implications for understanding the unique characteristics of myocardium from hibernating animals, such as the cold tolerance and the markedly different effects of cardioactive agents[19–21]. The present result is important for further understanding of the cardiac excitation-contraction coupling.

1 Kondo, N., and Shibata, S., Science *225* (1984) 641.
2 Bassingthwaighte, J.B., Fry, C.H., and McGuigan, J.A.S., J. Physiol. *262* (1976) 15.
3 Siegelbaum, S.A., and Tsien, R.W., J. Physiol. *299* (1980) 485.
4 Sutko, J.L., and Kenyon, J.L., J. gen. Physiol. *82* (1983) 385.
5 Vereecke, J., and Carmeliet, E., Pflügers Arch. *322* (1971) 60.
6 Kohlhardt, M., Haastert, H.P., and Krause, H., Pflügers Arch. *342* (1973) 125.
7 Brehm, P., and Eckert, R., Science *202* (1978) 1203.
8 Mitchell, M.R., Powell, T., Terrar, D.A., and Twist, V.W., Proc. R. Soc. London. *B219* (1983) 447.
9 Kass, R.S., and Sanguinetti, M.C., J. gen. Physiol. *84* (1984) 705.
10 Eaton, D.C., and Brodwick, M.S., J. gen. Physiol. *75* (1980) 727.
11 Trautwein, W., Physiol. Rev. *53* (1973) 793.
12 Coraboeuf, E., in: The Slow Inward Current and Cardiac Arrhythmias, p.25. Eds D.P. Zipes, J.C. Bailey and V. Elharrar. Martinus Nijhoff Publishers, Hague/Boston/London.
13 Kenyon, J.L., and Gibbons, W.R., J. gen. Physiol. *73* (1979) 139.
14 Lederer, W.J., and Tsien, R.W., J. Physiol. *263* (1976) 73.
15 Kass, R.S., Lederer, W.J., Tsien, R.W., and Weingart, R., J. Physiol. *281* (1978) 187.
16 Colquhoun, D., Neher, E., Reuter, H., and Stevens, C.F., Nature *294* (1981) 752.
17 Chapman, R.A., Prog. Biophys. molec. Biol. *35* (1979) 1.
18 Mullins, L.J., Am. J. Physiol. *236* (1979) C103.
19 Kondo, N., Jap. J. Pharmac. *39* (1985) 131P.
20 Charnock, J.S., Dryden, W.F., Skoog, C., and Lauzon, P.A., Comp. Biochem. Physiol. *65B* (1980) 675.
21 Duker, G.D., Olsson, S.-O., Hecht, N.H., Senturia, J.B., and Johansson, B.W., Cryobiology *20* (1983) 407.

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The function of intimal longitudinal smooth muscles of the human coronary artery[1]

K. Kawasaki, T. Iino, H. Hasegawa, I. Miyazawa and S. Hosoda

Department of Cardiology, Jichi Medical School, Kawachi-Gun, Tochigi 329-04 (Japan), 17 January 1986

Summary. Bundles of smooth muscles in the intimal layer of the human coronary artery contracted in a longitudinal direction on the application of vasoactive substances. The data indicate that the human coronary artery contracts not only transversely but also longitudinally.
Key words. Human coronary artery; smooth muscles in intimal layer; longitudinal contraction.

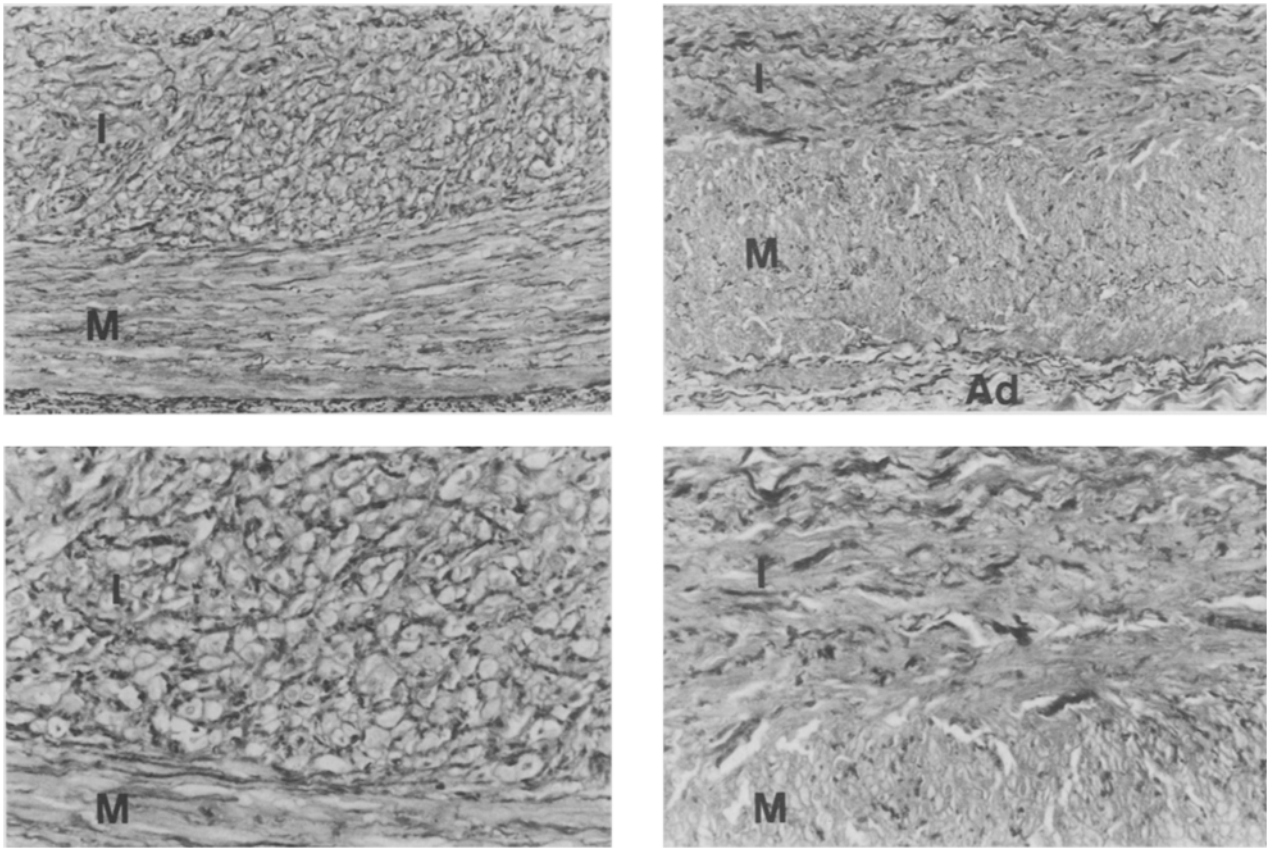

Figure 1. Left: a cross-section of human coronary artery for light microscopy. low magnification, × 35 (top); high magnification, × 70 (bottom). Right: a longitudinal-section of the same specimen. low magnification × 35 (top); high magnification, × 70 (bottom). Smooth muscle cells of the intima are arranged nearly parallel to blood stream. I, intima; M, media; Ad, adventitia. (elastica Van Gieson stain).

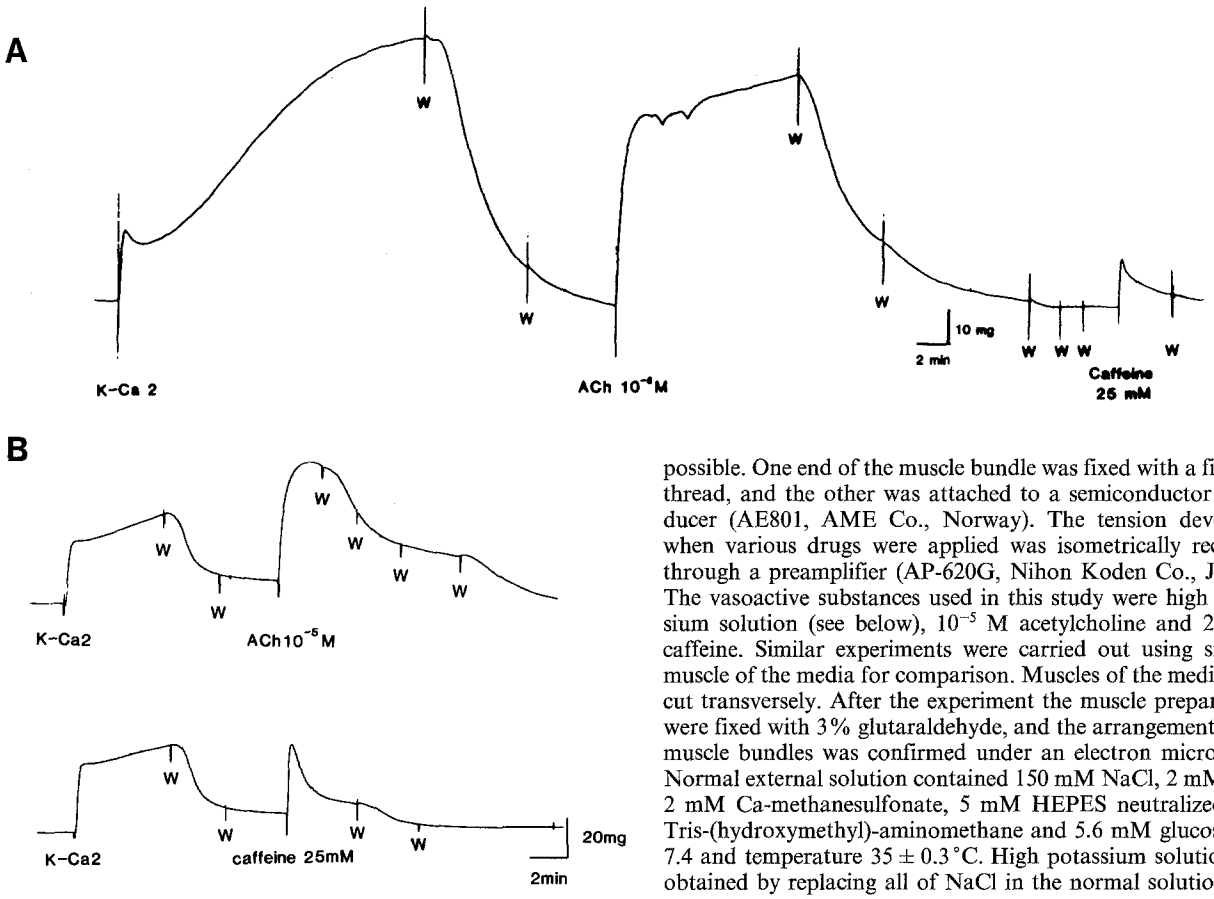

Figure 2. Isometric contractile responses of smooth muscle bundles of intima (A) and media (B) of human coronary artery to high potassium solution, 10^{-5} M acetylcholine, and 25 mM caffeine. The tensions developed by the intimal smooth muscle bundle were 77 mg, 68 mg, and 14 mg, respectively. The tensions developed by the medial smooth muscle bundle were 48 mg, 62 mg, and 36 mg, respectively. Na-Ca2, normal external solution; K-Ca2, high potassium solution; ACh, acetylcholine; W (wash), normal external solution.

The intimal layer of the coronary artery contains a number of smooth muscles, and its association with the development of atherosclerosis has attracted much attention[2]. However, the mechanical or pharmacological properties of the muscle have not been examined. To study the action of the muscle in the contraction of the human coronary artery, we isolated the bundles of intimal smooth muscles by a micro-dissection method, and measured the isometric tension developed when various drugs were applied.

Methods. The proximal portion of the right coronary artery was obtained from 5 cadavers (3 males, 2 females, aged from 45 to 70, average 61 years) within 2 h after death. The samples were taken from patients diagnosed as having cancer, peptic ulcer and toxicosis, in whom no heart disease had been clinically proved. The segment was cut longitudinally, fixed with pins, and then cut into rectangular sections (1 mm in width and 4 mm in length). Under a dark-field stereomicroscope, fibrous tissues and muscles were carefully removed from both the intimal and adventitial sides with a sharp knife (blade width, 2 mm) to produce a thin sheath-like specimen. Two different layers of a fibrous net that cross nearly at right angles were carefully detached from each other with forceps. From the intimal side containing the smooth muscles, a muscle bundle (25 μm in width and 1 mm in length) was isolated, and mounted in an organ bath with a volume of 0.9 ml in which rapid exchange of drug solutions was possible. One end of the muscle bundle was fixed with a fine silk thread, and the other was attached to a semiconductor transducer (AE801, AME Co., Norway). The tension developed when various drugs were applied was isometrically recorded through a preamplifier (AP-620G, Nihon Koden Co., Japan). The vasoactive substances used in this study were high potassium solution (see below), 10^{-5} M acetylcholine and 25 mM caffeine. Similar experiments were carried out using smooth muscle of the media for comparison. Muscles of the media were cut transversely. After the experiment the muscle preparations were fixed with 3% glutaraldehyde, and the arrangement of the muscle bundles was confirmed under an electron microscope. Normal external solution contained 150 mM NaCl, 2 mM KCl, 2 mM Ca-methanesulfonate, 5 mM HEPES neutralized with Tris-(hydroxymethyl)-aminomethane and 5.6 mM glucose (pH 7.4 and temperature 35 ± 0.3 °C. High potassium solution was obtained by replacing all of NaCl in the normal solution with K-methanesulfonate.

Results. Figure 1 shows a specimen for light microscopy prepared using half of the vessel segment obtained for this experiment. Smooth muscle cells of the intimal layer were arranged nearly parallel to the blood stream.

Longitudinal smooth muscle bundles of the intimal layer contracted in response to the application of high potassium solution, 10^{-5} M acetylcholine and 25 mM caffeine, and the tensions generated were 80 ± 3.5 mg (mean ± SE), 71 ± 2.8 mg, and 15 ± 1.5 mg, respectively. Smooth muscle bundles of the media also showed a contractile reaction to these substances, and the developed tensions were 49 ± 2.3 mg, 65 ± 3.2 mg, and 35 ± 1.2 mg, respectively (fig. 2).

As shown in figure 3, the arrangement of myofibrils in the intimal layer demonstrated that the contractile force of the muscle bundle was in the longitudinal direction.

Discussion. The intima of most muscular arteries of animals consists of endothelial cells and a basal lamina only[3]. However, lesions preceding fibrous plaque are known to occur in the intima of the human coronary artery at the early stage of atherosclerosis. This is known as the fibromusculoelastic lesion of the intima, which consists of proliferated smooth muscle cells surrounded by connective tissue[4]. Our study showed that smooth muscle bundles in the intimal layer of the human coronary artery, as well as those of the media, could be shown to contract when stimulated with various vasoactive substances. Blood vessels have been considered to contract only transversely. However, our findings show that on neuro-humoral or other stimulation human coronary arteries also contract longitudinally. In some special types of rat arteries, such as the coronary arteries and the helicine arteries of the ovary, uterus, penis[5] and lung[6], the intima contains bundles of longitudinally arranged smooth muscles. They have not been shown to change direction at various degrees of contraction[5]. Therefore, it has been emphasized that their function is clearly to block the blood flow, either partly or completely. Roach[7] discovered that bundles of longitudinal

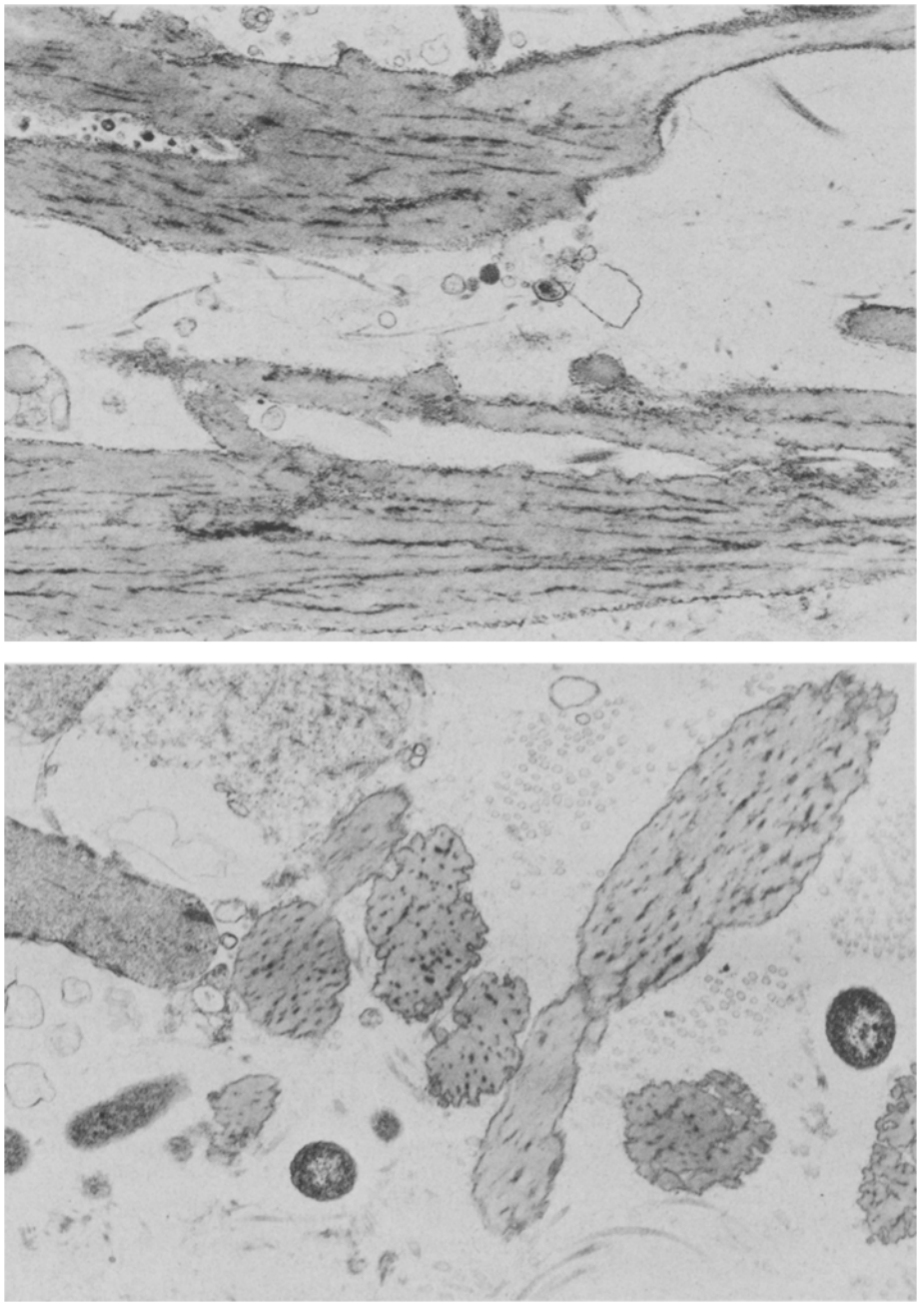

Figure 3. A longitudinal smooth muscle specimen for electron microscopy prepared after the measurement of tension. Top: Developed tension in the horizontal direction in this figure was measured. The array of myofibrils was longitudinal. Bottom: A cross-section of the same specimen. The ends of muscle fibers were circular to oval in shape. × 4000.

smooth muscles were present in the intima of the sheep's umbilical artery and proved that the muscle bundles pushed into the lumen and produced a stellate or crescentic appearance depending on the number of bundles. Some authors also consider that during longitudinal contraction, the increased volume of these smooth muscles may reduce the diameter of the vascular lumen, leading to complete occlusion upon additional transverse contraction of the smooth muscle of the media.

Smooth muscle of the intima reacted more strongly to potassium than to acetylcholine, while that of the media responded more strongly to acetylcholine. The contractile reaction of both types of smooth muscle was weakest with caffeine. Though only three agents were used in this study, responsiveness to various drugs seems to differ between these types of muscle.

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2 Parker, F., Am. J. Anat. *103* (1958) 247.
3 Rhodin, J. A. G., in: Handbook of Physiology, Eds D. F. Bohr et al. The Williams and Wilkins Company, Maryland 1980.
4 Haust, M. D., More, R. H., and Movat, H. Z., Am. J. Pathol. *37* (1960) 377.
5 Rhodin, J. A. G., in: Histology, Ed. J. A. G. Rhodin. Oxford Univ. Press, New York 1974.
6 Weibel, E., Z. Zellforsch. mikrosk. Anat. *47* (1958) 440.
7 Roach, M. R., in: Vascular Factors and Thrombosis, Eds. K. M. Brinkhous et al. Schattauer, Stuttgart 1970.

0014-4754/86/11/121222-03$1.50 + 0.20/0