Plasma Level of Norepinephrine and Cyclic Nucleotides Following Acute Myocardial Infarction

Takashi Kondo, M.D., Kouichi Ogawa, M.D., Masaaki Ban, M.D., Bunyu Ogasawara, M.D., Eisuke Watanabe, M.D., and Tatsuo Satake, M.D.

Summary
The plasma concentrations of norepinephrine (NE), adenosine cyclic 3', 5'-monophosphate (cyclic AMP), and guanosine cyclic 3', 5'-monophosphate (cyclic GMP) were measured serially for 2 weeks after the onset of symptoms in 17 patients with acute myocardial infarction (AMI).

The mean concentrations of NE in patients without complications were significantly elevated during the first 2 days following AMI. There was a significant correlation between the maximum concentration of plasma NE and of plasma CK. The mean concentrations of plasma cyclic AMP and cyclic GMP in patients without complications were significantly elevated on the first day and for 8 days respectively following AMI. The concentration of plasma cyclic AMP on admission in patients with complications was significantly higher than that in those without complications. There were significant correlations between the maximum concentration of plasma cyclic AMP and those of plasma CK, GOT, and LDH. Significant but weak correlations between the concentration of plasma NE and those of cyclic AMP and cyclic GMP were found.

The results of the present study suggest an enhanced sympathetic nervous system activity at an early stage of AMI, a prolonged enhancement of parasympathetic nervous system activity in the course of AMI, and the potential value of plasma cyclic AMP concentration as a useful index to estimate the seriousness and size of AMI.

Additional Indexing Words:
Autonomic nervous system Cyclic AMP Cyclic GMP CK LDH GOT

WeBB et al1) reported a high incidence of autonomic disturbance at the onset of acute myocardial infarction (AMI). Moreover, experimental studies in animals show that coronary occlusion increases activity in sensory and motor nerves of sympathetic and parasympathetic cardiac nerves.2)−5)
It has been proposed that changes in plasma norepinephrine (NE) concentration reflect the activity of the sympathetic nervous system, and that adenosine cyclic 3', 5'-monophosphate (cyclic AMP) is the mediator of the β-adrenergic effects of catecholamine on various tissues, including myocardium. On the other hand, it is postulated that guanosine cyclic 3', 5'-monophosphate (cyclic GMP) may mediate the physiological response to activation of muscarinic receptors by acetylcholine as well as cholinergic stimulation.

These previous studies suggest that the concentration of cyclic AMP could be used as a measurable biochemical indicator for sympathetic nervous activity, and that of cyclic GMP for parasympathetic nervous activity. However, the intracellular metabolism of cyclic nucleotides cannot be studied in human, the available information being restricted to observations of extracellular cyclic nucleotides. Since cyclic nucleotides can escape from cells, changes in plasma concentration of the cyclic nucleotides could reflect alterations in intracellular concentrations.

In this study, in order to investigate the pathophysiology of the autonomic nervous system in AMI, we measured the plasma concentrations of NE, cyclic AMP, and cyclic GMP serially for 2 weeks following AMI.

**Patients and Methods**

Seventeen patients admitted with AMI within 24 hours of the onset of the symptoms were studied. There were 10 males and 7 females from 46 to 81 years in age with a mean of 63 years. The diagnosis of AMI was made by characteristic chest pain, serial electrocardiographic and cardiac enzyme changes. The location of the infarction was anterior in 13, and inferior in 4 cases. Of these 17, 12 patients had no complication and 5 had complications such as shock, heart failure, and serious arrhythmias requiring therapy. During the study patients were given morphine, furosemide, digoxin, lidocaine, and other drugs when indicated, but those who received drugs with known autonomic effects, prior to or during the sampling of blood, were excluded.

Blood samples were taken for the simultaneous estimation of the plasma concentrations of NE, cyclic AMP, and cyclic GMP on admission and thereafter serially at the same time early every morning for 2 weeks.

The concentration of plasma NE was assayed by a sensitive radioenzymatic method devised by Henry et al. and modified by Lake et al. The concentrations of plasma cyclic AMP and cyclic GMP were measured by a sensitive radioimmunoassay devised by Cailla et al. and modified by Honma et al. Normal values for plasma NE were obtained from 12 age-matched normal volunteers, and for plasma cyclic AMP and cyclic GMP from 36 age-matched normal volunteers.

Other serial laboratory determinations included serum creatine kinase (CK), glutamic oxaloacetic transaminase (GOT), and lactate dehydrogenase (LDH) activities until return to normal. In 9 patients, measurements of free fatty acids (FFA)
RESULTS

Plasma NE concentration following AMI

Fig. 1 shows serial concentrations of plasma NE following AMI in patients without complication. While the mean concentration of plasma NE in normal subjects was 0.23±0.02 ng/ml, that in patients with AMI was 0.55±0.11 ng/ml during the first day, decreasing gradually to 0.54±0.10 on the 2nd day, 0.41±0.11 on the 3rd day, and 0.39±0.09 on the 4th day with a subsequent return to normal values. Statistically, the concentrations of NE on the 1st and 2nd day were significantly higher when compared with normal values (p<0.01). The concentration of plasma NE on admission in patients with complications was 0.94±0.18 ng/ml. This was higher than that in patients without complication, but because of the large variation in values, not significantly so.

When plotting the maximum concentration of plasma NE against that of plasma CK, a significant correlation was found (r=0.55, p<0.05) (Fig. 2). However, there was no significant correlation between the maximum concentration of plasma NE and that of plasma GOT or LDH.

Furthermore, a positive significant correlation was noted between the concentration of plasma NE and that of plasma FFA determined simultaneously for 7 days following AMI (r=0.73, p<0.05) (Fig. 3).

Concentration of cyclic AMP and cyclic GMP following AMI
Fig. 2. Correlation between maximum plasma NE concentration and maximum plasma CK concentration.

Fig. 3. Correlation between plasma NE and FFA concentration.

Fig. 4. Plasma cyclic AMP concentrations following AMI without complications.
Fig. 4 shows the serial concentrations of plasma cyclic AMP following AMI in patients without complication. The mean concentration of plasma cyclic AMP in normal subjects was 17.6±0.71 pM/ml, and that in patients with uncomplicated AMI was 20.9±1.07 pM/ml at the time of admission, 21.0±1.68 on the 2nd day, and 21.1±18.0 on the 3rd day, decreasing thereafter and remaining at the same as the control level after the 4th day. The elevation in the cyclic AMP concentration on the 1st day following AMI was significant (p<0.02). The concentration of cyclic AMP on the 2nd and 3rd days tended to be higher than normal values. However, the difference was not significant, because of a wide scatter of values.

Fig. 5 shows serial concentrations of plasma cyclic GMP following AMI in patients without complication. The mean concentration of plasma cyclic GMP in normal subjects was 4.3±0.22 pM/ml. The value in patients with uncomplicated AMI was 9.3±1.19 pM/ml during the first 24 hours after the onset, a significant increase compared with the value in normal subjects. This significant increase in cyclic GMP concentration continued for 8 days following AMI.

Fig. 6 shows serial changes of the ratio of the cyclic AMP to cyclic GMP
The mean A/G ratio in normal subjects was 4.4±0.22 and that in patients with uncomplicated AMI was 2.6±0.35 on the 1st day. A/G ratio was maintained significantly below the control level (p<0.001) for 8 days following AMI.

The concentration of plasma cyclic AMP on admission in patients with complications was 34.0±3.9 pM/ml, significantly higher than that in patients without complications (p<0.01). On the other hand, the concentration of plasma cyclic GMP on admission in patients with complications was 12.6±1.7 pM/ml. This was higher than that in patients without complications, but not significantly so. The A/G ratio at the time of admission in patients with complications did not differ significantly from that in patients without.

There were positive, significant correlations between the maximum concentration of plasma cyclic AMP and maximum plasma LDH concentration.

Fig. 7. Correlation between maximum plasma cyclic AMP concentration and maximum plasma LDH concentration.

Fig. 8. Correlation between maximum plasma cyclic AMP concentration and maximum plasma CK concentration.
centration of plasma cyclic AMP and that of plasma LDH \((r=0.73, \ p<0.005)\) (Fig. 7), that of plasma CK \((r=0.57, \ p<0.05)\) (Fig. 8) and that of plasma GOT \((r=0.63, \ p<0.02)\) (Fig. 9) respectively. There was no significant correlation between the maximum concentration of cyclic GMP and those of the enzymes.

Furthermore, significant but weak correlations between the concentration of plasma NE and those of plasma cyclic AMP \((r=0.45, \ p<0.005)\), and cyclic GMP \((r=0.30, \ p<0.05)\) were found for all determinations obtained simultaneously following AMI.

![Graph showing correlation](image)

**Fig. 9.** Correlation between maximum plasma cyclic AMP concentration and maximum plasma GOT concentration.

**DISCUSSION**

The phenylethanolamine-N-methyltransferase based assay for NE used in this study is highly specific and more sensitive than any other existing radioenzymatic procedures\(^6),13\). In the present study, we demonstrated by serial determinations of plasma NE using this sensitive method that the plasma concentration of NE was significantly increased in the first 2 days after the onset of AMI. This result confirms the findings of many earlier works showing the increase of catecholamine concentration in urine\(^17)\-19\) and blood\(^20)\-24\) during the acute stage of AMI.

The significant correlation reported here between the maximum concentration of plasma NE and that of plasma CK suggests that the anatomical extent of myocardial infarction may be related to the initial increase of plasma NE concentration to a certain extent. Experimental studies in dogs demonstrating that NE was released from the ischemic tissue into the circulation, and that the infarcted myocardium lost its catecholamine content within the first
24 hours, also support this possibility. However, the general body response to pain, anxiety, and circulatory disturbances which could result in increased sympathetic nervous activity may also be responsible for the increased plasma NE concentration.

Furthermore, the significant correlation between the plasma concentrations of NE and FFA may reflect the mobilization of FFA from the adipose tissue by catecholamine.

There appear to be few reports in relation to the changes in plasma concentrations of cyclic AMP and cyclic GMP in human AMI. Strange et al and Rabinowitz et al, using a protein binding assay, studied plasma cyclic AMP levels in man following AMI. They reported elevation of plasma cyclic AMP concentration during the first few or 24 hours after the onset of AMI. We demonstrated using a sensitive radioimmunoassay that the mean concentration of plasma cyclic AMP was significantly elevated on the first day following AMI. Significant correlations between the maximum concentration of plasma cyclic AMP and those of plasma CK, GOT, and LDH, considered indicators of the extent of myocardial damage, were also found. In experimental studies, it has been shown that the myocardial cyclic AMP was increased, presumably as a result of catecholamine release, in the early stage of the acutely ischemic heart, and decreased in the chronic ischemic heart. Moreover, clinically Rabinowitz et al noted that some patients with AMI had a significantly greater level of cyclic AMP in coronary sinus blood than in the arterial blood simultaneously obtained. These results suggest that at least some of the initially increased cyclic AMP could be released from the heart itself.

The concentration of plasma cyclic AMP in patients with complications was significantly higher than that in patients without on admission. This suggests the potential value of the admission level of cyclic AMP as an index to estimate the seriousness of AMI.

A significant correlation between the plasma concentrations of NE and cyclic AMP provides support for the hypothesis that endogenous release of catecholamine induced by a pathologic condition might be the mechanism for increased plasma cyclic AMP concentration. However, AMI is also associated with other hormonal changes; an increase in plasma concentrations of cortisol and glucagon, and a decrease in plasma insulin concentration. The intracellular cyclic AMP may be the common denominator of several metabolic changes, and many of the hormones involved act by changing target cell concentration of cyclic AMP. Therefore, the increased concentration of plasma cyclic AMP may reflect generalized metabolic responses to the stress of AMI.
On the other hand, there was an increase in plasma cyclic GMP concentration which remained elevated until the 8th day of the disease, and a weak, but significant correlation between the plasma concentrations of NE and cyclic GMP was demonstrated in this study. This increase in plasma cyclic GMP in AMI has not been described before. In experimental study in dogs, Stewart et al\(^{32}\) reported that myocardial cyclic GMP was either normal or in some cases elevated in the chronic ischemic heart. Nakamura et al\(^{36}\) reported that myocardial cyclic GMP was increased transiently at 15 min after coronary ligation in the infarcted area, returning to the control level at 1 hour after. These results suggest that the heart itself seems unlikely to play an important part in the increase of plasma cyclic GMP. The precise origin of the increased level of plasma cyclic GMP and the implications of the significant correlation between the plasma concentrations of NE and cyclic GMP are not clear at present. However, since the rise in plasma cyclic GMP concentration was observed during the treatment with $\alpha$-adrenergic agent, suggesting the possibility that cyclic GMP metabolism is responsive either directly to $\alpha$-adrenergic stimulation or indirectly to parasympathetic stimulation which might occur as a reflexive consequence,\(^{37}\) this correlation may lend support to this possibility.

The early increases of plasma NE and cyclic AMP, which rapidly return to normal as the patients recover, and the long-lasting elevation of cyclic GMP and decreased A/G ratio, may suggest an enhanced sympathetic nervous system activity at an early stage of AMI and a prolonged, more profound augmentation of parasympathetic nervous system activity in the course of AMI. Furthermore, positive, significant correlations between the maximum concentration of cyclic AMP and those of CK, GOT, and LDH indicate that plasma cyclic AMP concentration could be used as an index to estimate the seriousness and size of AMI.

### References

11. Lee TP, Kuo JF, Greengard P: Role of muscarinic cholinergic receptors in regulation of guanosine 3', 5'-cyclic monophosphate content in mammalian brain, heart muscle and intestinal smooth muscle. Proc Nat Acad Sci USA 69: 3237, 1972
Myocardial and plasma levels of adenosine 3', 5'-cyclic phosphate. Studies in experimental ischemia. Chest 68: 69, 1975

32. Stewart D, Kamiyama T, Mason D, Miller R, Wikman-coffelt J: Alteration in canine cardiac basal levels of cAMP and cGMP, and elevated tissue pO₂ levels, induced by coronary ligation. J Mol Cell Cardiol 10: 125, 1978


