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Underestimation of Risk Associations Due to Regression Dilution in Long-term Follow-up of Prospective Studies

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In prospective studies, disease rates during follow-up are typically analyzed with respect to the values of factors measured during an initial baseline survey. However, because of "regression dilution," this generally tends to underestimate the real associations of disease rates with the "usual" levels of such risk factors during some particular exposure period. The "regression dilution ratio" describes the ratio of the steepness of the uncorrected association to that of the real association. To assess the relevance of the usual value of a risk factor during particular exposure periods (e.g., first, second, and third decades) to disease risks, regression dilution ratios can be derived by relating baseline measurements of the risk factor to replicate measurements from a reasonably representative sample of study participants after an interval equivalent to about the midpoint of each exposure period (e.g., at 5, 15, and 25 years, respectively). This report illustrates the impact of this time interval on the magnitude of the regression dilution ratios for blood pressure and blood cholesterol. The analyses were based on biennial remeasurements over 30 years for participants in the Framingham Study (Framingham, Massachusetts) and a 26-year resurvey for a sample of men in the Whitehall Study (London, England). They show that uncorrected associations of disease risk with baseline measurements underestimate the strength of the real associations with usual levels of these risk factors during the first decade of exposure by about one-third, the second decade by about one-half, and the third decade by about two-thirds. Hence, to correct appropriately for regression dilution, replicate measurements of such risk factors may be required at varying intervals after baseline for at least a sample of participants. *Am J Epidemiol* 1999;150:341–53.

prospective studies; risk analysis; risk assessment

Much of our knowledge about the importance of risk factors such as blood pressure and blood cholesterol comes from prospective studies, in which various characteristics of a cohort are recorded at a "baseline" survey and the baseline characteristics of persons who subsequently develop a particular disease are then compared with those who do not (1, 2). However, because of the combined effects of measurement errors and longer-term fluctuations or changes within persons, this procedure does not reliably indicate the real association between the "usual" level of some risk factor during a particular period and the disease rate during that same (or some later) period (3–7). The uncorrected association between the baseline measurement of a risk factor and subsequent disease rates is generally shallower than the corrected association between the usual value of that risk factor during a particular period and disease (although exceptions are possible).

Replicate measurements of the risk factor in a reasonably representative sample of persons in a prospective study can be used to estimate the importance of, and correct for, this "regression dilution" effect (6). However, if the aim is to relate the disease rates to the usual risk factor levels during some much later exposure period, then corrections based on remeasurements made within just a few years of baseline may not take appropriate account of the effects of within-person variability over more prolonged periods. To assess the relevance of the usual levels of a risk factor during some particular exposure period (e.g., the second decade of follow-up) to disease risk, correction factors may need to be based on remeasurements made after an interval approximately equivalent to the midpoint of the relevant period (e.g., after about 15 years of follow-up).

This report describes appropriate ways of correcting prospective study results for regression dilution. Other aims were to 1) assess quantitatively the effects of regression dilution on blood pressure and cholesterol,

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according to interval of follow-up, by using repeated measurements of these factors during 30 years in the Framingham Study (1, 8); 2) determine, for these particular factors, whether regression dilution is importantly different for men and women or for people of different ages; 3) compare the findings from the Framingham Study after an interval of 26 years (i.e., the approximate midpoint of the third decade of follow-up) with those from the Whitehall Study of London civil servants after a similar interval (9–11); and 4) assess the extent to which, for these factors, the progressive increase in the importance of regression dilution with increasing duration of follow-up may be explained by the early deaths of those at highest risk.

MATERIALS AND METHODS

Study populations

The prospective Framingham Study initially included 5,209 men and women from Framingham, Massachusetts, who were aged 30–62 years when first examined between 1948 and 1952 (1, 8). Every 2 years during 16 consecutive surveys, the remaining participants were invited to have their blood pressure measured and a blood sample collected from a forearm vein. During each visit, a physician used a standard mercury sphygmomanometer to make two consecutive recordings of systolic and diastolic blood pressure, but only the first of the two measurements was used in our analyses. (Using the average of these two blood pressure measurements rather than just the first would have increased the correlations between measurements at different times by only about 0.03.) Total cholesterol was measured by using the colorimetric method of Sperry (12) during the first examination, but the Abell-Kendall method was used for all subsequent measurements (13).

The prospective Whitehall Study initially included 19,019 male civil servants from London, England, who were aged 40–69 years when first examined between 1967 and 1970 (9–11). During that baseline visit, a specially trained nurse made a single blood pressure measurement on the left arm by using the London School of Hygiene sphygmomanometer. Systolic blood pressure was recorded at the first appearance of the arterial flow sounds and diastolic blood pressure was recorded at both the muffling (phase IV) and the disappearance (phase V) of the sounds, but only the phase IV values were used in our analyses. During this first examination, blood samples were collected from earlobes into 2 mm glass tubing, and total cholesterol was measured by using a Technicon method (14); those values are thought to have been about 10 percent too low because the collection method used caused dilution of samples (10, 11).

In 1995, after an average interval of 26 years, a random sample of 401 participants in the Whitehall Study not known by the Office of National Statistics (London, England) to have died was selected for resurvey (15). Seventy-three percent completed a postal questionnaire; 61 percent had their blood pressure recorded twice after 5 minutes in the seated position, with a 2-minute interval between readings, and blood taken from a forearm vein at the office of the family physician. Whole-blood samples were mailed to the study laboratory (77 percent arrived within 24 hours and 92 percent within 48 hours), where total cholesterol was measured with a Beckmann auto-analyzer (Beckman Inc., Fullerton, California) by using an enzymatic method. Transport studies have shown that blood cholesterol levels can be measured reliably from samples collected in this way (16).

Statistical methods

Consider some adverse outcome, such as mortality from a certain disease during a particular time period (the "risk period"), whose rate of occurrence is to be related to the "usual" level of a certain risk factor during some particular time period (the "exposure period"). The risk period may well be the same as the exposure period, but it does not have to be: for example, mortality rates during the second decade of follow-up could be related to usual levels of the risk factor during the first decade. The magnitude of the regression dilution associated with a particular exposure period can be estimated either nonparametrically or parametrically from pairs of measurements made in the same persons on different occasions separated by an appropriate time interval. For example, to infer the real associations between the usual level of some particular exposure during the first decade of follow-up and disease rates (during the first decade or some later period), correction factors could be based on pairs of measurements separated by about 5 years (i.e., the length of time from baseline to the midpoint of the exposure period).

With the nonparametric method of estimating and correcting for the regression dilution associated with a particular exposure period, pairs of measurements of the relevant risk factor that are separated by an appropriate interval (e.g., about 5 years if the exposure period is the first decade) are subdivided into a few groups according to the value of just the first measurement in each pair. The range of the initial values (r_i) is defined as the difference between the means of these first measurements in the groups with the lowest and highest values. Even though the value of the second measurement in each pair did not determine in which group that pair belonged, the means of the second

measurements provide unbiased estimates of the "usual" levels of the risk factor in each group during the particular exposure period, against which the disease rates can be compared. Generally, the range of these mean usual values (r_u) will be substantially narrower than the range of the initial values (r_i), so the ratio of these two ranges ($R = r_u/r_i$) will be substantially less than 1. This "regression dilution ratio" provides an assumption-free estimate of the importance of regression dilution during the exposure period that is valid no matter what the sources of variation might have been over the particular time period (e.g., random error, measurement error, selective survival, or the "horse-racing" effect (17)) and is approximately independent of the number of groups chosen and of the boundaries used to define these groups. Moreover, the regression coefficient relating disease risk to the usual levels of the factor during the exposure period can be estimated as $1/R$ times the "uncorrected" regression coefficient relating risk to the baseline levels of the risk factor (6).

Under special circumstances, "parametric" statistical methods can also be used to estimate the regression dilution ratio. For example, when the variances of the recorded values at baseline and at remeasurement are similar (as for blood pressure and cholesterol in our study; refer to Results), the correlation coefficient between pairs of measurements separated by a particular interval (i.e., the "self-correlation") is about the same as the ratio of the ranges for that interval (6). Estimates of the importance of regression dilution that are based on self-correlations tend to have slightly less random variation than those based on the ratios of ranges and so may be preferred when only a limited amount of data is available (as in various subgroups by gender, age, and year of first measurement in our analyses). However, since the nonparametric method requires no assumptions about constant variances, the ratio of the ranges may generally be more appropriate than self-correlation coefficients for estimating the magnitude of regression dilution during longer-term follow-up.

Because measurements were made every 2 years in the Framingham Study, it was possible to classify measurements for each participant into multiple pairs separated by the same time interval. For example, to calculate the regression dilution ratio from measurements separated by a 6-year interval (i.e., the approximate midpoint of the first decade), up to 13 pairs of measurements per person were constructed by using data from the first (i.e., baseline) and fourth (i.e., year 6) examinations, the second and fifth, the third and sixth, and so on, up to the 13th and 16th examinations. Thus, the 5,209 persons initially in the Framingham Study contributed 44,000 pairs of measurements with a 6-year interval between them for assessment of regression dilution during the first decade of exposure. Pairs separated by 16- and 26-year intervals (i.e., the approximate midpoints of the second and third decades of exposure) were constructed in a similar way. (A missing value in either one of a pair of measurements resulted in a missing value for that pair.)

To explore the influence of gender, age, and secular trends on the regression dilution ratio, the self-correlation coefficients for blood pressure and blood cholesterol in the Framingham Study were calculated separately for men and women, for different age groups at the time of first measurement (ages 30–39, 40–49, 50–59, 60–69, and 70–79 years), and for pairs in which the first measurement was made in the first, second, and third decade of follow-up (i.e., at examinations 1–5, 6–10, and 11–15). The impact of early death and of antihypertensive medication use on the regression dilution ratios for blood pressure and blood cholesterol was investigated by considering separately those who survived to at least year 26 and those who did not receive such treatment. Estimates of the regression dilution ratio associated with the third decade of follow-up were also calculated from the Whitehall Study by comparing the baseline measurements made in 1967–1970 with the repeat measurements made about 26 years later, in 1995, on the same persons.

RESULTS

Characteristics of the study populations

Table 1 provides the mean values of selected characteristics of 2,336 men and 2,873 women at baseline (year 0) and after 6, 16, and 26 years of follow-up in the Framingham Study and of 19,019 men at baseline and 243 men after 26 years of follow-up in the Whitehall Study. The increase in mean age with follow-up was slightly smaller than the increased duration of follow-up because of higher mortality among older persons. There was little change in mean cholesterol, height, and weight, but a modest decline occurred in mean diastolic blood pressure and, in the Whitehall Study, in mean systolic blood pressure. (However, the absolute values of the initial cholesterol measurements in the Whitehall Study might well have been somewhat too low; refer to Materials and Methods.)

Differences in blood pressure and cholesterol over time

Table 2 shows the mean values of blood pressure and cholesterol at baseline and after 6, 16, and 26 years of follow-up among men in Framingham who were aged 30–39, 40–49, and 50–59 years at the baseline survey

TABLE 1. Selected characteristics* of the study populations at baseline (year 0) and at later remeasurements in the Framingham and Whitehall studies

	Framingham Study: year of measurement				Whitehall Study: year of measurement	
	0	6	16	26	0	26†
No. of persons‡						
Men	2,336	2,240	1,929	1,458	19,019	243
Women	2,873	2,818	2,592	2,182		
Age (years)						
Men	44 (9)	50 (9)	59 (8)	67 (8)	52 (7)	75 (7)
Women	44 (9)	50 (9)	59 (8)	68 (8)		
Systolic blood pressure (mmHg)						
Men	136 (20)	132 (20)	138 (21)	135 (19)	136 (21)	131 (17)
Women	135 (24)	134 (25)	140 (24)	137 (21)		
Diastolic blood pressure (mmHg)						
Men	86 (12)	84 (12)	82 (11)	78 (10)	85 (14)	81 (12)
Women	84 (13)	83 (12)	82 (11)	76 (10)		
Total cholesterol (mmol/liter)						
Men	5.7 (1.1)	6.1 (1.1)	6.0 (1.1)	5.6 (1.0)	5.1 (1.2)	5.1 (1.3)
Women	5.7 (1.2)	6.2 (1.2)	6.5 (1.2)	6.2 (1.1)		
Height (m)						
Men	1.72 (0.07)			1.70 (0.07)	1.76 (0.07)	1.73 (0.07)
Women	1.59 (0.06)			1.57 (0.06)		
Weight (kg)						
Men	76 (11)	77 (11)	77 (12)	77 (12)	76 (11)	76 (11)
Women	64 (12)	65 (11)	65 (12)	64 (12)		
No. of paired comparisons‡						
Men		18,813	9,874	3,030		243
Women		25,210	13,751	4,492		

* Values expressed as total number or as mean (standard deviation).
† Mean values after 26 years in the Whitehall Study are based on responses by 243 persons to a pilot resurvey.
‡ Each person in the Framingham Study was to be seen biennially for 30 years and so could contribute up to 13 paired comparisons with a 6-year interval between measurements and up to 8 and 3 paired comparisons with intervals of 16 and 26 years, respectively.

(left side of table 2, all men; right side, all men with these measurements from all of these resurveys). Either for all men initially screened at baseline or for just those with measurements at all surveys, comparisons can be made between people of different ages at the same survey or between people in the same birth cohort at different surveys. However, none of these comparisons provides a simple description of the ways in which a person's blood pressure or cholesterol typically changes with age. For example, cross-sectional comparisons of all men who attended a particular survey (i.e., within columns in table 2) showed some differences between those in different age groups, but such comparisons were between different generations with different life experiences rather than between the same persons. Also, the prior effect of selective mortality among those with high blood pressure or cholesterol would have been greater for those aged 50–59 years at the time of the survey than for those aged 30–39 years (particularly if, as in the right half of table 2, attention was restricted to men still alive at year 26 of follow-up).

Comparisons within birth cohorts of all who attended the baseline survey with those who attended later surveys (i.e., within rows in the left half of table 2) might likewise have been biased by selective removal of those with high blood pressure or cholesterol. This bias can be avoided by restricting such comparisons to those persons who survived to attend all four surveys (i.e., within rows in the right side of table 2). However, even these longitudinal comparisons among the same persons might be biased by changes in measurement techniques (including subject habituation) and by differences over time in risk factor management.

Thus, in terms of the natural history of blood pressure, it was difficult to interpret the observation that among the 1,091 persons who attended all surveys, systolic blood pressure decreased slightly between years 0 and 6, increased by an average of 8 mmHg between years 6 and 16, and then remained constant between years 16 and 26, whereas diastolic blood pressure hardly changed between baseline and 16 years and had then decreased by an average of 5 mmHg at

TABLE 2. Effect of age on blood pressure and cholesterol at baseline (year 0) and at 6, 16, and 26 years of follow-up* among all men in the Framingham Study and among those men with these measurements from all four of the resurvey visits

	All men in the Framingham Study: year of measurement				Subset with all four measurements: year of measurement			
	0	6	16	26	0	6	16	26
	Systolic blood pressure (mmHg)							
No. of men	2,259	1,954	1,619	1,169	1,091	1,091	1,091	1,091
Age at baseline (years)								
30-39	130	127	132	133	129	126	131	133
40-49	135	133	140	138	132	129	138	138
50-59	141	138	144	138	132	131	141	138
	Diastolic blood pressure (mmHg)							
No. of men	2,259	1,954	1,619	1,169	1,091	1,091	1,091	1,091
Age at baseline (years)								
30-39	83	83	83	79	83	82	83	79
40-49	87	85	83	77	85	84	83	78
50-59	88	85	81	74	84	83	81	74
	Total cholesterol (mmol/liter)							
No. of men	1,440	1,908	1,583	628	628	628	628	628
Age at baseline (years)								
30-39	5.6	6.0	6.0	5.7	5.5	6.1	5.9	5.7
40-49	5.8	6.1	6.0	5.6	5.7	6.2	5.9	5.7
50-59	5.8	6.1	5.9	5.3	5.7	6.1	5.8	5.2

* Mean values at each follow-up visit are shown for each age group.

year 26. Likewise, among those with measurements from all surveys, it was difficult to interpret either the average increase of 0.5 mmol/liter in blood total cholesterol by year 6 or the later 0.6 mmol/liter decrease. These uncertainties about absolute values and trends do not, however, affect the epidemiologic comparisons of risk between persons of the same age at the same survey, which were the chief concern of this report.

Shrinkage of ranges with increasing intervals between measurements

In contrast to the relatively modest changes in the overall mean values of blood pressure and cholesterol measurements that occurred over time in the Framingham Study (table 2), large artifactual differences were observed in the mean values at subsequent follow-up of persons subdivided into five similar-sized groups according to the value of just the first of each pair of measurements (figure 1). In general, there was a sharp convergence by year 2 due to the combined effects of measurement error and of short-term fluctuations in the baseline measurements, which was followed by a more modest convergence over the next few decades. For example, the mean systolic blood pressure of persons who were in the top fifth initially (i.e., those with a systolic blood pressure value of 146 mmHg or more at the first measurement) declined from 162 mmHg at year 0 to 156 mmHg after 2 years, 155 mmHg after 6 years, 153 mmHg after 16 years, and 147 mmHg after 26 years (figure 1 and left side of table 3). Similarly, the mean systolic blood pressure of persons in the bottom fifth initially (i.e., those with a value of less than 118 mmHg at the first measurement) increased from 111 mmHg at year 0 to 117 mmHg after 2 years, 120 mmHg after 6 years, 126 mmHg after 16 years, and 130 mmHg after 26 years. The absolute difference, or range, between the mean values in the top and bottom groups declined throughout the three successive decades of follow-up from 51 mmHg at year 0 to 35 mmHg after 6 years, 27 mmHg after 16 years, and only 18 mmHg after 26 years. Similarly, for diastolic blood pressure and total cholesterol, there was a sharp change after 2 years, followed by more modest shrinkage in the absolute differences between the mean values in the top and bottom baseline-defined groups. After an interval of 26 years, the changes in these ranges were similar in the Framingham and Whitehall studies (table 3).

Changes in regression dilution ratios with increasing intervals between measurements

The convergent patterns in figure 1 (and table 3) illustrate the progressive increase over time in the importance of regression dilution for certain measurements. Table 4 provides nonparametric estimates of the

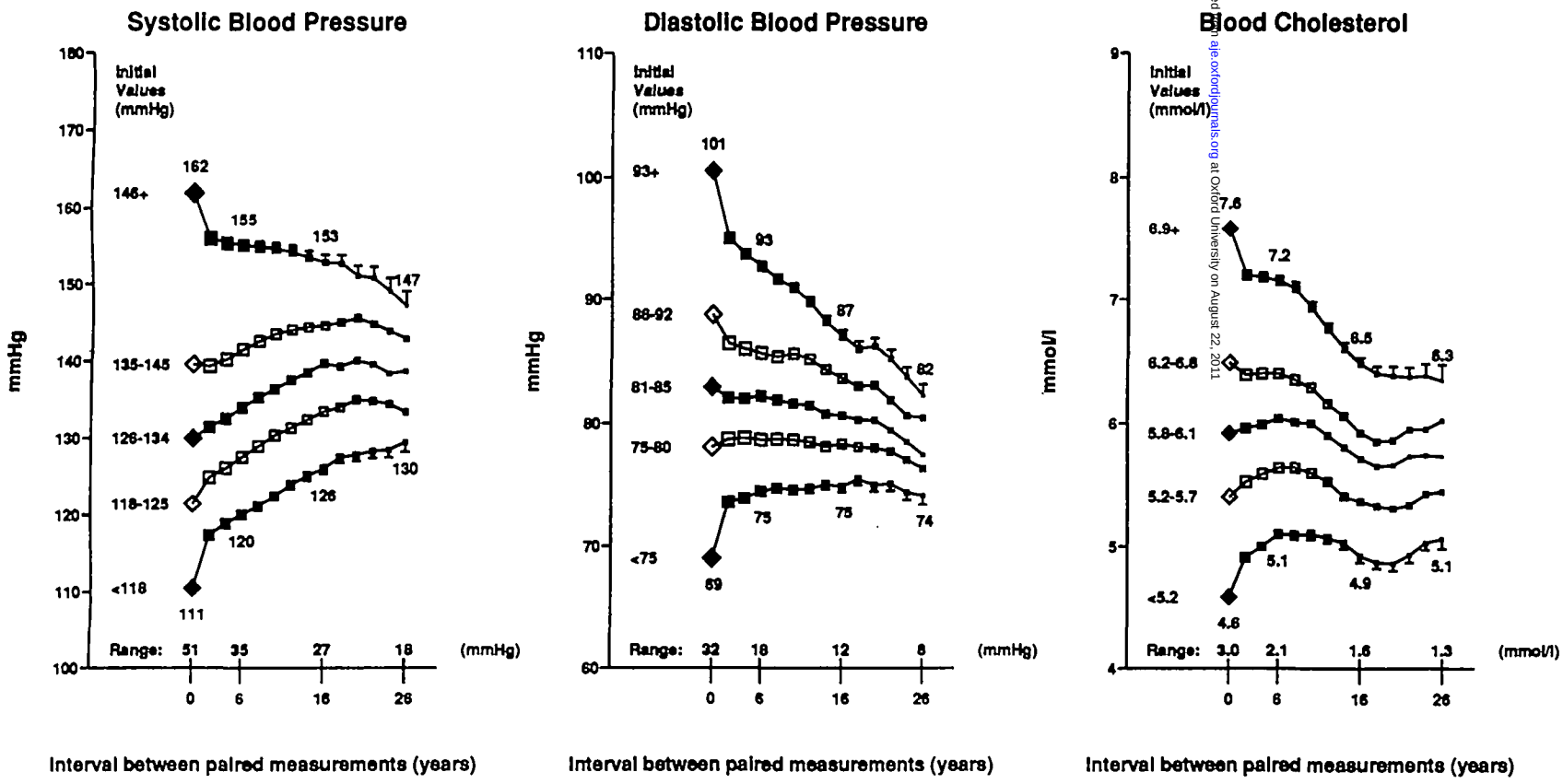


FIGURE 1. Serial shrinkage of the ranges for blood pressure and cholesterol values in groups defined by the first of a pair of measurements in the Framingham Study. Mean values initially and at subsequent follow-up are plotted for persons subdivided into five similar-sized groups according to the value of just the first of each pair of measurements. The size of each square is proportional to the quantity of statistical information available, and the 95% confidence intervals for the top and bottom groups are represented by the vertical bars. Mean values in the top and bottom groups, and the absolute differences (ranges) between them, are given at year 0 (i.e., the initial measurement of the pairs) and at years 6, 16, and 26 (i.e., the approximate midpoints of the first, second, and third decades, respectively, after the initial measurement).

TABLE 3. Mean values of blood pressure and cholesterol after different intervals between pairs of measurements among men in the Framingham and Whitehall studies

	Framingham Study: years between measurements				Whitehall Study: years between measurements	
	0	6	16	26	0	26
	Systolic blood pressure (mmHg)					
No. of measurements	18,813*	18,813	9,874	3,030	243	243
Baseline fifths						
I	111	120	126	130	112	141
II	122	128	134	133	124	146
III	130	134	140	139	134	152
IV	140	142	145	143	145	149
V	162	155	153	147	164	158
Difference (V-I)	51	35	27	18	52	17
	Diastolic blood pressure (mmHg)					
No. of measurements	18,813*	18,813	9,874	3,030	243	243
Baseline fifths						
I	69	75	75	74	68	77
II	78	79	78	76	77	82
III	83	82	81	77	83	84
IV	89	86	84	80	90	82
V	101	93	87	82	103	88
Difference (V-I)	32	18	12	8	35	11
	Total cholesterol (mmol/liter)					
No. of measurements	12,013*	12,013	5,604	1,687	239	239
Baseline fifths						
I	4.6	5.1	4.9	5.1	3.5	5.3
II	5.4	5.6	5.4	5.5	4.4	5.3
III	5.9	6.0	5.7	5.7	5.0	5.9
IV	6.5	6.4	5.9	6.0	5.7	6.0
V	7.6	7.2	6.5	6.4	6.9	6.2
Difference (V-I)	3.0	2.1	1.6	1.3	3.4	1.0

* Values for the Framingham Study are based on the means of multiple pairs of measurements with the same interval between measurements, grouped according to the value of the first measurement in each pair (refer to Materials and Methods). The values at year 0 are the means of the first measurements for those pairs with second measurements also available after a 6-year interval (and would differ slightly if based only on pairs with a 16- or 26-year interval).

regression dilution ratio derived from the ratio of the range of values in the baseline-defined groups after a particular interval to the range at baseline (refer to Materials and Methods). For blood pressure and cholesterol, the regression dilution ratios became substantially more extreme with increasing duration of follow-up, whereas the ratios for height and weight remained close to 1 (as might be expected, since these latter measures involve substantially less within-person variability). Table 4 also shows parametric estimates of the regression dilution ratios derived from correlation coefficients between pairs of measurements separated by a particular interval ("self-correlations"; refer to Materials and Methods). In general, the nonparametric and parametric methods yielded fairly similar results. For example, for systolic blood pressure in the Framingham Study, the regression dilution ratio associated with measurements made 16 years apart (i.e., the approximate midpoint of the second decade of exposure) was estimated to be 0.52 by using the ratio of the ranges and 0.45 by using the self-correlation. With either method, this value of about 0.5 implies that the regression coefficient relating risk in the second decade or later to a baseline measurement of systolic blood pressure is only about half as steep as the corrected regression coefficient relating this risk to the usual systolic blood pressure during the second decade.

Gender, age, and the regression dilution ratio

For systolic blood pressure, diastolic blood pressure (not shown), and blood cholesterol, the decrease in the

TABLE 4. Nonparametric and parametric estimates of regression dilution for blood pressure, cholesterol, height, and weight among men in the Framingham and Whitehall studies

	Framingham Study: years between measurements			Whitehall Study: years between measurements
	6	16	26	26
Systolic blood pressure				
Ratio of follow-up to baseline ranges	0.68	0.52	0.34	0.32
Correlation with initial measurement	0.63	0.45	0.31	0.26
Diastolic blood pressure				
Ratio of follow-up to baseline ranges	0.58	0.38	0.26	0.29
Correlation with initial measurement	0.57	0.38	0.27	0.30
Cholesterol				
Ratio of follow-up to baseline ranges	0.70	0.52	0.43	0.28
Correlation with initial measurement	0.68	0.53	0.46	0.38
Height				
Ratio of follow-up to baseline ranges	0.97	1.01	0.97	0.83
Correlation with initial measurement	0.96	0.97	0.96	0.84
Weight				
Ratio of follow-up to baseline ranges	0.93	0.85	0.79	0.86
Correlation with initial measurement	0.91	0.82	0.73	0.75

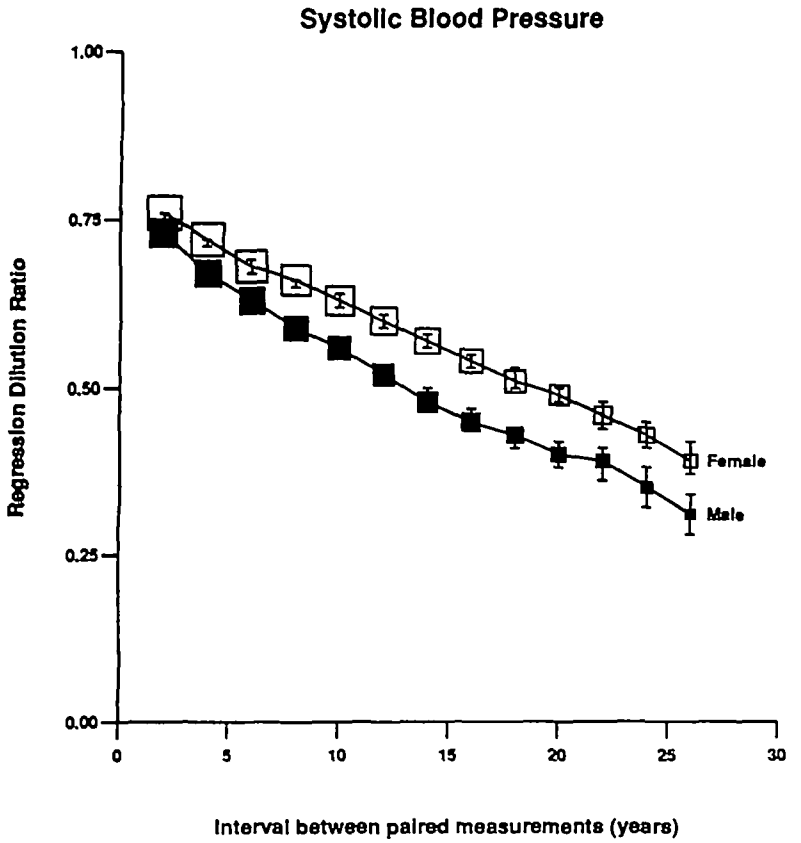
regression dilution ratio with increasing duration of follow-up was similar for men and women in the Framingham Study (figure 2). For example, the regression dilution ratios derived from the self-correlation coefficients for men decreased from 0.68 after 6 years to 0.53 after 16 years to 0.46 after 26 years; in women, they decreased from 0.68 to 0.52 to 0.42, respectively. A similar decrease in the regression dilution ratios with increasing duration of follow-up was also observed in the five 10-year age groups at baseline (figure 3). For example, the regression dilution ratio estimates for cholesterol after an interval of 6 years were 0.71 for those aged 30–39 years, 0.65 for those aged 40–49 years, 0.69 for those aged 50–59 years, 0.70 for those aged 60–69 years, and 0.71 for those aged 70–79 years. Similar patterns, but with slightly more random variation, were obtained by using regression dilution ratios derived from the ratios of ranges (data not shown).

Secular trends and the regression dilution ratio

For systolic blood pressure, the regression dilution ratio derived from the correlation coefficient between measurements made at 6-year intervals was 0.72 if the first of the two measurements was made in the 1950s (visits 1–5), 0.65 if made in the 1960s (visits 6–10), and 0.51 if made in the 1970s (visits 11–15). This finding suggests that there may have been a somewhat lower signal-to-noise ratio during the 1970s than during the 1950s (with, in this context, the signal being the extent to which people's usual blood pressures really differ from each other, and the noise being the extent to which particular measurements of blood pressure are liable to differ from that person's usual blood pressure). By contrast, there was no such trend in the regression dilution ratios for blood cholesterol: self-correlation coefficients between measurements made at 4-year intervals were 0.74 if the first of the two measurements was made in the 1950s, 0.72 if made in the 1960s, and 0.71 if made in the 1970s. (Cholesterol was not measured at visits 11, 12, or 16, so estimates of the regression dilution ratios for 6-year intervals could not be derived for the 1970s, but they were 0.72 for the 1950s and 0.68 for the 1960s.)

Early deaths, treatment, and the regression dilution ratio

Table 5 shows the extent to which estimates of the regression dilution ratio during the third decade of follow-up in the Framingham and Whitehall studies may have been influenced by differential mortality across the baseline-defined groups. The mean values at baseline for the total study population were compared with the mean values at baseline for those persons who were still alive after 26 years. For blood cholesterol, these mean values were similar in the top group and in the bottom group, whereas for systolic blood pressure the range was somewhat narrower among the survivors.



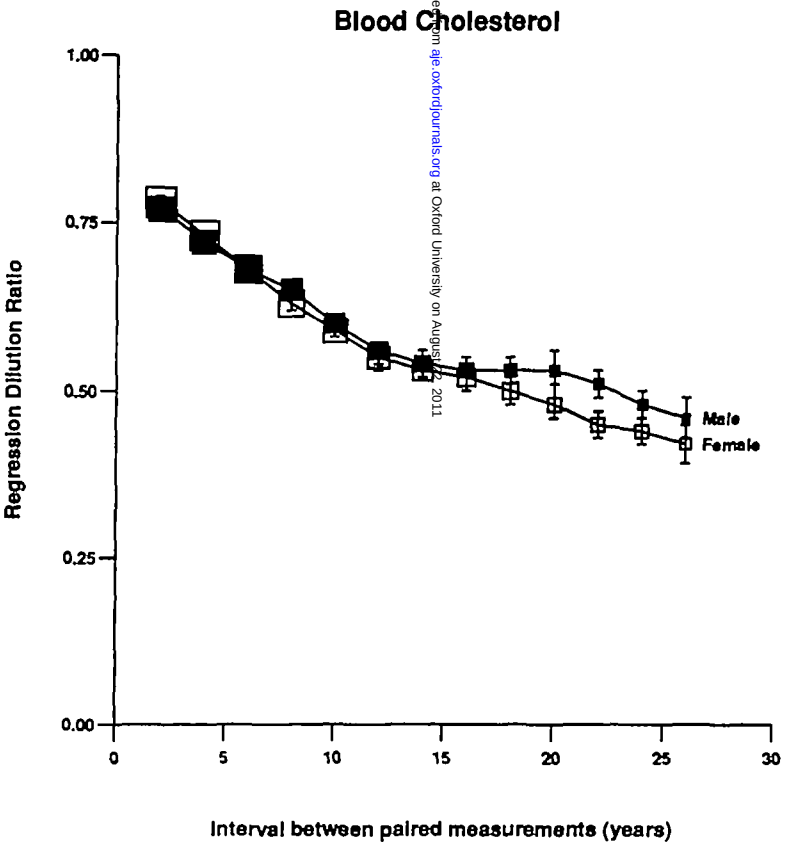


FIGURE 2. Serial changes in the regression dilution ratios for systolic blood pressure and blood cholesterol with increasing duration of follow-up among men and women in the Framingham Study. Parametric estimates of regression dilution ratios, derived from the correlation coefficients between pairs of measurements in persons (self-correlations), are plotted against the intervals between measurements. Symbols and conventions as shown in figure 1.

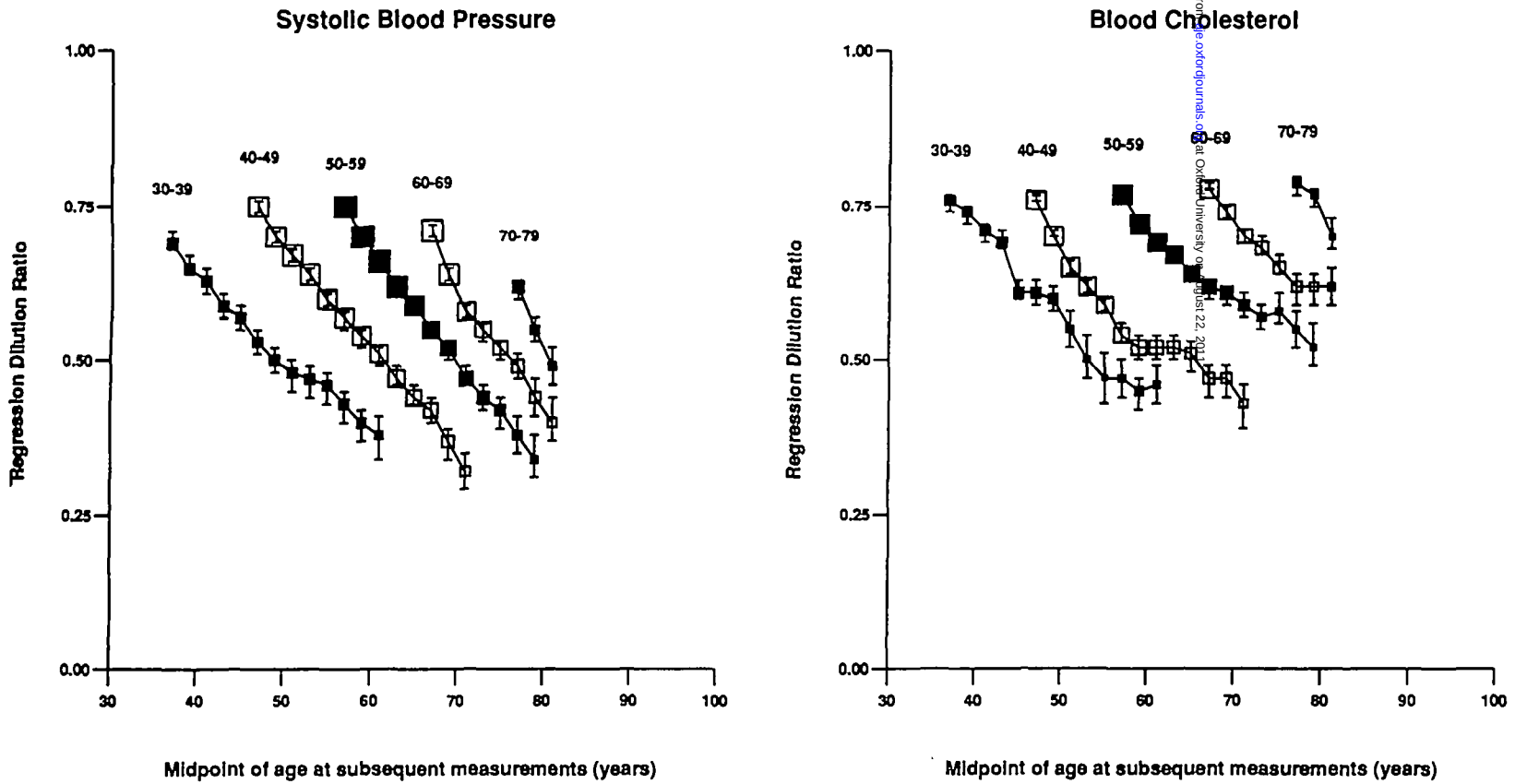


FIGURE 3. Serial changes in the regression dilution ratios for systolic blood pressure and blood cholesterol for participants at different ages in the Framingham Study. Parametric estimates of the regression dilution ratios, derived from the correlation coefficients between pairs of measurements in persons (self-correlations), are plotted against the estimated ages at the times of remeasurement. Symbols and conventions as shown in figure 1.

TABLE 5. Mean values of baseline measurements: influence of selective loss due to early deaths on systolic blood pressure and cholesterol among men in the Framingham and Whitehall studies

	Framingham Study		Whitehall Study	
	Total population	Survivors at year 26	Total population	Survivors at year 26
	Blood pressure (mmHg)			
No. of men	2,336	1,458	19,013	9,839
Baseline fifths				
I	112	111	111	111
II	123	121	125	124
III	132	129	134	134
IV	142	137	145	145
V	166	152	168	165
Difference (V-I)	54	42	57	54
	Total cholesterol (mmol/liter)			
No. of men	1,502	933	18,310	9,490
Baseline fifths				
I	4.3	4.2	3.5	3.5
II	5.1	4.9	4.4	4.4
III	5.7	5.5	5.0	5.0
IV	6.3	6.1	5.6	5.6
V	7.5	7.3	6.9	6.9
Difference (V-I)	3.2	3.1	3.4	3.4

However, these differences between the total population and the survivors were much less extreme than the convergence in the ranges shown in table 3, which suggests that selective mortality of persons with high blood pressure or cholesterol does not account for much of the shrinkage over 26 years.

In the Framingham Study, any treatment with a diuretic or other antihypertensive medication was recorded from year 6 onward. The proportion of survivors who received such treatment increased throughout the follow-up period, but treatment did not materially affect the progressive decrease in the regression dilution ratio for blood pressure with increasing duration of follow-up. For example, the estimates for systolic blood pressure, derived from self-correlations, of 0.64, 0.50, and 0.37 for all survivors after 6, 16, and 26 years, respectively, were similar to the corresponding values of 0.61, 0.47, and 0.33 for those survivors who did not receive any antihypertensive therapy.

Other factors and the regression dilution ratio

Although the multiple pairs of measurements from the Framingham Study are not independent, estimates of the regression dilution ratio derived from these multiple pairs were similar to those obtained when the baseline measurement was always the first of a relevant pair (i.e., when the data from any one visit were used only once). For example, for systolic blood pressure, the regression dilution ratio derived from the self-correlation at 6 years was 0.71 when year 6 was compared with baseline for 2,009 men, whereas the ratio was 0.63 when obtained from the 18,813 pairs with a 6-year interval between measurements (table 4). Estimates of the regression dilution ratio were also largely unaffected by the change in the method used to measure cholesterol after baseline in the Framingham Study. For example, among men, the correlation between pairs of measurements with a 6-year interval between them was 0.69 for year 6 compared with baseline (the only examination for which the Sperry method (12) was used; refer to Materials and Methods) and 0.71 for year 8 compared with year 2. Similarly, the estimates of the regression dilution ratio were unaffected by the initial decrease in mean systolic blood pressure in the Framingham Study (table 2). For example, among men, the correlation between pairs of measurements with a 6-year interval between them was 0.71 for year 6 compared with baseline and 0.68 for year 8 compared with year 2.

DISCUSSION

The present analyses addressed the extent to which within-person variability in risk factor measurements distorts assessment of the strength of the association

between the usual level of a risk factor during some particular exposure period and the incidence of disease during the same or a later period. Fluctuations in the values of risk factors may be due to measurement error and to short-term biologic variability (such as diurnal or seasonal variation), or they may be due to longer-term systematic changes. The effects of the former can be limited by repeating baseline measurements several times and by controlling the conditions under which measurements are taken. However, underestimation of the strength of the association of the usual level of a risk factor during some later exposure period with disease will still occur unless replicate measurements made some years later are used to correct directly for regression dilution. The effects of shifts in mean blood pressure or cholesterol due to long-term physiologic changes (such as the progressive increase in systolic blood pressure with age or the decrease due to the onset of disease or to the effects of treatment) are included in what is referred to as "regression dilution," as are the effects of selective mortality.

In our analyses of the Framingham and Whitehall studies, the progressively smaller ranges of mean values in the baseline-defined groups for blood pressure and cholesterol with longer intervals of follow-up (table 3) indicate that the effects of within-person variability become progressively more important with longer follow-up. It has been suggested that this finding might be due largely to selective death of those at highest risk (18), but analyses restricted to survivors in these studies indicated that only a small fraction of the shrinkage of these ranges could have been due to selective mortality (and, in any case, this would still need to be allowed for in the way demonstrated above). Moreover, we found that unless appropriate corrections are made for the regression dilution associated with particular exposure periods, the strength of associations of disease with usual blood pressure or cholesterol levels might be underestimated by about one-third during the first decade, one-half during the second decade, and two-thirds during the third decade of exposure. The corrections that are needed are independent of any assumptions about the constancy of relative risks over time or about the biologic mechanisms by which exposures affect disease (and are not much affected by age or gender).

Both the nonparametric and parametric statistical methods that have been used to estimate the magnitude of the regression dilution ratio should provide similar results when the variances of the values recorded at baseline and at remeasurement are similar. However, since the nonparametric method based on the ratio of ranges requires no such assumptions, it may be more suitable for longer intervals of follow-up. Appropriate adjustment for regression dilution in prospective studies requires remeasurements to be available at intervals during follow-up from at least a subsample of the survivors. If particular studies lack such data, then applying the present results from the Framingham and Whitehall studies for blood pressure and cholesterol may provide reasonable approximations for the corrections needed for each of the first three decades of exposure.

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