High Accumulation of Elements in the Arteries of the Lower Limbs with Aging

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ABSTRACT

To elucidate compositional changes of the arteries in the upper and lower limbs with aging, the authors investigated age-related changes of elements in both the arteries of the upper and lower limbs by inductively coupled plasma-atomic emission spectrometry. After the ordinary dissection at Chiang Mai University, the brachial, radial, femoral and posterior tibial arteries were resected from the subjects, ranging in age from 25 to 88 yr. The subjects consisted of 15 men and 5 women (average age = 67.5 $\pm$16.0 yr). It was found that although an accumulation of Ca and P hardly occurred in the brachial and radial arteries with aging, a high accumulation of Ca and P occurred in both the femoral and posterior tibial arteries with aging. Accumulation of Ca and P in the femoral and posterior tibial arteries began to occur in the sixties and thereafter increased remarkably. As for the arteries over 60 yr, the average content of Ca was about 4-fold higher in the femoral and posterior tibial arteries than in the brachial and radial arteries.

Key Words: Artery, Calcium, Phosphorus, Magnesium, Atherosclerosis

INTRODUCTION

There are a number of reports (Kanabrocki et al., 1960; Yu and Blumenthal, 1963; Elliott and McGrath, 1994; Tohno, S. et al., 1997; Tohno and Tohno, 1998a; Tohno, S. et al., 1998b; Tohno, Y. et al., 2001d) in which direct chemical analysis of elements in human arteries was utilized. Based on age-related changes of elements in the arteries, it was found that in Japanese, an accumulation of Ca and P did not occur uniformly in any arteries with aging and a higher accumulation occurred in both the aorta and the arteries of the lower limb with aging (Tohno and Tohno, 1998a). Few works have been done to study an accumulation of elelments in human arteries except for Japanese.

Therefore, the authors investigated age-related changes of elements in the arteries of Thai and found that a higher accumulation of Ca and P occurred in the arteries of the lower limb with aging, whereas it hardly occurred in the arteries of the upper limb.

MATERIALS AND METHODS

Sampling of Arteries

Cadavers were treated by injection of a mixture of 26% methanol, 14% glycerin, 3% phenol, 14% formalin, 0.34 M potassium nitrate and 14 mM arsenic oxide through the femoral artery (Tohno, Y. et al., 2001a). After the ordinary dissection at Chiang Mai University was finished, the brachial, radial, femoral and posterior tibial arteries were resected from the subjects and washed thoroughly with distilled water.

Determination of Elements

The samples of arteries were dried at 80˚C for 16 hr. After 1 ml conc. nitric acid was added, they were heated at 100˚C for 2 hr. After the addition of 0.5 ml conc. perchloric acid, they were again heated at 100˚C for an additional 2 hr. The samples were adjusted to a volume of 10 ml by adding ultrapure water and were filtered through the filter paper (No.7; Toyo Roshi Co., Osaka, Japan). The resulting filtrates were analyzed with an inductively coupled plasma-atomic emission spectrometer (ICPS-1000 III; Shimadzu Co., Kyoto, Japan) (Tohno, S. et al., 1998a). The conditions were 1.2 kW of power from a radio-frequency generator, a plasma argon flow rate of 1.2 l/min, a cooling gas flow of 14 l/min, a carrier gas flow of 1.0 l/min, an entrance slit of 20 µm, an exit slit of 30 µm, a height of observation of 15 mm and integration time lapse of 5 s. The amount of element was expressed on a dry-weight basis.

Statistical Analysis

Statistical analyses were performed, using the GraphPad Prism Version 2.0 (GraphPad Software Inc., San Diego, CA, USA). Pearson's correlation was used to investigate the association between parameters. A p-value of < 0.05 was considered to be significant. Data were expressed as the mean ± standard deviation.

RESULTS

The subjects for the femoral and posterior tibial arteries consisted of 15 men and 5 women, ranging in age from 25 to 88 yr (average age = 67.5 ± 16.0 yr). The brachial and radial arteries were resected from the same subjects, but as the brachial and radial arteries could not be resected from the four subjects, they were not analyzed in the present study. The average age of the 16 subjects for the brachial and radial arteries was 67.7 ± 16.4 yr.

Age-Related Changes of Elements in the Arteries

From a viewpoint of anatomically-corresponding regions, the brachial artery was compared with the femoral artery, whereas the radial artery was compared with the posterior tibial artery.

Figure 1A shows age-related changes of Ca in the brachial and femoral arteries. The correlation coefficients between age and the Ca content were estimated to be 0.309 (p = 0.244) in the brachial arteries and 0.574 (p = 0.008) in the femoral arteries, indicating that there was a very significant direct correlation in the femoral arteries, but there was no significant correlation in the brachial arteries. Figure 1B shows age-related changes of Ca in the radial and posterior tibial arteries. The correlation coefficients between them were estimated to be 0.015 (p = 0.955) in the radial arteries and 0.500 (p = 0.025) in the posterior tibial arteries. A significant direct correlation was found between them in the posterior tibial arteries, whereas no significant correlation was found in the radial arteries.

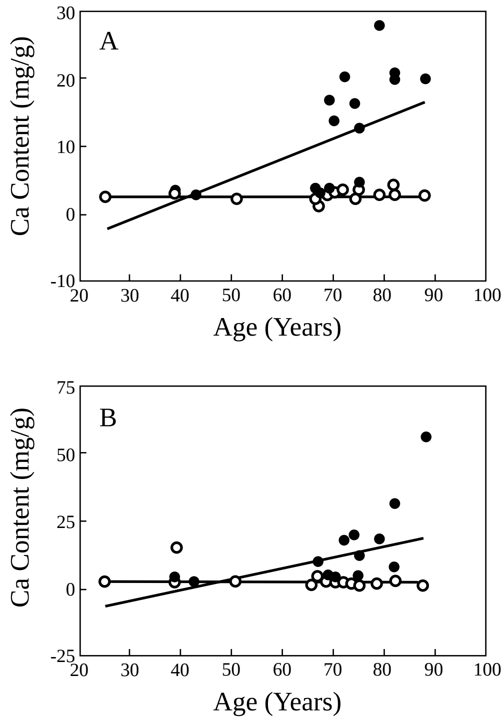

Figure 1. Age-related changes of Ca in the brachial and femoral arteries (A) and in the radial and posterior tibial arteries (B). In Fig. 1A, the open and closed circles denote the brachial and femoral arteries, respectively. In Fig. 1B, the open and closed circles denote the radial and posterior tibial arteries, respectively.

Figure 2A shows age-related changes of P in the brachial and femoral arteries. The correlation coefficients between age and the P cotent were estimated to be 0.494 (p = 0.052) in the brachial arteries and 0.497 (p = 0.026) in the femoral arteries. A significant direct correlation was found between age and the P content in the femoral arteries, whereas no significant correlation was found in the brachial arteries. Age-related changes of P in the radial and posterior tibial arteries are shown in Fig. 2B. The correlation coefficients between them were estimated to be -0.005 (p = 0.986) in the radial arteries and 0.466 (p = 0.039) in posterior tibial arteries. A significant direct correlation was found between them in the posterior tibial arteries, but no significant correlation was found in the radial arteries.

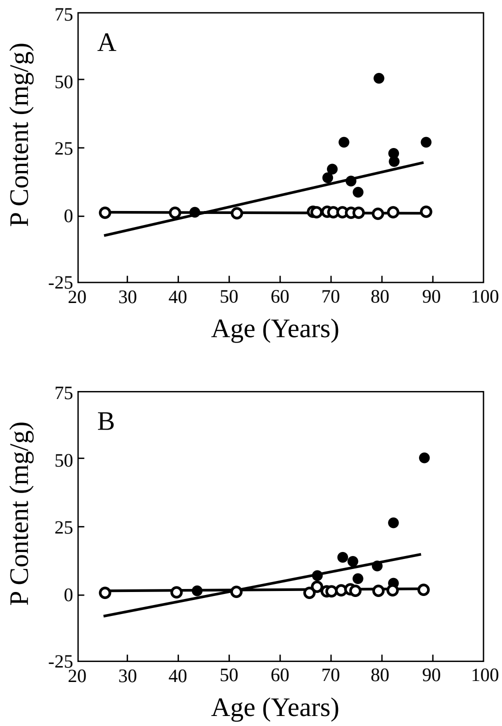

Figure 2. Age-related changes of P in the brachial and femoral arteries (A) and in the radial and posterior tibial arteries (B). In Fig. 2A, the open and closed circles denote the brachial and femoral arteries, respectively. In Fig. 2B, the open and closed circles denote the radial and posterior tibial arteries, respectively.

Age-related changes of S in the four arteries are shown in Fig. 3. The correlation coefficients between them were estimated to be 0.117 (p = 0.666) in the brachial arteries, -0.250 (p = 0.288) in the femoral arteries (Fig. 3A), -0.153 (p = 0.571) in the radial arteries and -0.255 (p = 0.278) in the posterior tibial arteries (Fig. 3B). No significant correlations were found between age and the S content in the four arteries.

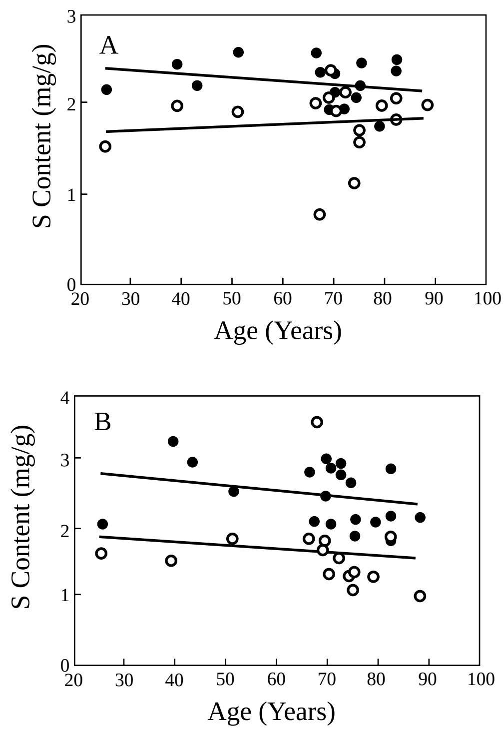

Figure 3. Age-related changes of S in the brachial and femoral arteries (A) and in the radial and posterior tibial arteries (B). In Fig. 3A, the open and closed circles denote the brachial and femoral arteries, respectively. In Fig. 3B, the open and closed circles denote the radial and posterior tibial arteries, respectively.

Figure 4A show age-related changes of Mg in the brachial and femoral arteries. The correlation coefficients between age and the Mg content were estimated to be 0.334 (p = 0.207) in the brachial arteries and 0.575 (p = 0.008) in the femoral arteries. There was a very significant direct correlation between age and the Mg content in the femoral arteries, but there was no significant correlation in the brachial arteries. Figure 4B shows age-related changes of Mg in the radial and posterior tibial arteries. The correlation coefficients between them were estimated to be -0.045 (p = 0.870) in the radial arteries and 0.427 (p = 0.060) in the posterior tibial arteries. Therefore, no significant correlations were found between them in the radial and posterior tibial arteries.

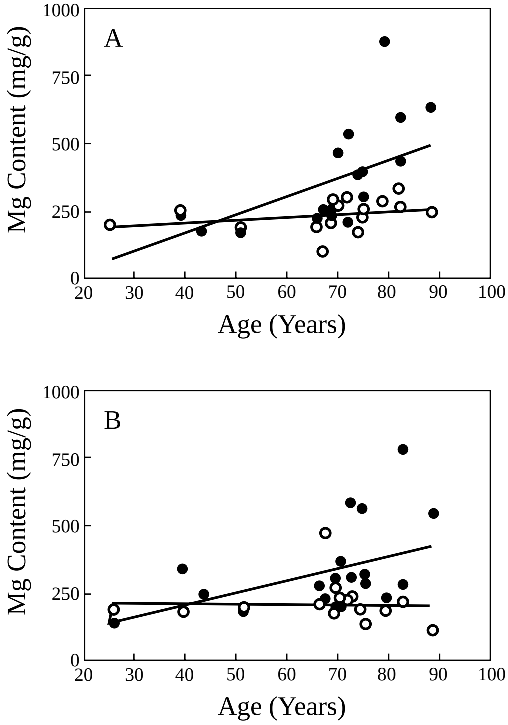

Figure 4. Age-related changes of Mg in the brachial and femoral arteries (A) and in the radial and posterior tibial arteries (B). In fig. 4A, the open and closed circles denote the brachial and femoral arteries, respectively. In Fig. 4B, the open and closed circles denote the radial and posterior tibial arteries, respectively.

Regarding the correlations between age and either Zn or Fe content, no significant correlations were found in the four arteries, except for age and the Zn content in the femoral arteries, in which the correlation coefficient was estimated to be 0.518 (p = 0.019).

Figure 5 shows comparisons of the average content of Ca and P among the four arteries by age group. In both the femoral and posterior tibial arteries, Ca and P began to increase in the sixties and thereafter increased to 2 to 9 times higher in the seventies and eighties compared with those in the sixties. By contrast, Ca and P hardly increased in the brachial and radial arteries with aging.

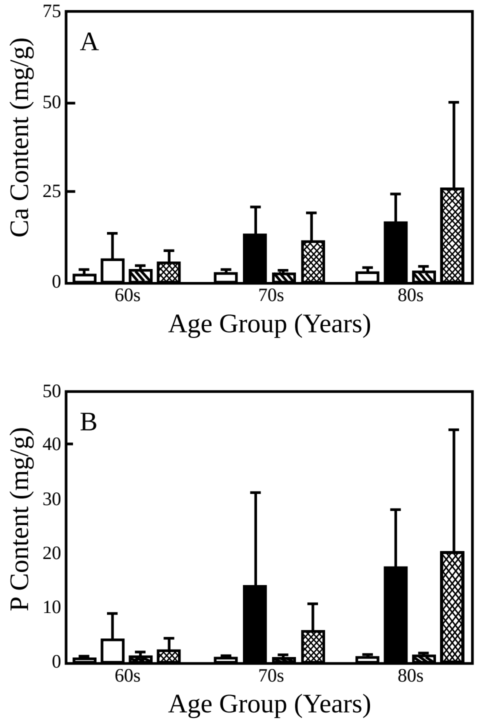

Figure 5. Comparison of the average content of Ca (A) and P (B) among the four arteries by age group. The open, shaded, hatched and crossed bars indicate the brachial, femoral, radial, and posterior tibial arteries, respectively.

Comparison of the Element Content Between the Arteries of the Upper and Lower Limbs

Table 1 shows the average content of elements in the arteries of the subjects over 60 yr. As for the arteries over 60 yr, the average contents of Ca were about 4-fold higher in the femoral and posterior tibial arteries in comparison with those in the brachial and radial arteries. Likewise, the average contents of P were 16- and 11-fold higher in the femoral and posterior tibial arteries compared with those in the brachial and radial arteries, respectively.

Table 1. Comparison of the Element Contents Among the Arteries over 60 Yr.

Artery	Average Content (mg/g)			
	Ca	P	S	Ng
Brachial	2.89±0.69	0.78±0.17	1.83±0.44	0.24±0.06
Radial	3.02±1.01	0.78±0.37	1.66±0.64	0.22±0.09
Pemoral	12.22±8.41	12.46±13.74	2.20±0.25	0.40±0.19
Post. Tibial	13.46±14.37	8.37±12.87	2.46±0.39	0.35±0.17

Relationships Among Elements in the Femoral and Posterior Tibial Arteries

Table 2 indicates the relationships among elements in the femoral arteries. Extremely-significant direct correlations were found among the contents of Ca, P, Mg, and Zn in the femoral arteries, whereas extremely significant inverse correlations were found between S and element contents, such as Ca, P, and Zn, and a very significant inverse correlation was found between S and Mg contents. In addition, significant correlations were found between Fe and element contents, such as Ca, P, and S.

Table 2. Relationships Among Element Contents in the Femoral Arteries.

Element	Correlation Coefficient and *p*-Value				
	P	S	Mg	Fe	Zn
Ca	0.935 (<0.0001)	-0.773 (<0.0001)	0.904 (<0.0001)	0.480 (0.032)	0.906 (<0.0001)
P		-0.768 (<0.0001)	0.954 (<0.0001)	0.536 (0.015)	0.900 (<0.0001)
S			-0.628 (0.003)	-0.449 (0.047)	-0.721 (0.0003)
Mg				0.415 (0.069)	0.884 (<0.0001)
Fe					0.374 (0.105)

Note: *p*-Values are indicated in parentheses.

The relationships among elements in the posterior tibial arteries are shown in Table 3. Extremely or very significant direct correlations were found among the content of Ca, P, and Mg in the posterior tibial arteries. In addition, very significant direct correlations were found between S and Fe contents and between Mg and Zn contents. These results indicated that regarding the relationships among the elements, there was a little difference between the femoral and posterior tibial arteries.

Table 3. Relationships Among Element Contents in the Posterior Tibial Arteries.

Element	Correlation Coefficient and *p*-Value				
	P	S	Mg	Fe	Zn
Ca	0.991 (<0.0001)	-0.321 (0.168)	0.690 (0.0008)	-0.108 (0.650)	0.367 (0.112)
P		-0.290 (0.215)	0.681 (0.001)	-0.113 (0.634)	0.274 (0.242)
S			0.113 (0.634)	0.584 (0.007)	-0.184 (0.439)
Mg				0.171 (0.470)	0.614 (0.004)
Fe					-0.030 (0.902)

Note: *p*-Values are indicated in parentheses.

Comparison of the Ca Content of the Femoral and Radial Arteries Between Thai and Japanese

Figure 6 shows the average content of Ca in both the femoral arteries of Thai and Japanese by age group. In the Sixties, the average content of Ca in the femoral arteries of Thai corresponded to three-tenths of that in the femoral arteries of Japanese. Although the average content of Ca was a little higher in the femoral arteries of Thai than in that of Japanese in the seventies, the average content of Ca was higher in femoral arteries of Japanese than in the arteries of Thai in the eighties. It should be noted that a high accumulation of Ca occurred in both the femoral arteries of Thai and Japanese with aging

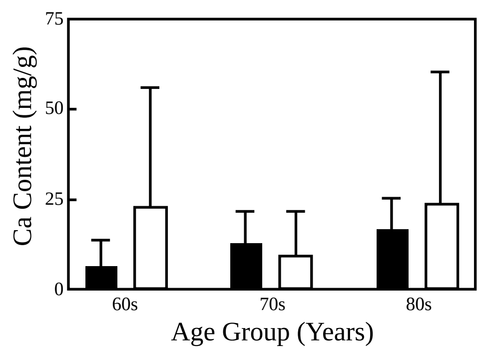

Figure 6. Comparison of the average content of Ca between femoral arteries of Thai and Japanese by age group. The shaded and open bars indicate Thai and Japanese, respectively.

The average content of Ca in both the radial arteries of Thai and Japanese by age group is shown in Fig. 7. The average content of Ca hardly increased in the radial arteries of Thai and Japanese with aging. No significant difference was found in the average content of Ca between the radial arteries of Thai and Japanese.

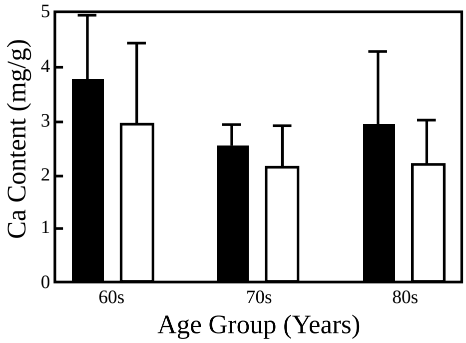

Figure 7. Comparison of the average content of Ca between the radial arteries of Thai and Japanese by age group. The shaded and open bars indicate Thai and Japanese, respectively.

DISCUSSION

The present study revealed that a high accumulation of Ca and P occurred in both the femoral and posterior tibial arteries with aging, but it hardly occurred in the brachial and radial arteries with aging.

Blumenthal et al., (1950) studied the rate of calcification in several major human arteries (aorta, coronary, hepatic, renal, and iliac arteries) under conditions of average intensity and frequency of calcification and found that calcification was most intensive in the iliac artery, followed in descending order by the coronary, renal, hepatic arteries, and aorta.

Kawasaki et al., (1987) studies static mechanical properties of major branches of the human arteries (abdominal aorta, common carotid, femoral, and brachial arteries) using an ultrasonic phase-locked echo-tracking system. They found that the stiffness in the brachial and femoral arteries increased with age more than in the proximal arteries and that these changes were not statistically significant, considering the wide range of individual values for stiffness. Their finding that the stiffness in the brachial artery increased with age is not compatible with our finding.

Regarding the popliteal artery, Cichocki et al., (1989) investigated the concentrations and localizations of elements in the artery by means of the proton-induced X- ray-emission (PIXE) and micro-PIXE methods and reported that the amount of Ca and P increased with age, approaching 20% and 9% at places, and mineral deposits were detected in the tunica media. Nemetschek-Gansler et al., (1979) studied the thickening intima of femoral arteries with special reference to elastin and found that deposits of Ca salt occurred in the interstices of the tunicae intima and media in subjects over 40 yr.

The authors (Tohno and Tohno, 1998a) previously investigated age-related changes of elements in the axillary and radial arteries of the upper limbs and the external iliac, femoral, popliteal, and dorsalis pedis arteries of the lower limbs in Japanese. It was found that a higher accumulation of Ca and P occurred in the arteries of the lower limbs with aging, whereas their accumulation hardly occurred in the arteries of the upper limbs. The present study is in agreement with the finding of the arteries in the upper and lower limbs of Japanese.

To examine whether the accumulation of elements in the aged arteries was related to the way of walking, the authors (Tohno, S. et al., 2001; Tohno, S. et al., 2003) recently investigated age-related changes of elements in the arteries of the upper and lower limbs in Japanese monkeys and found that an accumulation of Ca occurred slightly in both the arteries of the upper and lower limbs with aging and a Ca accumulation was 20-60% higher in the arteries of the lower limb than in the arteries of the upper limbs. It is clear that the difference in a Ca accumulation between the aged arteries of the upper and lower limbs is much smaller in Japanese monkeys than in Thai and Japanese human.

The authors (Tohno, Y. et al., 2001b and c; Tohno, Y. et al., 2003) previously investigated the relationships among the elements in the arteries of Japanese and found that as Ca and P increased in the arteries, such as the thoracic aorta, basilar, coronary, radial, femoral, and common iliac arteries, Mg increased simultaneously in all of the arteries. The present study demonstrated that as Ca and P increased in the femoral arteries, Mg increased simultaneously in the femoral arteries. The same finding was also obtained in the femoral arteries of Thai.

ACKNOWLEDGEMENT

The authors thank Dean Supot Wudhikarn, Faculty of Medicine, Chiang Mai University for providing an Opportunity of the study.

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