CHANGES IN BRADYKININ LEVEL IN CORONARY SINUS BLOOD AFTER EXPERIMENTAL CORONARY OCCLUSION, AND THEIR RELATIONSHIPS TO HEMODYNAMIC CHANGES

By Keiichi Hashimoto, Soichi Furukawa, Masaru Hirose and Hirokazu Hayakawa

The cause of chest pain in angina pectoris and myocardial infarction is not yet clear at present. As one of its mechanisms Lewis, Katz, Raab and other authors ascribed it to the pain producing substances originated from the ischemic region of the myocardium.

More recently bradykinin, which is known to have strong pain producing and hypotensive actions, was suggested by Burch and DePasquale[1] as a substance responsible for the pain in angina pectoris. Sicuteri and co-workers[2] also stated the concept that bradykinin would be the cause of pain and cardiogenic shock in myocardial infarction. However, in these studies bradykinin itself was not determined.

The present work has been undertaken to study whether bradykinin is a possible cause of anginal pain and also whether it affects the cardiac function in ischemic heart disease. For this purpose the changes of bradykinin levels in the coronary sinus blood and of hemodynamics were examined after the experimental occlusion of a coronary artery in dogs.

Method: Dogs were anesthetized by the intravenous administration of thiopenthal sodium at a dose of 30 mg/Kg under respiratory control by the Bird's respirator. The chest was opened and the pericardium was incised along the right vagus nerve. A canula was inserted into the coronary sinus from the right auricle, and the origin of the left anterior descending artery was ligated. Through this canula, the blood was drawn before and 1, 2, 5, 10, 15, 30 and 60 minutes after ligation, respectively. For comparison, femoral venous blood was also drawn before the thoracotomy. ECG was monitored continuously from the beginning of the experiment to the end.

Blood samples thus obtained were then analyzed not only for bradykinin, but also for bradykininogen, an inactive precursor of bradykinin, and bradykininase, a bradykinin destroying enzyme. Bradykinin and bradykininase were measured by the method of Abe and co-workers[3,4] with some modifications and bradykininogen by the method of Brocklehurst and Zeitlin[5].

In order to study the changes of hemodynamics after coronary ligation, a 8F Cournand catheter was inserted into the cavity of left ventricle through the femoral artery. Heart rate, left ventricular systolic pressure, left ventricular end-diastolic pressure, and the maximum dp/dt were measured and the Veragut's index for myocardial contractility (max. dp/dt/IP)[6] was calculated before and 1, 2, 5, 10 minutes after coronary ligation together with measurement bradykinin. In addition, changes of these parameters were also examined in dogs after intravenous administration of 20.000 units/Kg of aprotinine (Trasylol), an inhibitor of bradykinin forming enzyme. The changes of these parameters after ligation were compared between the groups with and without pretreatment of aprotinine.

Results: Changes in bradykinin level after ligation of coronary artery. Bradykinin was measured in 14 dogs. The mean value of bradykinin in coronary sinus blood before ligation was 15.1 ng/ml, approximately twice the value of 7.4 ng/ml in the femoral venous blood before the thoracotomy. After the ligation of anterior descending artery, bradykinin levels in coronary sinus blood increased in almost all cases within 2 to 10 minutes. In 5 cases out of 14 examined, the maximum value was attained at 2 minutes after ligation, whereas in 6 cases it was attained at 5 minutes and in 2 cases at 10 minutes after the ligation. Only in one case, no elevation of bradykinin level was observed during the period. The maximum mean value of the elevated bradykinin was obtained at 5 minutes after the ligation and was calculated to be 52.2 ng/ml, which was three times higher than the mean value before ligation. In most cases the bradykinin level fell after 15 minutes and returned gradually to the level prior to the ligation after 60 minutes.

Changes in bradykininogen level after the ligation of coronary artery. The bradykininogen level in coronary sinus blood after thoracotomy was higher than that in femoral vein before thoracotomy. Following the ligation of coronary artery, it decreased in 7 out of 15 cases after 2 minutes and in 5 cases after 5 to 10 minutes. In other 3 cases no distinct changes were observed.

On an average, the bradykininogen level in femoral venous blood was 4.6 μg/ml before thoracotomy, 5.5 μg/ml in coronary sinus blood before ligation, 4.4 μg/ml at 2 minutes, 5.0 μg/ml at 5 minutes, 4.1 μg/ml at 10 minutes, 4.5 μg/ml at 15 minutes, 4.7 μg/ml at 30 minutes, and 4.0 μg/ml at 60 minutes after the ligation of coronary artery.

The changes in bradykininase activity after the ligation of coronary artery. Bradykininase was measured in 5 dogs. In 3 out of 5 cases the bradykininase activity in coronary sinus blood after thoracotomy was observed significantly higher than in femoral venous blood measured prior to thoracotomy. At 2 minutes after coronary ligation, it increased in 3 cases and was not altered in one case. In the remaining one case the activity decreased 2 minutes after ligation, but increased after 10 minutes. On an average, the bradykininase activity of 74% in coronary sinus blood after thoracotomy increased to 77.5% in 2 minutes after ligation, 77% in 10 minutes after, and then decreased gradually.

Changes in bradykinin level after ligation of coronary artery with pretreatment of aprotinine. No increase in bradykinin level was observed when the coronary artery was ligated after the intravenous administration of 20,000 units/Kg of aprotinine (Trasylol), an inhibitor of kinin forming enzyme.

The relationships between the changes of bradykinin and hemodynamics after the coronary ligation. Changes in hemodynamics after coronary ligation were compared between the groups with and without pretreatment of aprotinine in order to study whether these changes were due to bradykinin itself or not. The results obtained were as following: (1) no significant change in heart rate occurred in either group after the ligation of coronary artery, (2) left ventricular systolic pressure tended to rise after the ligation in the group without pretreatment of aprotinine, while no distinct change was observed in the aprotinine-treated group, (3) left ventricular end-diastolic pressure showed a remarkable rise in the former group, while any observable change scarcely occurred in the aprotinine-treated group after the ligation of coronary artery, (4) left ventricular maximum dp/dt had a tendency to rise in

Department of Internal Medicine, Nippon Medical School, Tokyo, Japan.

the former group, and, on the contrary, to decrease in the aprotinine-treated group after the ligation, and (5) Veragut's index fell 1 to 2 minutes after the ligation and then recovered to the previous value in the group without pretreatment, while in the aprotinine-pretreated group, it showed a remarkably high value after the ligation of coronary artery, indicating a different behavior.

Discussion: Marked elevation of bradykinin level in coronary sinus blood occurred at 2 to 5 minutes after the ligation of coronary artery. On the other hand, bradykininogen level showed a gradual decrease. Bradykininase activity slightly increased in 2 to 10 minutes and then showed a downward tendency. These findings indicate that when coronary artery was ligated, bradykininogen changed into bradykinin which was then decomposed by bradykininase.

The pain threshold dose of bradykinin has been reported by some authors that the pain occurs by intraarterial injection of 0.5 – 1 μg of synthetic bradykinin in human and 0.1 – 6.3 μg in dog. Uchida et al[7] showed the bradykinin level in the cubital venous blood to cause pain by ischemic arm exercise test was 20 – 30 ng/ml in human.

In our experiment, the average bradykinin level in coronary sinus blood was 52.2 mg/ml at 5 minutes after coronary artery ligation and the mean coronary sinus outflow was 15 ml/min. at this time. Therefore, the quantity of bradykinin, which flowed into coronary sinus, was calculated at approximately 0.8 μg/min. This quantity indicates that bradykinin level contained in the ischemic area was enough to cause pain. In addition, the elevated bradykinin level continued for about 10 minutes, almost coinciding with the duration of the attack of clinical angina pectoris.

Analysis of the relationships between bradykinin level and the hemodynamics revealed that the bradykinin level increased after the ligation of coronary artery, accompanied with the rise of left ventricular end-diastolic pressure and lowering of myocardial contractility as indicated by decrease of Veragut's index, while after administration of aprotinine no changes were observed in both of the bradykinin level and left ventricular end-diastolic pressure by coronary ligation, and the myocardial contractility showed a tendency rather to elevate. These findings suggest that bradykinin might be responsible for negative inotrophic state observed in clinical myocardial infarction.

In conclusion, when coronary artery was ligated bradykinin was produced in the ischemic myocardial tissues and its level is sufficiently high to produce pain, and, in addition, it might be a cause of cardiogenic shock.

REFERENCES

1. Burch, G. E. and DePasquale, N. P.: Am. Heart J., 65, 116, 1963.
2. Sicuteri, F. *et al.*: Malattie Cardiovasculari, 8, 343, 1967.
3. Abe, K. *et al.* Tohoku J. Exp. Med., 89, 103, 1966.
4. Abe, K. *et al.*: Tohoku J. Exp. Med., 86, 77, 1965.
5. Brocklehurst, W. E. and Zeitlin, I. J: J. Physiol., 191, 417, 1967.
6. Veragut, U. P. and Krayenbühl, H. P.: Cardiologia 47, 96, 1965.
7. Uchida, Y. and Ueda, H.: Jap. Heart J., 10, 503, 1969.