypercholesterolemia was defined as a serum concentration ≥240 mg/dl and/or the use of antilipidemic medications.

Outcome

A follow-up survey assessing mortality, migration, and the incidence of heart failure, acute myocardial infarction and sudden death, and stroke was carried out after the baseline study. All deaths and migrations were confirmed by the official resident registration data issued by the local government offices.

Admission cases of heart failure in the cohort were checked by the regional registration survey data, which records primary hospital discharge diagnoses in the study area. The cases of heart failure were objectively defined by the Framingham criteria.[16] Details of this register have been described previously.[17] The event of non-sudden fatal myocardial infarction was also based on hospital registration survey data. The diagnosis of acute myocardial infarction was based on the Monica study criteria.[18] Sudden cardiac death within 1 h of the onset of acute illness was examined using death records and then checked against medical records of the hospitals within the survey areas. Stroke registry was used for the outcome study.[19] Stroke was defined as a sudden onset of focal neurological deficit ≥24 h duration and confirmed by brain computed tomography or magnetic resonance imaging.

Statistical Analysis

Continuous variables are shown as mean±SD. CKD subjects were divided into quartiles according to their baseline plasma BNP levels. To compare results among quartiles, ANOVA or chi-square test was used as appropriate. Survival from entry into the study was estimated using the Kaplan-Meier method, followed by a trend test (log rank). The association between baseline plasma BNP levels and endpoint CV diseases (new onset of heart failure, acute myocardial infarction/sudden cardiac death, and stroke) was evaluated. Using a Cox proportional hazards regression model, hazard ratios (HR) for plasma BNP with CV events were assessed. In this multivariable proportional-hazards regression model,

Table 1. Clinical Characteristics by BNP Quartile in Each CKD Definition

	CKD (definition-1), BNP quartile and range					P value	CKD (definition-2), BNP quartile and range					P value
	Total	Q1 ≤11.2	Q2 11.3–22.7	Q3 22.8–42.9	Q4 ≥43.1		Total	Q1 ≤11.9	Q2 12.0–23.5	Q3 23.6–43.4	Q4 ≥43.6	
n	1,901	478	473	475	475		1,578	395	394	395	394	
Age (years)	67.9±9.0	62.7±9.4	67.0±8.1	69.0±7.8	72.8±7.3	<0.001	68.7±8.4	64.4±8.9	67.7±7.8	69.7±7.4	72.9±7.1	<0.001
M/F	727/1,174	220/258	161/312	155/320	191/284	<0.001	552/1,026	159/236	131/263	118/277	144/250	<0.02
BMI	24.5±3.4	25.0±3.3	24.6±3.4	24.0±3.3	24.2±3.4	<0.001	24.4±3.3	24.8±3.2	24.5±3.2	23.9±3.3	24.2±3.4	<0.002
eGFR (ml·min^{-1}·1.73 m^{-2})	57.4±12.7	61.8±16.5	57.2±11.4	55.1±10.6	55.4±10.6	<0.001	52.9±5.4	54.0±4.9	53.2±5.0	52.4±5.7	51.8±5.6	<0.001
Proteinuria (%)	22.7	28.2	20.9	17.1	24.6	<0.001	6.9	4.8	6.6	5.6	10.7	<0.01
Blood hemoglobin (g/dl)	13.6±1.5	14.2±1.4	13.6±1.3	13.3±1.3	13.4±1.6	<0.001	13.5±1.4	13.9±1.4	13.6±1.3	13.3±1.3	13.3±1.5	<0.001
Hypertension (%)	53.8	47.1	50.7	50.7	66.5	<0.001	52.8	45.8	51.5	48.6	65.2	<0.001
Antihypertensive drugs (%)	37	27	36	35	49	<0.001	38	29	37	34	50	<0.001
Hyperlipidemia (%)	19	28.5	18.4	15.8	13.3	<0.001	19.1	28.1	19.3	14.4	14.5	<0.001
Diabetes (%)	7.5	9.2	8	4.6	8.2	<0.05	5.3	3.5	6.6	3.5	7.6	<0.02
Smoking (%)	11.8	16.7	9.3	8.8	12.2	<0.001	9.1	10.6	8.1	7.6	9.9	0.396
Atrial fibrillation (%)	3.1	0.4	0.2	1.1	10.5	<0.001	2.9	0.5	0.3	1.0	9.9	<0.001

BNP, B-type natriuretic peptide; CKD, chronic kidney disease; BMI, body mass index; eGFR, estimated glomerular filtration rate.

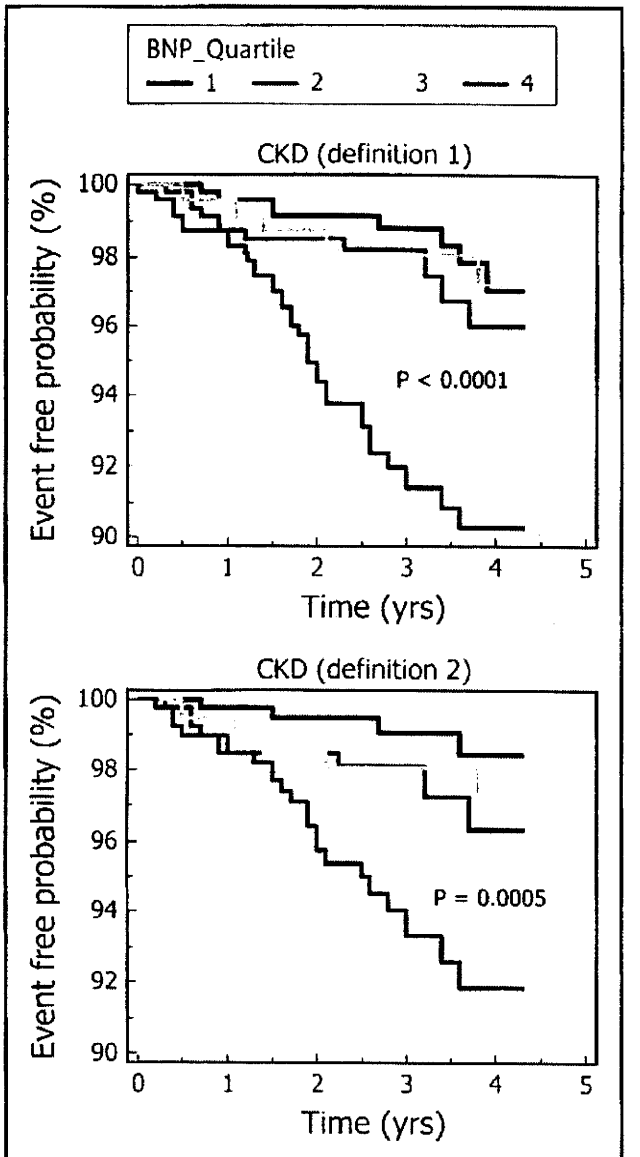

Figure. Kaplan-Meier curves of cardiovascular event-free probability according to quartile of plasma B-type natriuretic peptide (BNP) in chronic kidney disease (CKD) definition-1 (**Upper**) and CKD definition-2 (**Lower**).

adjustments were made in the analyses for age, body mass index, and the presence or absence of hypertension, diabetes, hypercholesterolemia, current smoking, and atrial fibrillation. For analyses of CV incidence, person-years were censored at the date of CV events, the date of emigration from the study area, the date of death or the end of the follow-up period, whichever came first. All statistical analyses were performed using SPSS software (Chicago, IL, USA). A significant difference was defined as $P<0.05$.

Results

As shown in **Table 1**, the number of cases of CKD definition-1 was 1,901 (727 in men, 1,174 in women). In this type of CKD, the prevalence within the community-based population was 14% (16% in men, 13% in women). The mean age was 67.9 years, and the mean eGFR was 57.4 ml·min^{-1}·

Table 2. Event Rates and HR for CVD by BNP Quartile in CKD

BNP quartile (pg/ml)	All CVD events/ 1,000 person-years	Age-sex adjusted HR (95%CI)	P value	Multivariate adjusted HR* (95%CI)	P value
CKD (definition 1)					
Q1 (≤11.2)	5.7	1.0		1.0	
Q2 (11.3–22.7)	8.6	1.77 (0.70–4.49)	0.226	1.83 (0.72–4.66)	0.203
Q3 (22.8–42.9)	7.1	1.47 (0.55–3.93)	0.439	1.62 (0.60–4.37)	0.341
Q4 (≥43.1)	25.9	4.71 (2.04–10.90)	<0.001	4.59 (1.97–10.73)	<0.001
CKD (definition 2)					
Q1 (≤11.9)	3.5	1.0		1.0	
Q2 (12.0–23.5)	8.4	2.58 (0.79–8.48)	0.118	2.48 (0.75–8.19)	0.135
Q3 (23.6–43.4)	7.7	2.39 (0.70–8.12)	0.164	2.56 (0.75–8.73)	0.134
Q4 (≥43.6)	20.3	5.56 (1.83–16.90)	<0.003	5.54 (1.81–16.97)	<0.003

*Adjusted for age, sex, BMI, current smoking, hypertension, diabetes, hypercholesterolemia, eGFR, and atrial fibrillation. HR, hazard ratios; CVD, cardiovascular disease; CI, confidence interval. Other abbreviations see in Table 1.

1.73 m^{-2}. Proteinuria was found in 23% of the subjects. The percentages of cases of hypertension, diabetes, and atrial fibrillation were 54%, 7.5%, and 3.1%, respectively. The median plasma BNP level was 22.7 pg/ml.

The number of cases of CKD definition-2 was 1,578 (552 in men, 1,026 in women), and the prevalence was 12% (12% in men, 11% in women) within the community-based population. The percentages of hypertension, diabetes, and proteinuria were 53%, 5.3%, and 6.9%, respectively. The prevalence of atrial fibrillation was 2.9%. The median plasma BNP level was 23.5 pg/ml (Table 1).

The cohorts were followed for 5,275 person-years in CKD definition-1, and for 4,350 person-years in CKD definition-2, respectively. Composite CV events (heart failure, acute myocardial infarction, sudden cardiac death, stroke) during the follow-up period (mean, 2.8 years) occurred in 62 cases in the CKD definition-1 group and in 43 cases in the CKD definition-2 group. The number of CV events/1,000 person-years was 11.7 and 9.9 in the CKD definition-1 and definition-2 groups, respectively.

Kaplan-Meier curves for the CV event-free rate according to the BNP quartiles in both CKD cohorts are shown in Figure. The CV event-free rate was significantly lower in the highest quartile of BNP (>43 pg/ml) in both CKD cohorts (CKD definition-1, P<0.0001; CKD definition-2, P<0.0005 by log-rank test).

As shown in Table 2, in the CKD definition-1 group, the number of CV events/1,000 person-years among BNP quartiles (Q) was 5.7 in Q1, 8.6 in Q2, 7.1 in Q3, and 25.9 in Q4. Similarly, in the CKD definition-2 group, the number was 3.5, 8.4, 7.7, and 20.3, respectively. CV events occurred in the highest quartile group of each CKD cohort (P<0.001 for both definitions).

After adjustment for age and sex, Cox regression analysis was performed to analyze the relationship between plasma BNP level and the risk of CV events (Table 2). The HR obtained from the Cox proportional model for the highest quartile of plasma BNP was significantly higher than that for the lowest quartile for CKD definition-1 (HR 4.71; 95% confidence interval (CI) 2.04–10.9; P<0.001) and for CKD definition-2 (HR 5.56; 95%CI 1.83–16.9; P<0.003). In addition, after multivariate adjustment of the models (age, sex, BMI, smoking, hypertension, diabetes, hypercholesterolemia, atrial fibrillation, and eGFR), similar results were obtained (Table 2). The HR in the highest quartile was significantly higher compared with the lower quartile groups (HR 4.59 in CKD definition-1 group, P<0.001; HR 5.54 in CKD definition-2 group, P<0.003).

Discussion

The present study demonstrates that for the first time in CKD cohorts defined by different criteria and selected from a community-based population, the subgroup with the highest plasma BNP quartile had a 4- to 5-fold higher CV risk, including heart failure, stroke, myocardial infarction, and sudden cardiac death compared with the subgroup with the lowest plasma BNP quartile. This relationship was robust even after adjustment for classical CV risk factors. Our observations suggest that the plasma BNP level is a useful tool for stratifying CV risk within a CKD cohort selected from a general population.

In cohort studies without renal dysfunction, Wang et al reported that the subgroup with plasma levels of BNP over the 80th percentile had a 3-fold higher risk of new onset of heart failure and a 2-fold higher risk of brain transient ischemic attack than subjects showing plasma levels below the 80th percentile.[5] Similarly, in a general population without subjects with elevated serum creatinine levels, Kistorp et al demonstrated that subjects who had higher plasma NT-proBNP levels above the 80th percentile had a 3-fold higher risk of CV diseases than the subjects who had plasma NT-proBNP levels below the 80th percentile.[6] However, no studies have yet examined whether plasma levels of natriuretic peptides might be effective for stratifying the CV risk within a large number of CKD subjects selected from the general population. This may have been because of concerns that plasma natriuretic peptide levels might increase in the absence of organic cardiac disorders, and thus confound the relationship between the plasma level and CV events in this setting, as the important clearance site of the natriuretic peptide family protein is the kidney.[20]

There are several possible explanations for the fact that an elevated plasma BNP level was associated with a high risk for CV events, as demonstrated in the present study. First, the increased level of plasma BNP might be a marker for more advanced renal dysfunction, and deterioration of renal function is usually associated with an accumulation of traditional CV risk factors[21] and there may be related increases in homocysteine, inflammation, oxidative stress, and thrombogenic factors.[1,4] These factors may impair endothelial function, lead to progression of atherosclerosis, and thus increase