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Nephrosclerosis and aortic atherosclerosis from age 6 to 70 years in the United States and Mexico

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Summary. With increasing age, the thoracic aorta shows progressive fibroplastic intimal thickening, which is thought to be pre-atheromatous. A similar progressive intimal thickening in the renal cortical arteries is the distinguishing feature of the nephrosclerosis which underlies essential hypertension. Therefore, the earliest detectable youthful precursors of atherosclerosis and hypertension show strong morphological resemblances to each other. In this study, close statistical associations have been shown between the two types of arterial intimal fibroplasia. Both conditions show similar sigmoid growth curves from ages 6 to 70 years, thereby generating correlations across age groups of $r=0.99$ in New Orleans and $r=0.95$ in Mexico City. Specimens gathered in New Orleans were found to have about 1.4 times greater arterial intimal thickening than specimens from Mexico City, and this excess was seen at all ages in both the aortas and the renal cortical arteries. It seems likely that intimal fibroplasia of arteries is reflecting similar biological principles at all levels of the vascular tree. Whatever etiological factors vary between New Orleans and Mexico City, those factors appear to act directly at a tissue level to promote the early precursors of atherosclerosis and of the nephrosclerosis that underlies hypertension.

Key words: Hypertension – Aging – Arteriolosclerosis – Nephrosclerosis – Atherosclerosis

Introduction

With increasing age, the thoracic aorta shows progressive fibroplastic intimal thickening. The thickness of intima was recently found to be approximately proportional to age, from 15 to 55 years, with an average growth rate of about 6 μm per year; from 55 to 70 years, the growth rate slackened to about 3 μm per year (Tracy et al. 1987b). Small arteries of the renal cortex also show progressive fibroplastic intimal thickening with age, which is readily measured as intimal thickness in proportion to outer diameter (% OD) (Tracy et al. 1988b). In a mixed group of black and white males and females in New Orleans, a growth rate of about 0.25% OD per year was found from 15 to 55 years of age; the growth rate slackened to 0.13% OD per year from 55 to 70 years (Tracy et al. 1990). The similarities in the behavior of these two forms of arterial change with age, along with their morphological resemblances to each other, suggest the possibility of a pathogenetic linkage between them.

Fibroplastic thickening of arterial intima in the renal cortex is one of the defining feature of nephrosclerosis, whereby this condition is distinguished from other renal diseases; it is a feature of nephrosclerosis that relates well to blood pressure in subjects with essential hypertension (Bell 1950; Katafuchi and Takebayashi 1987; Sommers et al. 1958; Tracy and Toca 1974; Tracy et al. 1981, 1988a, b; Ueda et al. 1976; Yamaguchi et al. 1969). Fibroplastic intimal thickening of the aorta is sometimes said to be pre-atheromatous, in that the necrotic core of atherosclerosis is thought to emerge within the fibrotic substrate (Geer and Haust 1972; Ross et al. 1984; Stary 1989; Tracy and Kissling 1985; Tracy et al. 1983, 1987b). These aging changes, seen in two levels of the arterial tree, may constitute early steps that initiate a life-long course of progression toward clinically significant cardiovascular disorders (Freedman et al. 1988; Katafuchi and Takebayashi 1987; McGill 1968; Stary 1989; Tracy et al. 1987b).

An opportunity to examine the statistical associations between aortic and renal arterial fibroplasia further arose in association with studies of coronary heart disease (CHD; Tracy et al. 1983) and of childhood precursors of atherosclerosis (Freedman et al. 1988). Those studies provided samples of aortas and kidneys from a collection of coroners' autopsies in New Orleans. An opportunity to assess these matters in Mexico City also arose

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fortuitously. A series of coroners' autopsies constructed to compare coronary arteries in Mexico with those in New Orleans also provided samples of kidney tissues, but not aortas (Cueto-Garcia et al. 1989). Aortas from Mexico City had, however, been examined in an earlier study, and the data from that source are used here to provide a first look at Mexico for comparison with New Orleans (Tracy et al. 1986; Tracy and Kissling 1988).

The intimal fibroplasia of aorta and of renal cortical arteries have morphological similarities to each other. The present study was undertaken to explore some statistical similarities between the two pathological conditions. Autopsies conducted in a coroner's service on victims of sudden and violent deaths are often used to obtain a rough idea about the prevailing severities of arterial disorders in the general population (Freeman et al. 1988; McGill 1968; Strong and Restrepo 1978; Vanecek 1976). The use of this approach here is, therefore, in keeping with established practice.

Materials and methods

The 119 cases from the New Orleans CHD series were sampled from those examined at the Orleans Parish Coroner's Office from June 1979 to August 1980. The objective was to obtain cases of death due to CHD and other cases of death due to non-cardiovascular causes (basal) matched by age, race and sex to the CHD cases. Details are given elsewhere (Tracy et al. 1983). The sample contained 58 white males, 11 white females, 37 black males, and 13 black females; 52 had CHD and 67 had basal causes of death. The diagnosis of CHD was based upon the presence of coronary thrombosis or of arteriosclerotic occlusion of the coronary artery with myocardial scar. Basal cases had no evidence of cardiovascular disease by history or on gross inspection at the time of autopsy. Cases with anatomical evidence of CHD but dying of some other cause, and cases with other cardiovascular disease, such as cerebrovascular or renal disease, were excluded from this series.

The Bogalusa heart study is an epidemiological study of cardiovascular risk factors from birth to the age of 26 years (Freedman et al. 1988). Five cross-sectional surveys were conducted, covering more than 8000 subjects in Bogalusa, La. from 1973 to 1985. Of 104 autopsies conducted from 1978 to 1986 in the basis population, 40 supplied adequate samples of both kidney and aorta. Ages at death were 6–25 years. The sample contained 22 white males, 7 white females, 8 black males, and 3 black females. All cases were classed as basal causes of death.

Specimens were collected in 1960–1964 in New Orleans and Mexico City as a part of the International Atherosclerosis Project (IAP) (McGill 1968). Aortas from those sources have been previously examined microscopically, and the results have been reported elsewhere (Tracy et al. 1986). Those data are plotted graphically here to allow comparison of results between the two studies. Race, sex, and cause of death composition of the IAP data previously reported were somewhat different from the newly collected data. However, race and sex have proved to have little impact on the current results, and cause of death effects can be judged from the tabulated findings.

A study of coronary atherosclerosis in Mexico City was undertaken in 1988, by assembling a collection of specimens from 111 males dying of violent causes, and transporting them to New Orleans for evaluation (Cueto-Garcia et al. 1989). As a component of this undertaking, samples of kidney tissue were also assembled, and evaluated by the methods described here. These kidney samples have been measured only by observer A. Ages 12–54 years were represented. Microscopy of aortas was not done in this series; rather, data from the IAP series were used in their place.

Samples of kidney tissue were managed as for a routine autopsy without special handling. Bits of tissue cut perpendicular to the capsular surface were fixed and stored in acetate-buffered 10% formaldehyde. Samples were embedded in paraffin, sectioned at 6 μm, and stained with periodic acid-Schiff (PAS)-alcian blue. Generally, a total of 2–4 cm^2 area of renal cortex were represented in two to six sections of tissue. The method for morphometry of the kidney has been described at length elsewhere (Tracy et al. 1988b) and is summarized here. A microscope with ×10 and ×40 objective lenses and mechanical stage controlled by the left hand was equipped with an eyepiece ruler marked in units equivalent to 10 μm, under the ×10 objective lens. All arterial profiles in the section were examined systematically. The OD of the least axis of the elliptic profile was measured under the ×10 objective lens, excluding the adventitia, and measuring from one outer media to the other. The thicknesses of intima were measured under the ×40 lens, also along the least axis, with the better presented of the two opposite walls (i.e., lacking tangential sectioning, branch ostium, or artifact). If both opposite walls were equally well presented, then an average of the two was used. Vessels with OD less than 80 μm (i.e., arterioles) were excluded because they are often hyalinized, thus obliterating the intima and media. Vessels over 300 μm were excluded because it is impractical to obtain them in sufficient numbers in all specimens. The average ratio of intima to diameter in the "close" level vessels (OD 150–300 μm) is R_c, and in the "remote" level vessels (OD 80–149 μm) is R_r; units of measure are %OD. These size ranges are named with respect to the heart. Relative to each other, the remote level vessels are thought to offer greater resistance to blood flow, while the close level vessels have more of a strictly conduit function.

Three instances of diabetic nephropathy were encountered in the New Orleans CHD study. These were retained in the total pool. No chronic pyelonephritis, glomerulonephritis, malignant nephrosclerosis, or conditions other than benign nephrosclerosis were found besides the three diabetics. In Bogalusa and Mexico, no instance was encountered of kidney disease other than nephrosclerosis.

Aortas were opened longitudinally, formalin fixed, and sampled along the left and right lateral walls from the fourth to the twelfth intercostal ostia, as detailed elsewhere (Tracy et al. 1983). Longitudinally oriented segments were decalcified with acetic acid, blocked in paraffin, sectioned at 11 μm, and stained with hematoxylin and eosin. Ink marks were placed on the cover slips at intervals to define 1/20 of the sample length approximately 0.7 cm in most aortas.

Positions affected by atheronecrosis, operationally defined as having cholesterol clefts easily discerned under the 40× objective lens, were marked with black ink; the percentage of the length of the specimen marked positive for atheronecrosis was called P_A. The mean fibroplastic thicknesses of the intima (in micrometers) averaged over the non-necrotic points, were called F. Details of these methods are discussed elsewhere (Tracy et al. 1983).

Kidney and aortic sections of the 119 New Orleans CHD cases and 40 Bogalusa Heart Study cases were independently graded by a pathologist (A) and a biochemistry graduate student (B). The mean values of R_r for observers A and B were 4.5 and 3.9 %OD respectively; the mean values of R_c for observers A and B were 8.6 and 8.7 %OD, showing less grader difference for R_c than for R_r. The correlations between observers were 0.84 and 0.88 for R_r and R_c respectively, which are not significantly different. The mean values of F for observers A and B were 312 and 288 μm respectively; the mean values of P_A for observers A and B were 9.2 and 8.0% of the aortic specimen respectively. The correlations between observers were 0.96 and 0.91 for F and P_A respectively. The readings of the two observers were averaged into a single value for all variables in each case in subsequent analyses. Mexican renal slides, and the aortic slides from the IAP, were evaluated only by observer A.

In order to make age adjustments for R_c, R_r and F, the three variables were related to age (A) by cubic regression equations. In all three situations, the "intercept" term was not significantly

Table 1. Descriptive statistics for selected variables in 159 cases, total: New Orleans-Bogalusa combined sample

Name	Symbol	Units	Mean	SD	Correlation coefficients							
					R_c	R_r	F	P_A	R_{ca}	R_{ra}	F_a	P_{Aa}
Unadjusted												
Age	A	Years	44.0	17.2	0.784*	0.601*	0.763*	0.409*	−0.008	−0.010	−0.011	0.002
Nephrosclerosis												
Close	R_c	%OD	8.8	4.9		0.793*	0.608*	0.298*	0.606*	0.388*	−0.010	0.002
Remote	R_r	%OD	4.3	3.8			0.486*	0.361*	0.517*	0.789*	0.032	0.122
Aorta												
Fibroplasia	F	μm	281.1	159.4				0.688*	−0.008	0.019	0.630*	−0.002
Atheronecrosis	P_A	%	7.9	14.3					−0.021	0.138	0.601*	0.621*
Adjusted												
Close	R_{ca}	%OD	0	3.0						0.653*	−0.008	0.013
Remote	R_{ra}	%OD	0	3.0							0.019	0.158*
Fibroplasia	F_a	μm	0	101.6								0.007
Atheronecrosis	P_{Aa}	$\sqrt{}$%	0	1.4								

* Correlations significantly different from zero, $P<0.05$
R_{ca} is residual from $R_c=0.0393\ A+0.00742\ A^2-0.0000784\ A^3$ ($R^2=0.637$)
R_{ra} is residual from $R_r=-0.0407\ A+0.00499\ A^2-0.0000420\ A^3$ ($R^2=0.375$)
F_a is residual from $F=1.35\ A+0.231\ A^2-0.00243\ A^3$ ($R^2=0.512$)
P_{Aa} is residual from $\sqrt{P_A}=0.000282\ A\cdot F-0.0462\ A-0.00344\ F+0.66$ ($R^2=0.613$)

different from zero; therefore, the equations were fit again, forcing the curves through the origin (zero intercept). Results are given in the footnotes of Table 1, and the age-adjusted terms, R_{ra}, R_{ca}, and F_a, are calculated from the equations in that footnote. The first-order term on age was not statistically significant; elimination of that term yielded:

$$F=0.288\ A^2-0.00300\ A^3\ (R^2=0.593) \quad (1)$$
$$R_c=0.0313\ (0.290\ A^2-0.00303\ A^3)\ (R^2=0.626) \quad (2)$$
$$R_r=0.0152\ (0.215\ A^2-0.00163\ A^3)\ (R^2=0.366) \quad (3)$$

The constant factors in the right sides of Eqs. 2 and 3 represent the ratios of the means of R_c to F (8.81/281.1 = 0.0313) and of R_r to F (4.26/281.1 = 0.0152); these are introduced to facilitate comparisons of the regression coefficients between equations. Equations 1 and 2 are nearly identical except for the constant factor, whereas Eq. 3 has a smaller coefficient for A^3 than for A^2.

A strong correlation holds between P_A and F ($r=0.688$). Yet in some settings, it is desirable to examine the behavior of P_A after correcting for its association with F. For this purpose, residuals from the equation given in the footnote to Table 1 were used. The term, $-0.00344\ F$, was not statistically significant. After eliminating that term, the simplified result can be rendered.

$$\sqrt{P_A}=0.000221\ A\ (F-181)+0.30\ (R^2=0.611) \quad (4)$$

P_{Aa} is used to symbolize residuals from this equation. The reason for using the square root has been explained elsewhere (Tracy and Kissling 1985); in that former report, which deals with a different series of cases than the present paper, $\sqrt{P_A}=0.000166\ A\ (F-187)+0.47$ ($R^2=0.564$) was found. Yet another third series of cases reported $\sqrt{P_A}=0.000164\ A\ (F-185)+0.35$ ($R^2=0.542$) (Tracy and Kissling 1988). Those former results, together with Eq. 4, offer a first look at the likely range of variation that will be encountered in the parameter estimates from one series of cases to another. The quantity associated with F inside of the parentheses represents a threshold; if the average intimal thickness in an aorta fails to exceed this value, then each intimal position is expressed as having a negative probability of atheronecrosis. This would seem to imply that necrotic cores typically cannot exist in aortas with average intimal thickness less than 181, 187, or 185 μm, even at infinite age.

To explore the interaction of CHD with arterial variables, multiple linear regression was used, setting $C=1$ for the CHD, and $C=0$ for the non-CHD condition. For construction of Fig. 1, where

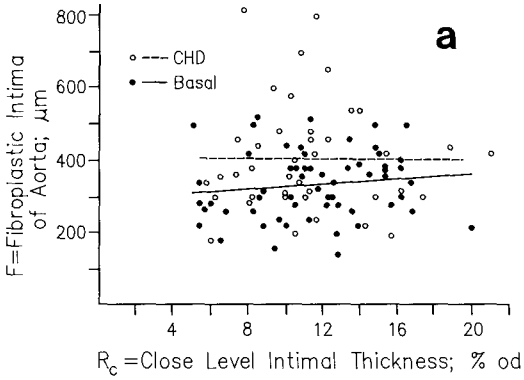

Fig. 1. Nephroclerosis (R_c) and aortic intimal fibroplasia (F) are plotted for the 100 cases aged 45–70 years in the New Orleans-Bogalusa series. *Solid* and *broken lines* represent the regression equation relating F to R_c and cause of death (coronary heart disease = CHD = 1; non-CHD = basal = 0) which had multiple $r=0.336$, reflecting a significant difference between cause of death groups. Region *a* is marked to aid discussion

lines depicting Eq. 5 are drawn, ages 45–70 years were used because this age stratum had ample cases of CHD. The relevant outcomes can be rendered.

$$F=3.29\ (R_c-34.3)\ (C-1.0)+408\ (R^2=0.090) \quad (5)$$
$$F=3.61\ (R_r-26.3)\ (C-0.9)+415\ (R^2=0.092) \quad (6)$$

These equations state that F does not change with R_c and R_r when $C=1$, i.e., in CHD cases; in basal cases, $C=0$, F actually *declines* with R_c and R_r. These relationships, however, are not statistically significant. Only the correlation between F and C is statistically significant; the partial correlations of $F\cdot R_r$ and $F\cdot R_c$ being $r=0.062$ and $r=0.055$ respectively. The percentage of the aortic sample affected by necrotic cores was expressed as P_{Aa}, the residuals from Eq. 4. Since this variable is age adjusted, the total pool of 159 cases was used. The relevant outcomes can be rendered.

$$P_{Aa}=0.112\ (R_r+0.9)\ (C-0.2)-0.14\ (R^2=0.081) \quad (7)$$
$$P_{Aa}=0.089\ (R_c-1.9)\ (C-0.4)+0.04\ (R^2=0.071) \quad (8)$$

The terms 0.9, −0.2, −1.9, and −0.4 associated with the variables in parentheses are small and are not of statistical significance. These equations state that atheronecrosis (P_{Aa}) significantly correlates with nephrosclerosis (R_r, R_c) but only in the CHD cases (i.e., the association is not significant if $C=0$). Indeed, a slight *inverse* correlation was seen in the basal cases (i.e., $C=0$), as if severe nephrosclerosis may tend to shift the high P_{Aa} cases out of the basal into the CHD cause of death group by precipitating CHD.

Results

Subjects who were alike in age showed great variation among each other in nephrosclerosis (R_c) and aortic intimal fibroplasia (F) (Fig. 1). Only a small part of the variation in aortic intima among age-matched subjects was due to membership in cause of death groups; CHD subjects averaged significantly more F than those who lacked CHD (basal subjects), but the overlap between cause of death groups was great. The measures of nephrosclerosis did not differ significantly between cause of death groups.

The thickness of intima in renal cortical arteries correlated strongly with aortic intimal fibroplasia ($r=0.608$ and 0.486 for R_c and R_r respectively, Table 1). These correlations became insignificant after age adjustment, ($r=-0.003$ and 0.047 for R_{ca} and R_{ra} respectively, Table 1). Within both CHD and basal cause of death groups, of ages 45–70 years, the absence of a significant correlation between R_c and F was found (partial $r=$ 0.055 corrected for the CHD correlations, Fig. 1). The similar partial r for R_r was 0.066, and also is not significantly different from zero. Indeed, the correlation between aortic and renal intima was not of statistical significance in any of the age groups from 6 to 70 for either R_r or R_c (Table 2). The region marked "a" in Fig. 1 is conspicuously empty, as if the co-existence of maximal F and R_c is impossible. If mortality prevents occupancy of region "a", then some artifactual distortion of the pattern could be encountered in the oldest age groups. To compensate for this effect, the subjects of ages 6–50 years can be examined by averaging the correlations within age groups 6–8, 10–19, 20–29, 30–39, and 40–49 years in Table 2. The resulting estimates for $F \cdot R_c$ and $F \cdot R_r$ respectively are $r=0.022$ and $r=0.140$; with 74 cases, these correlations are not significantly different from zero.

When assessing nephrosclerosis and fibroplasia using group mean data the mean values of F and R_c from Table 1, 281.1 and 8.8, were found to have a ratio of 32:1, implying that the average intima tended to be about 32 times thicker in the aorta than in the 100-μm-diameter renal cortical arteries. Figure 2 is constructed so that the two vertical scales on the left and right have a ratio of 32:1; each unit of R_c corresponds to 32 units of F. The plotted overlapping 10-year age group averages of F and R_c (CHD and basal cases combined) very nearly coincide with each other from age 6 to 70 years (open and closed circles respectively in Fig. 2, $r=0.990$); their average ratio ± one standard deviation was 32.9 ± 2.2. Data on cases from Mexico City also reveal a close correspondence between group average F and R_c (open and closed squares respectively in Fig. 2), even though the kidneys were collected 25 years later than the aortas, and from an entirely different set of subjects ($r=0.956$); their average ratio was 36.9 ± 7.5, which does not differ significantly from the New Orleans cases. Data from a group of aortas assembled in New Orleans in 1960–1964 are also plotted in Fig. 2 (open diamonds). All of the curves in Fig. 2 are alike in following a sigmoid shape which rises about 4-fold in a nearly linear manner from 20 to 55 years of age; the curves are flatter during the childhood growth period from 6 to 20 years, and again when entering old age, from 55 to 70 years. The aortic intimal thickness in New Orleans averages 1.36 ± 0.17

Table 2. Means of selected variables by cause of death according to *overlapping* 10-year age groups in 159 cases, total sample[a]

Age	Number of cases		F; μm		P_A; %		R_c; %OD		R_r; %OD		Correlation $R_c \cdot F$
	CHD	Basal	CHD	Basal	CHD	Basal	CHD	Basal	CHD	Basal	
6–8	0	3	X	49	X	0	X	0.3	X	0.1	−0.181
6–14	0	6	X	62	X	0	X	1.4	X	0.4	0.141
10–19	0	26	X	77	X	0	X	2.8	X	0.8	0.016
15–24	0	33	X	89	X	0	X	2.8	X	0.9	0.029
20–29	0	11	X	112	X	0	X	3.1	X	1.2	−0.190
25–34	5	3	214*	118	3.8	0	6.0	4.0	1.7	1.9	0.103
30–39	5	5	214	191	3.8	0.6	6.0	6.1	1.7	2.0	0.368
35–44	5	7	320	262	4.6	0.9	7.7	9.9	2.9	3.3	−0.350
40–49	12	12	350	293	8.2	3.0	9.2	10.7	4.6	4.5	0.006
45–54	18	26	403*	311	17.9*	4.9	10.6	11.0	6.2	4.9	0.142
50–59	20	37	387	330	16.1*	5.8	11.7	11.6	6.5	5.5	0.130
55–64	22	30	415*	353	20.0**	8.4	11.5	12.5	6.4	6.4	−0.077
60–69	15	12	455*	369	28.1**	11.8	11.3	12.2	7.0	6.1	−0.217
65–70	2	2	383	357	28.0	5.8	12.4	11.5	5.0	5.9	−0.530
70	0	1	X	401	X	2.5	X	14.1	X	6.8	X

[a] Variable symbols are keyed in Table 1

* $P<0.05$, ** $P<0.01$ from t-tests of differences between coronary heart disease (CHD) and basal groups; correlation coefficients all are not significantly different from zero

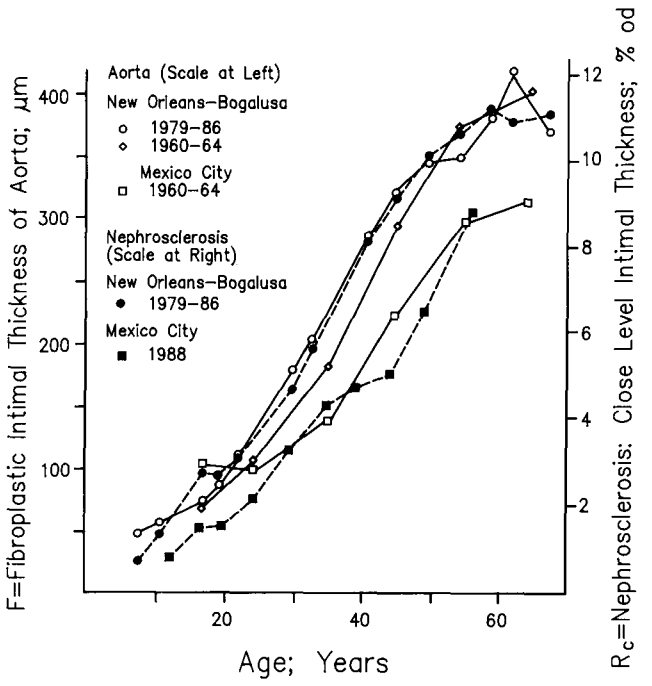

Fig. 2. Mean values for nephrosclerosis (R_c, scale at right) and aortic intimal fibroplasia (F, scale at left) by overlapping 10-year age groups are plotted against age

(SD) times thicker in New Orleans than in Mexico City at ages 20–54 years; the intima of renal arteries averages 1.51 ± 0.24 times thicker; the weighted average ratio combining all of the age-grouped specimens from 20 to 54 years was 1.4.

Aortic intimal fibroplasia was about 15% thicker in males than in females, as shown by ANOVA performed upon age-adjusted data (Table 3). Blacks exceeded whites in the intimal thicknesses of renal arteries and aortas, but the differences were confined to the CHD groups (Table 4); no significant race difference was found in the basal cause of death groups. No significant race or sex differences in atheronecrosis were found by ANOVA carried out on the age-fibroplasia adjusted variable, P_{Aa} (data not tabulated).

A thin, youthful aortic intima (Fig. 3A) is compared with an elderly fibrotically thickened intima of a renal cortical artery (Fig. 3B). Whereas the age-specific, average intimal thicknesses have a ratio of 32:1, these photographic examples have only a 2:1 ratio. The examples were chosen from the thin end of the aortic range and the thick end of the renal arterial range to facilitate comparisons of structure. The aortic examples were also chosen to exclude areas of infiltration by foam cells and leukocytes, so that nearly all observed cells are presumably smooth muscle. In both levels of the arterial tree, elongated smooth muscle cells, seen in longitudinal sections cut perpendicular to the vessel wall, are oriented in the direction of blood flow (white arrows). Those cells are invested with PAS-positive basement membranes (dark lines outlining cells), which become thickened and blurred as the intima thickens with age. Note how the media of renal arteries shrinks in areas of intimal thickening (Fig. 3). Planes of sectioning taken tangential to the vessel wall are shown for an elderly aortic fibrous plaque (Fig. 4A) and for an elderly renal arterial intima (Fig. 4B). In this plane of presentation, cells sometimes retain the same narrow, elongated contour as in the perpendicular plane of section (white arrows). These cells are inferred to be cylindrical in three dimensions. Yet other cells are rounded, angulated, or stellate in the tangential plane (curved arrows). These cells are inferred to be flattened, pancake-like, in three dimensions. Lipid deposits (Fig. 4, "L") are commonly seen in the aorta but are always absent in the thickened intima of small renal arteries.

Table 3. Mean aortic intimal fibroplasia (F_a) by cause of death and sex, age adjusted to age 44

	CHD		Non-CHD (basal)	
	Male	Female	Male	Female
Number of cases	39	13	86	21
$F_a+281.1$; μm	333	278	270	235
ANOVA Causes	9.54 (0.00)			
Sexes	8.85 (0.00)			
Interactions	0.59 (0.44)			

ANOVA: F-test (probability of being zero) was computed on the age-adjusted variable, F_a, (footnote Table 1) and entered here by adding 281.1, which is the mean fibroplasia at mean age 44. ANOVA is type III sums of squares from PROC GLM of SAS

Table 4. Mean aortic intimal fibroplasia (F_a) and nephrosclerosis measured from intimal thicknesses of close level (R_{ca}) and remote level (R_{ra}) renal cortical vessels by cause of death and race; age adjusted to age 44

	CHD		Non-CHD (basal)	
	Black	White	Black	White
Number of cases	23	29	38	69
$F_a+281.1$; μm	361	286	257	266
$R_{ca}+8.8$; %OD	9.8	7.5	8.9	9.0
$R_{ra}+4.3$; %OD	5.7	3.4	4.0	4.4

ANOVA	Fibroplasia	Close intima	Remote intima
Causes	13.98 (0.00)	0.27 (0.60)	0.44 (0.51)
Races	3.85 (0.05)	4.44 (0.03)	3.29 (0.07)
Interactions	6.32 (0.00)	5.45 (0.02)	7.10 (0.01)

ANOVA: F-test (probability of being zero) was computed on the age-adjusted variables (footnote Table 1) and entered by adding 281.1 to mean F_a, 8.8 to mean R_{ca}, and 4.3 to mean R_{ra}, which are the means of the three variables at mean age 44. ANOVA is type III sums of squares from PROC GLM of SAS

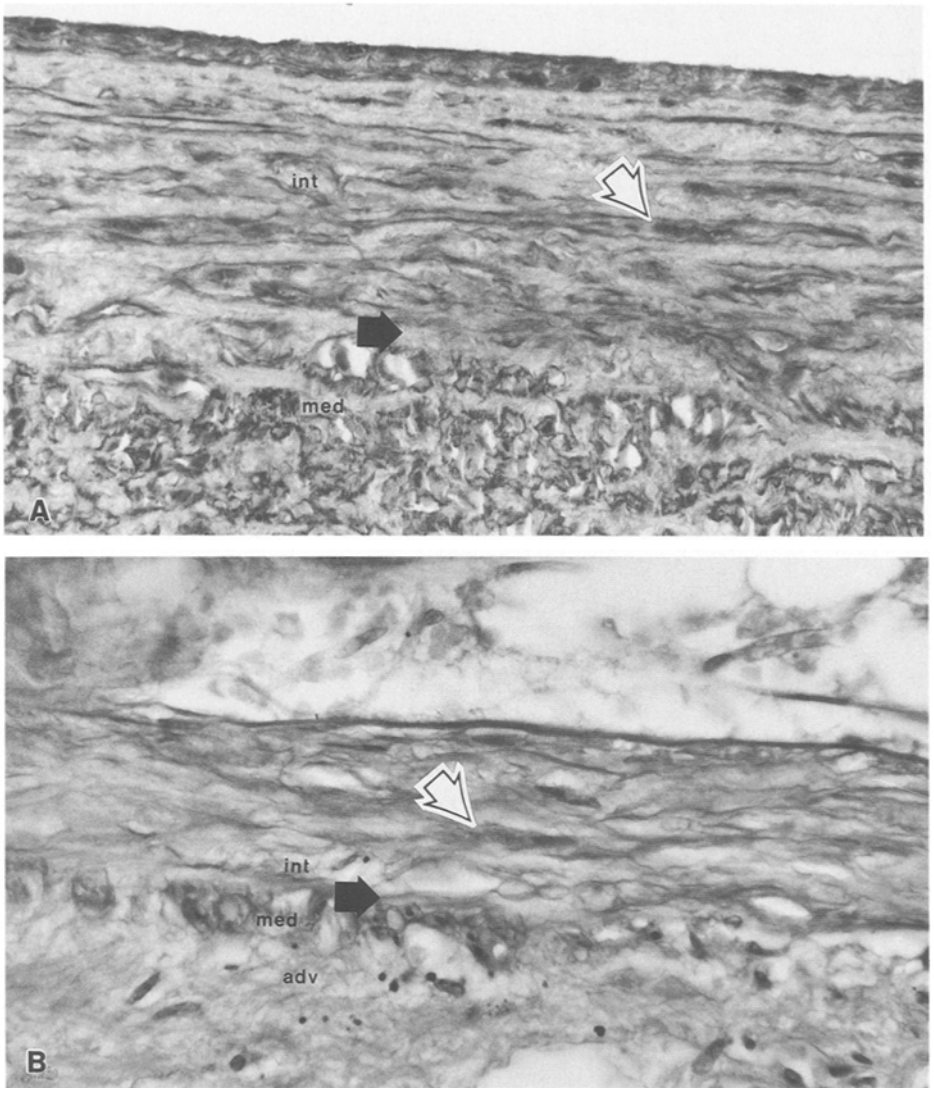

Fig. 3. Longitudinally oriented planes of cut perpendicular to the vessel walls are shown for a youthful aorta (**A**) and an elderly renal cortical artery (**B**). *Dark arrows* mark intima-media boundaries. *Int*, *med* and *adv* denote the intima, the media and adventitia respectively. The media in **B** is shrunken in company with intimal thickening. *White arrows* mark smooth muscle cells with their darkly staining basement membranes. Periodic acid Schiff, × 700

Discussion

Intimal fibroplasia of the aorta, seen in areas of tissue sections which lack infiltration by foam cells and leukocytes, resembles the intimal thickening of microscopic renal arteries. In both situations, layers of dense collagen alternate with loose matrix materials (Bell 1950; Geer and Haust 1972; Katafuchi and Takebayashi 1987), thickening of the intima with age is marked by increasing numbers and thicknesses of layers (Tracy et al. 1987a, 1988b), and the initially longitudinal spindle cells of the intima become widely spread apart with age as they flatten and lose their longitudinal orientation. In the aorta, these flattened cells have been called "stellate" (Geer and Haust 1972; Orekhov et al. 1986) or "pancake" (Ross et al. 1984) cells; in the small renal arteries they have not been described before. Figure 2 illustrates some of the statistical properties that aorta and renal vessels share; the sigmoid growth curves for the two tissues, marked off by data on group averages, are nearly identical to each other in the 1979–1986 New Orleans-Bogalusa data, wherein aortas and kidneys were drawn from the same sets of cases. Even in the Mexican data, wherein kidneys were drawn from a different set of cases than those which supplied the aortas, the two curves show some similarities to each other and to the New Orleans-Bogalusa data. These findings suggest that the evolution of intimal thickening may be governed by a common set of biological principles which apply alike to all levels of the arterial tree. Moreover, for both tissues the differences between New Orleans and Mexico City can be approximately described as a difference of a constant factor: the intima of all types of vessels averaged about 1.4 times greater in New Orleans at all ages. Whatever etiological agents differ between the two cities, they seem to act solely by altering the magnitude of a single rate parameter within a common growth process. Part of the population difference could be due to different sizes of aortas, smaller organs of Mexicans presumably invested with smaller intimal structures. A similar effect could be postulated for the 15% excess of males over females in aortic intima. This possibility remains to be tested.

When individuals within a broad age group were examined, as in Fig. 1, only a tiny and statistically insignifi-

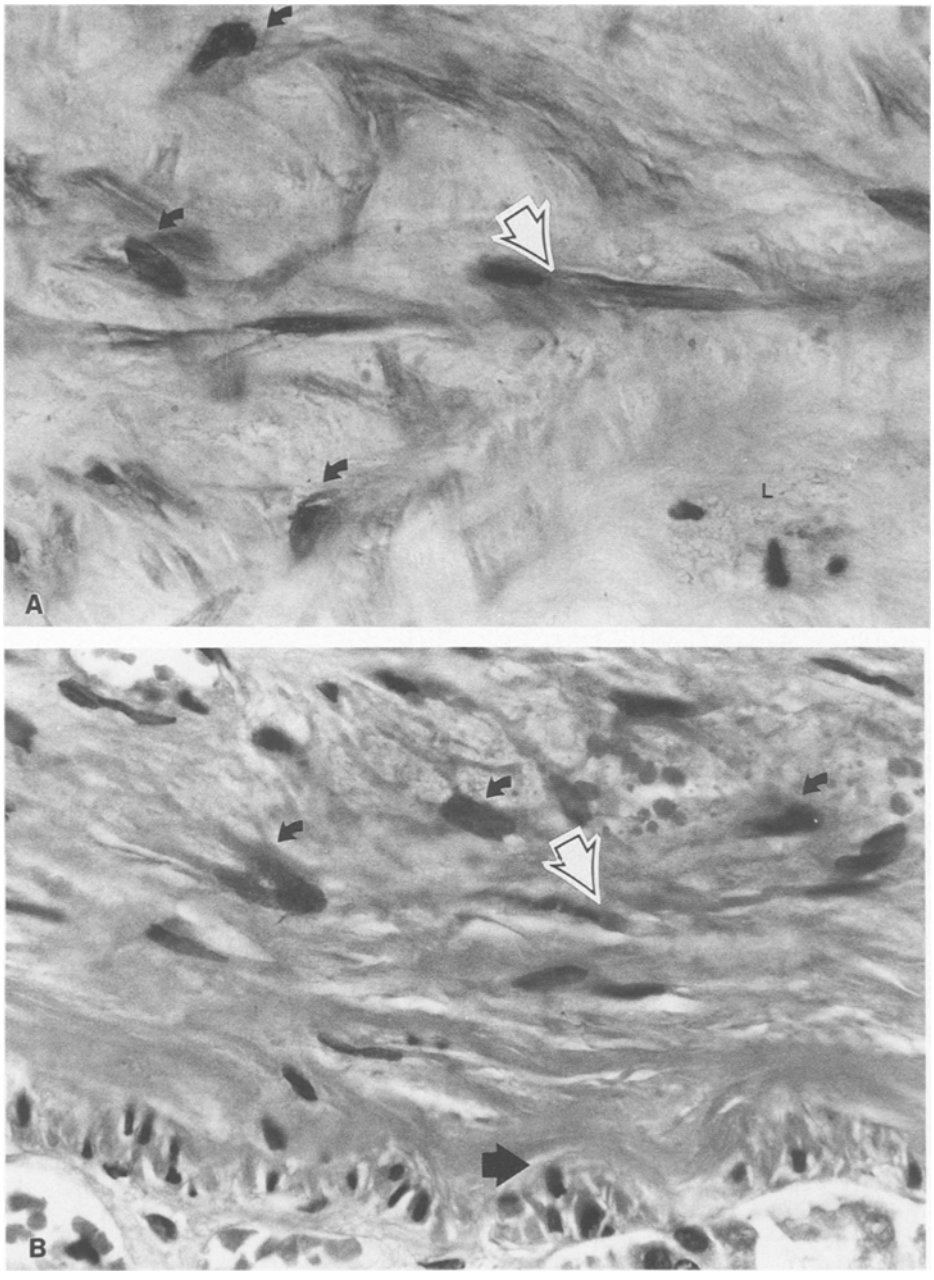

Fig. 4. Planes of cut tangential to a thick aortic fibrous plaque (**A**) and a thickened renal cortical artery (**B**) are oriented with blood flow from left to right. *White arrows* mark elongated smooth muscle cells, while *curved arrows* mark rounded, angulated, or stellate ones. *Course arrow* in (**B**) marks intima-media boundary. Region *L* has lipid-laden cells. Hematoxylin and eosin, ×700

cant correlation was found between aortic and small renal arterial intimal thicknesses ($r=0.055$). Some individuals had much thickening in the aorta and little in the renal vessels, while other individuals reversed this pattern; still other individuals had simultaneously low or high values in both sets of vessels. This outcome seems to say that one or another level of the vasculature can grow old alone, leaving its opposite member still young; and the choice of which level will grow old appears to be random.

The two outcomes taken together imply that the aging of arteries is governed by two classes of etiological agents: one class can act to promote aging changes at all levels of the vasculature, while the other class acts to deflect the aging process toward the gross or the microscopic but not both levels. The agents which act at all levels of the arterial tree appear to be the ones which vary in strength between cities. The strong correlation shown between age groups and between the United States and Mexico appears to be offset by an equally strong inverse correlation within individuals due to previously unsuspected physiological mechanisms which now, because of these results, need to be explained. A mathematical development of this argument is given in appendix A.

In a former study of aortas collected in 1960–1964, no sex difference was found for aortic fibroplasia in New Orleans blacks or whites or other populations (Tracy and Kissling 1988), whereas a significant sex difference was found here. No reason is apparent to explain the contrary outcomes in these two studies. In two previous studies of nephrosclerosis at ages 50–90 years, the reno-

vascular alterations were greater in CHD than in basal cause of death groups (Tracy et al. 1988a, b). Two former studies of men aged 25–44 years (Strong et al. 1984) and of men and women aged 25–54 years (Tracy et al. 1991) found no significant difference in renovasculopathy between CHD and basal cause of death groups, in agreement with the results obtained here. These results suggest that hypertension is not often a cause in most instances of CHD up to 50 years of age, but becomes a frequent cause thereafter.

Specimens gathered in New Orleans were found to have about 1.4 times greater arterial intimal thickening than specimens from Mexico City, and this excess was seen at all ages in both the aortas and the renal cortical arteries. Hence the early fibroplastic stage of atherosclerosis along with the early stages of nephrosclerosis were both found to vary significantly between populations, and to parallel each other across geographic lines. This phenomenon is reported here for the first time. Moreover, the commencement of intimal fibroplasia in childhood, and its subsequent evolution into old age along a sigmoid growth curve, were found to follow identical patterns in the aorta and renal cortical arteries. These results confirm previous reports from New Orleans (Tracy et al. 1987b, 1990) and extend them to Mexico. It seems likely that intimal fibroplasia of arteries is reflecting similar biological principles at all levels of the vascular tree. Whatever etiological factors vary between New Orleans and Mexico, those factors appear to act directly at a tissue level to promote simultaneously the early precursors of atherosclerosis and of the nephrosclerosis that underlies hypertension.

The following conclusions can be drawn from our study. The fibrotically thickened aortic intima in aging individuals was found to be greater in subjects with CHD than in subjects with non-cardiovascular causes of death. Intimal thickening of renocortical arteries showed no significant difference between cause of death categories. A strong correlation between intimal thicknesses of the aorta and renocortical arteries was attributed entirely to their common association with age within the sample of cases assembled in New Orleans. Between New Orleans and Mexico, intimal thicknesses of the aorta closely paralleled those of renocortical arteries at all ages. In New Orleans and Mexico the patterns of change with age in the aorta are remarkably similar to those in the renocortical arteries. A mathematical model is used to reconcile these apparently paradoxical results, and to show that positive correlations between groups of subjects can be consistent with absence of correlation between individuals within groups.

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Appendix A

Between the renal arterial (R_c) and aortic (F) intimal fibroplasias, three correlations can be drawn from the data. The correlation among all 159 individuals in the study is read from Table 1, $r_i = 0.608$. The correlation among individuals within age groups 6–8, 10–19, 20–29, 30–39, 40–49, 50–59, and 60–69, in the last column of Table 2, has weighted average $r_w = 0.020$. Finally, the means of F and R_c in the seven age groups in Table 2 are correlated with $r_g = 0.987$. The fact that these three correlations are roughly consistent with each other can be shown as follows.

If the true population correlations between group means and among individuals within age groups are denoted ρ and γ respectively, then, following McGill et al. [1981], we can write

$$r_i = \frac{\rho + \gamma \dfrac{\delta_x \delta_y}{\sigma_x \sigma_y}}{\sqrt{\left[\left(1 + \dfrac{\delta_x^2}{\sigma_x^2} + \dfrac{\varepsilon_x^2}{\sigma_x^2}\right)\left(1 + \dfrac{\delta_y^2}{\sigma_y^2} + \dfrac{\varepsilon_y^2}{\sigma_y^2}\right)\right]}} \tag{A1}$$

$$r_g = \frac{\rho + \gamma \dfrac{\delta_x \delta_y}{m\sigma_x \sigma_y}}{\sqrt{\left[\left(1 + \dfrac{\delta_x^2}{m\sigma_x^2} + \dfrac{\varepsilon_x^2}{m\sigma_x^2}\right)\left(1 + \dfrac{\delta_y^2}{m\sigma_y^2} + \dfrac{\varepsilon_y^2}{m\sigma_y^2}\right)\right]}} \tag{A2}$$

$$r_w = \frac{\gamma}{\sqrt{\left[\left(1 + \dfrac{\varepsilon_x^2}{\delta_x^2}\right)\left(1 + \dfrac{\varepsilon_y^2}{\delta_y^2}\right)\right]}} \tag{A3}$$

Subscripts x and y refer to R_c and F, respectively. A hypothetical error free variance among individuals within age groups is δ^2, and σ^2 is the variance among the means of the groups. The number of individuals, m, is taken to be the same in all groups. The quantity, ε^2, is a hypothetical variance of repeated measures within individuals. In reality, fibroplasia cannot be measured at more than one point in time for each individual. Theoretically, however, the relative position of each individual in a group might drift upward or downward over time, and this source of variation, measured by ε^2, would contribute to attenuating the observed correlation, r_w, below the true value, γ. These relationships call for the usual assumptions about lack of correlation among the components of variation.

The quantities, σ_x, σ_y, $(\delta_x^2 + \varepsilon_x^2)$, and $(\delta_y^2 + \varepsilon_y^2)$, are open to estimation from observations, while δ_x, δ_y, ε_x, and ε_y are not. Observed variances within the seven age groups of Table 2 are pooled to estimate the composite sum of error plus error-free variance among individuals within age groups, $(\delta_x^2 + \varepsilon_x^2) = (3.21\ \mu m)^2$ and $(\delta_y^2 + \varepsilon_y^2) = (101.2\ \mu m)^2$. Among the seven chosen age group *means* in Table 2, $\sigma_x = 4.64$ and $\sigma_y = 145.9$ are found. There are now six unknown quantities, ρ, γ, δ_x, δ_y, ε_x, and ε_y. To solve for these unknowns, additional information is needed. Noting that $(\delta_y^2 + \varepsilon_y^2)/(\delta_x^2 + \varepsilon_x^2) = (101.2/3.21)^2 = 31.5^2$ and that $\sigma_y/\sigma_x = 145.9/4.64 = 31.4$, it seems reasonable to guess that

$$\frac{\varepsilon_y}{E_x} \simeq \frac{\delta_y}{\delta_x} \simeq \frac{\sigma_y}{\sigma_x} = 31.4 \tag{A4}$$

The number of subjects in group i, n_i, is not the same for the N age groups in Table 2. Hence, the constant, m, in Eq. A2 must be replaced by $(1/N)\sum(1/n_i) = 1/m$, which is found to yield $m = 10.6$ for the $N = 7$ age-groups, excluding one subject of age 70.

Substituting into Eqs. A1 and A2 the results from Eq. A4 plus $\sigma_x = 4.64$ and $(\delta_x^2 + \varepsilon_x^2) = 3.21^2$ gives

$$\rho = 1.479\ r_i - 0.0464\ \gamma\delta_x^2 \tag{A5}$$

$$\rho = 1.045\ r_g - 0.00438\ \gamma\delta_x^2 \tag{A6}$$

$$\gamma = r_w\ (1 + \varepsilon_x^2/\delta_x^2) \tag{A7}$$

Eliminating ρ from Eqs. A5 and A6, and introducing observed estimates $r_i = 0.608$ and $r_g = 0.987$ yields $\gamma\delta_x^2 = -3.17$. Putting this result into Eq. A7, along with $\varepsilon_x^2 = 3.21^2 - \delta_x^2$, yields expected $r_w = -0.308$. The observed values of the correlation among individuals within the seven age groups of Table 2 range from $r_w = -0.22$ to $r_w = 0.37$, with a weighted average of 0.020. The calculated value,

-0.308, does not differ significantly from the observed point estimate, 0.020.

We can go on to estimate $\rho = 1.043$, which is not far from unity or slightly less, and is consistent with a nearly perfect correlation among age group means in the hypothetical absence of measurement error. We can also explore the maximal ranges of the hypothetical quantities, γ and δ_x, using the result, $\gamma\delta_x^2 = -3.17$ along with $(\varepsilon_x^2 + \delta_x^2) = 10.3$. The outcome is shown in Table A. Values of these parameters outside of the ranges in Table A are mathematically imaginary. These considerations lead to some surprising conclusions. The coexistence of a very high correlation between the fibroplasias observed in the aorta and the renal arteries among age groups, together with large variability of these measures among individuals within age groups, can be reconciled only by invoking an antagonism between the variables within individuals, thus generating a negative γ. Furthermore, the observed correlation, r_w, is presumed to be increasingly attenuated by greater amounts of measurement error, ε^2, so that our estimate of the hypothetical true correlation, γ, grows increasingly negative as ε^2 increases.

Table A. Parameters of variance and correlation under certain hypotheses

δ_x^2	ε_x^2	γ
10.3	0	-0.308
8.3	2	-0.382
6.3	4	-0.503
4.3	6	-0.737
3.17	7.13	-1.00

Appendix B

Since blood pressure correlates with grossly evaluated atherosclerosis, it seems reasonable to ask whether blood pressure might correlate with microscopically evaluated aortic intimal fibroplasia, one of the variants of atherosclerosis. Direct testing of this matter by measuring blood pressure and fibroplasia in a series of cases has never been attempted. An indirect approach, however, can be pursued here, using the rationale of path analysis, which offers a way to explore the consequences of assumptions about causality. The partial correlation coefficient can be set to zero under certain assumptions, and this allows tractable calculations about the expected values of other correlations. If we assume no association between the aorta and the kidney except through the sole mediation of mean blood pressure, then we can set the partial correlation to zero.

$$r_{xy.z} = \frac{r_{xy} - r_{xz}\, r_{yz}}{\sqrt{(1 - r_{xz}^2)(1 - r_{yz}^2)}} = 0 \qquad (9)$$

where x = nephrosclerosis, y = aortic fibroplasia, and z = blood pressure. This equation states that all of the correlation between x and y, later documented to be $r_{xy} = 0.055$, is taken to be caused by the hypothetical action of blood pressure upon the aorta. In various assortments of cases, age-adjusted correlations between nephrosclerosis and blood pressure ranging from $r = 0.4$ to 0.7 have been reported, with a most frequent value near 0.6 (Katafuchi and Takebayashi 1987; Tracy et al. 1983, 1987a, 1988a, b, 1990; Yamaguchi et al. 1969). These assumptions allow us to estimate the hypothetical correlation to be $r_{yz} = r_{xy}/r_{xz} = 0.055/0.6 = 0.092$. If this correlation is derived from a group of 100 cases aged 45–70 years, then the 95% confidence interval of the estimated correlation coefficient is -0.11 to 0.292, using Fisher's z-transform. The provisional conclusion, therefore is that mean blood pressure is unrelated to aortic intimal fibroplasia between individuals within a population.

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