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Acute Cardiovascular Consequences of Anterior Descending Coronary Artery Occlusion in Unanesthetized Monkey[1, 2] (40157)

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It has been suggested that the monkey would provide a superior model for the study of experimental myocardial infarction since data could be projected more confidently to man. This contention is based principally upon anatomical evidence rather than upon studies of the physiological consequences of coronary occlusion (CO) in these animals. For instance, McNamara *et al.* (1) have demonstrated that ligation of the left anterior descending (LAD) coronary artery in rhesus monkeys produces an anterior wall infarct similar to that observed in the analogous situation in man. The distribution of this vessel is similar in man and monkey (1, 2). Nevertheless, the cardiovascular effects of ligation of this artery in the monkey have not been well documented. With respect to man, Webb *et al.* (3) have documented that the majority (54%) of those patients seen within 30 min of the onset of acute anterior myocardial infarction (MI) show tachycardia and transient hypertension. Conversely, most observers report decreases in blood pressure during CO in awake dogs and cats. It would obviously be instructive to determine the nature of the corresponding response in the non-human primate. It is, therefore, the purpose of this report to describe the acute cardiovascular response to occlusion of the LAD coronary artery in intact, unanesthetized monkeys.

Materials and methods. The following data are from eight chair restrained rhesus monkeys (*Macaca mulatta*, average weight = 7 kg) and 1 baboon (*Papio anubis*, weight = 25.5 kg). The experiments were conducted in isolation chambers in which the animals were observed via closed circuit television. Each subject was placed inside the chamber for 2 hr daily for a minimum of 2 weeks prior to surgical preparation for CO to adapt to the experimental environment. Careful attention to this phase of the study insured that the monkey would sit quietly during subsequent experiments with a heart rate which in most subjects seldom exceeded 140 per min during rest.[3]

The animals were sedated with 1 mg/kg phencyclidine hydrochloride and anesthetized with halothane in preparation for catheter and coronary artery snare implantations. Standard guidelines for the care of experimental animals were followed throughout. Catheters (0.03 in., ID) were inserted into the right axillary artery to measure arterial blood pressure and/or retrogradely into the left ventricle via the right carotid artery to measure left ventricular pressure. Konigsberg P4-S5 pressure transducers were also implanted through the apex of the heart in three monkeys. Stainless steel ECG electrodes were placed subcutaneously on the upper anterior right and left margins of the chest and the lower left anterior margin of the abdomen. The heart was visualized through a left thoracotomy so that a ligature could be placed loosely around the LAD coronary artery about ⅓ to ½ the distance from its origin to the apex of the heart. One animal had a

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[3] In conjunction with another experiment (20), the monkeys were also trained in a classical aversive conditioning paradigm during this preparation period by following a 1 min tone, the conditional stimulus to shock (CSs), by a short electric current (1 sec., 1 ma, 60 Hz pulsed-DC). The conditional and unconditional responses consisted of transient tachycardia, a pressor response and an increase in the rate of rise of left ventricular pressure. The CSs-shock combination was presented once or twice within the hour following coronary occlusion (cf Fig. 1), and may have consequently exacerbated the myocardial ischemia.

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second snare placed immediately distal to the origin of the LAD artery. None of the vessels was intramural at the point of snare placement. Spasm of the artery was prevented by discreetly painting the vessel with lidocaine prior to dissecting it free from the corresponding vein and connective tissue. Care was taken to minimize damage to nerves which coursed along the vessel; however, even though extraneous tissue was excluded when placing the snare, it may be presumed that nerves intimately associated with the artery were unintentionally included. The ends of the ligature were passed around the vessel and then through a length of silastic tubing filled with petroleum jelly. The length of suture which needed to be displaced to produce complete occlusion was measured; no artery was occluded for a protracted period during this procedure and all vessels were patent upon closure of the chest. The snare was stabilized against the myocardium such that the tip of the silastic tube was close to the vessel at the point of intended occlusion, thereby minimizing the mechanical strain experienced by the artery upon ligation. The coronary snare, catheters and ECG leads were tunneled under the monkey's skin and exited at its back. The incisions were closed and the monkeys dressed in a vest to protect the implants. The animals were rechaired and allowed 4 weeks to recover from the operation. They were placed inside the isolation booth for *ca* 2 hr daily during recuperation to maintain their familiarity with the experimental setting. The animal was placed inside the isolation chamber as usual on the day selected for CO, and allowed to sit quietly for at least 15 min. The door to the booth was then opened briefly to pull the snare, permanently occluding blood flow through the vessel; the procedure required less than 1 min. Medication for cardiac arrhythmias was withheld until it became apparent that treatment was necessary to stabilize the animal's condition. The experiment was considered to be complete upon initiation of this treatment or following ventricular fibrillation (VF).

All data were recorded on a Beckman model R dynograph and a Tandberg 115 fm tape recorder. Left ventricular pressure (LVP) was recorded from the Konigsberg transducer and/or by filling the catheter and Statham P23Gb transducer with a 60% glycerine solution in heparinized saline. The response of the catheter system was uniform to approximately 35 Hz with a damping of 0.6 of critical. LVP was differentiated using an active, high-pass filter with a corner frequency of 300 Hz. Standard limb leads of the ECG were recorded from the implanted electrodes. The number of ventricular premature beats per minute was determined from the ECG. Three ml of blood were drawn prior to CO and at 6, 24, 48 and 72 hr thereafter for measurement of serum enzyme concentrations (SGOT, LDH, CPK). The values of LVP, d(LVP)/dt and heart rate (HR) were read at successive one minute intervals starting 10 min prior to CO and continuing until the study was terminated. Data were examined for 60 min post-CO or until VF occurred. Statistical analysis was performed using a paired t test.

Eight animals survived the experiment, were sacrificed several weeks later, and their hearts cut into 5 mm thick slices. Forty μ thick sections were cut from each slice, stained (H + E), and examined microscopically to outline the infarction.[4] One monkey died within 2 hr of CO and microscopic examination of its heart revealed no obvious evidence of the MI. Alternatively, the hearts of two monkeys were scanned using a gamma scintillation camera following injection of Thallium-201 to outline the infarction by myocardial imaging (4).

Results. Postmortem examination (or imaging) revealed that all the animals had sustained a transmural infarction on the antero–lateral and anterior septal walls of the left ventricle. Some animals also evidenced a small involvement of the anterior wall of the right ventricle. Our findings in this respect are consistent with those of Hill *et al.* (5), who have provided a comprehensive description of the anatomical extent of comparable LAD coronary occlusions in *M. mulatta.* The production of acute myocardial infarction was confirmed shortly after CO by elevations

[4] We express our sincere appreciation to Dr. Grover Hutchins and his colleagues in the Department of Pathology, The Johns Hopkins Hospital, and Mrs. Jane Estes, Department of Physiology and Biophysics, University of Kentucky, for their help in the histology and postmortem examination of the hearts.

in serum enzyme levels in all monkeys. CPK increased from an average (±SD) of 18 ± 8 U prior to CO to a peak, 6 hr later, of 233 ± 155 U. SGOT changed from 23 ± 6 U to 103 ± 77 U at 24 hr, and LDH from 1176 ± 688 U to 1893 ± 1091 U, also at 24 hr. $P < 0.02$ in each instance. The monkeys also evidenced alterations in the ECG similar to those described by others (1, 5, 6).

The immediate hemodynamic consequences of LAD CO in monkey #4 are illustrated in Fig. 1. Each variable is plotted for consecutive 1 min intervals starting 10 min prior to CO (at 0 min), and extending some 90 min thereafter. Figure 2 shows selected polygraph tracings from this same monkey. Control data (open circles, Fig. 1; left panel, Fig. 2) were taken while the monkey rested quietly inside the booth. Blood pressure was stable and heart rate was approximately 100/min; LVEDP was about +6 mm Hg. The door was opened at 0 min to occlude the artery. Sustained changes in pressure and HR never resulted from such a disturbance in this or any other monkey. There was a period of bigeminal rhythm (BR) less than one-half min after occlusion. The BR lasted for about 1 min, after which the ventricular ectopic beats disappeared. Similar instances of short

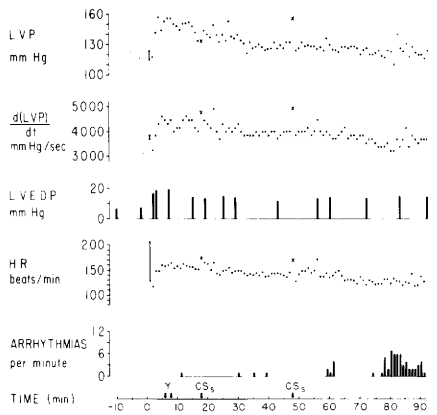

FIG. 1. Observations of cardiovascular variables before (open circles) and after (closed circles) LAD coronary occlusion in monkey #4. Bottom panel illustrates the number of arrhythmias recorded during any given 1 min interval. Maximum and minimum value for LVP, d(LVP)/dt and HR during bigeminal rhythm shortly after CO are shown connected by line. "Y" = periods of yawning. ("X" indicates observations made during a CSs - see footnote 3).

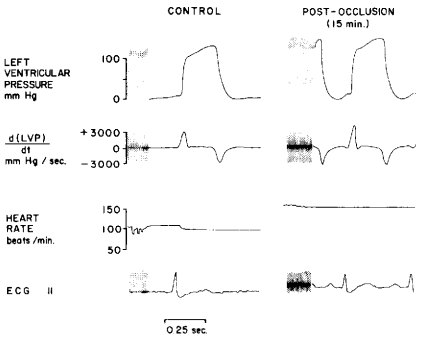

FIG. 2. Selected polygraph recordings for portions of record in Fig. 1. Control data are from 1 min prior to CO; post-CO from 15 min after occlusion. Data show pressor response, tachycardia and elevated d(LVP)/dt following CO.

latency, transitory arrhythmias were observed in three other animals. Note that by 3 min, LVP, d(LVP)/dt and HR were elevated above control. LVEDP was increased by more than 10 mm Hg at 2 min after CO. Although there may have been a transient depression in blood pressure immediately after CO in some animals, each monkey evidenced a pressor response associated with elevated filling pressure 3–5 min later. Visual observation of the subjects during the experimental session revealed that the cardiovascular responses were not associated with excessive movement. The right panel of Fig. 2 illustrates elevations in LVP, d(LVP)/dt and HR when monkey 4 was sitting virtually motionless. Scattered arrhythmias, due principally to premature ventricular ectopic complexes, were observed with increasing frequency after approximately 60 min. These arrhythmias occurred even as HR, LVP and d(LVP)/dt were gradually returning toward control. The increasing frequency of the arrhythmias eventually prompted termination of the study.

Results in Table I were computed from data for all animals and show mean ± SE for each variable from recordings made 1 min prior to CO (i.e., "control") and at selected intervals thereafter. The figure for 15 min is based upon only seven monkeys, and at 30 and 60 min, from six monkeys since three animals experiencing VF were eliminated from the pool. One animal had no LVP recording so that arterial systolic pressure was used; d(LVP)/dt was not available. These

TABLE I. ACUTE CARDIOVASCULAR RESPONSE TO ANTERIOR DESCENDING CORONARY OCCLUSION IN MONKEY.[a]

Time	Control	5 min	15 min	30 min	60 min
HR (/min)	130 ± 7	163 ± 10**	160 ± 6**	158 ± 8**	158 ± 8**
LVP (mmHg)	100 ± 4	120 ± 6**	122 ± 7**	115 ± 8*	114 ± 6*
d(LVP)/dt (mmHg/sec)	2580 ± 237	3600 ± 386*	3696 ± 467*	3296 ± 363*	3203 ± 375*
Number of animals	9	9	7	6	6

[a] Values are mean ± SE; * = $P < 0.05$; ** = $P < 0.01$.

findings confirm that HR, LVP and d(LVP)/dt were significantly elevated over preocclusion control values during the first hr after LAD CO. With respect to the "late" phase of arrhythmias, all but one animal (excluding those with early VF) experienced premature ventricular contractions ranging in time of onset from 40 to 90 min (mean 56 ± 18 min, SD).

One monkey was implanted with two snares. Pulling the first snare, placed approximately halfway down the LAD artery, produced the usual increases in LVP, d(LVP)/dt and HR; these data were used for calculating the results in Table I. When the remaining snare was pulled 3 weeks later, however, blood pressure dropped precipitously, whereas heart rate rose above 200/min. The animal died shortly thereafter from symptoms resembling cardiogenic shock.

There is considerable interest as to whether animals experience pain following experimental CO. None of the animals vocalized excessively or became unusually agitated during the experiment. Each of the monkeys did display the unusual behavior of exaggerated and prolonged episodes of "yawning" starting about 5 min after CO. Several of these episodes in monkey 4 have been designated on Fig. 1 by a "Y".

Discussion. This paper describes cardiovascular changes during the first hr after LAD coronary artery occlusion in the non-human primate. The responses which we have described were intimately associated with the CO. For instance, LVEDP was elevated very shortly after CO with the other hemodynamic variables responding concurrently or soon thereafter. The "yawning," which became a behavioral hallmark of the procedure, invariably occurred within 3–5 min of CO. Increases in blood serum enzyme concentrations were recorded in each animal during the hours immediately following these experiments. The response pattern just described seems to be characteristic of LAD occlusion. One other monkey with a snare around the great cardiac vein, but excluding the LAD artery, evidenced no sustained changes in pressure, HR or d(LVP)/dt. Likewise, three other monkeys which underwent occlusion of the marginal branch of the left circumflex (LC) artery, a small vessel supplying a restricted portion of the lateral left ventricular free wall, also showed no significant responses (excepting infrequent arrhythmias) under otherwise identical conditions. Conversely, two monkeys subject to complete occlusion of the LC artery evidenced a depressor response and bradycardia with little or no change in d(LVP)/dt (unpublished observations). It, therefore, seems safe to conclude that each animal did experience myocardial ischemia concomitant with pulling the snare, and that the phenomenon we observed was directly linked with the specific act of LAD coronary occlusion.

The literature on the cardiovascular effects of CO in the monkey is meager. To date, the largest body of data has been provided by Hill *et al.* (5, 7), who described ECG and hemodynamic changes during LAD ligation in 26 rhesus monkeys. They also described premature ventricular contractions immediately after CO. Our results diverge, however, with respect to hemodynamic findings since they report a decrease in blood pressure and no change in heart rate during the several hours following occlusion. This discrepancy appears to be associated with the unusually high preocclusion HR (>200/min) for their animals. The authors stated that sufficient time was probably not allotted for recovery from surgery prior to participation in the experiments. In addition, their monkeys were not previously adapted to chair restraint, which probably precipitated considerable stress in these animals with a consequent

elevation of sympathetic discharge prior to CO.

Our report of an increase in LVP and d(LVP)/dt following coronary occlusion appears to be unique in the animal literature. Hood *et al.* (8) reported a decline in LVP and d(LVP)/dt following gradual CO in dogs. The changes they observed in HR and LVEDP resemble our findings. Heyndrickx *et al.* (9) and Theroux *et al.* (10) also reported an increase in HR during an acute, circumflex CO in unanesthetized dogs; LVP and d(LVP)/dt were slightly depressed. The exact nature of the hemodynamic response will obviously vary with the experimental conditions. For instance, probable differences between LC and LAD occlusions have already been mentioned. More extensive infarctions due to occlusions closer to the left main coronary artery might be expected to yield different results from those reported here (11). Results from our one animal with two snares support this possibility, since occluding the LAD near its origin produced a marked decrease in pressure rather than the usual pressor response.

In addition to the hemodynamic changes described above, we also observed instances of cardiac arrhythmias following CO. Four animals evidenced two phases, one occurring within minutes of occlusion with a second, more sustained period, following within an hour or less. This pattern is similar to that described in the unanesthetized pig following LAD occlusion (12). Two phases of arrhythmias have also been described in dog (13). The first phase begins almost immediately after coronary ligation; after a period of quiescence, the late phase in dog begins at about 6–9 hr. The early arrhythmias we saw may have been due to the incipient myocardial ischemia. However, although minimal mechanical strain was placed upon the vessel during the process of occlusion, it is possible that even a small deformation of the artery could have activated those few nerve fibers still associated with the artery, thereby triggering the arrhythmia. We have no evidence to support or refute this hypothesis. It seems less likely that this factor could have been associated with the later phase of arrhythmias. Four of our animals experienced VF. Our data in this respect, however, are not representative of what might be expected of LAD CO in monkey, since appropriate drug therapy was initiated when arrhythmias became frequent.

Though somewhat different from the animal literature, the changes in LVP and HR described herein do resemble those reported in acute MI in man. Webb *et al.* (3) described tachycardia and/or pressor responses in 54% of those patients seen within 30 min of onset of acute anterior MI; only 32% evidenced bradycardia and/or depressor responses. Posterior MI typically resulted in decreased HR and blood pressure. They attributed the predominate response to anterior MI to exaggerated sympathetic cardiac drive. Corr *et al.* (14) have also demonstrated that the nature of the cardiovascular responses to CO in the cat depends upon the location of the lesion, as also seems to be the case in man (3), but even their data do not show pressor/tachycardia responses characteristic of elevated sympathetic drive.

Elevations in pressure, d(LVP)/dt, HR and the frequency of cardiac arrhythmias are suggestive of augmented sympathetic nervous activity. Reflex increases in sympathetic activity have been described (15, 16) subsequent to CO. The response which we observed to LAD CO may represent a physiologically significant activation of these (cardio-cardiac) reflexes. The possibility that a general sympathetic discharge results secondary to pain induced by CO must also be considered, particularly since no sedatives were used. It is difficult to assess pain in these animals, but we noted no subjective evidence of sustained discomfort. The only consistent behavioral pattern following CO which we can reliably report is the excessive yawning. Yawning has been observed in monkeys during mild stress or "uneasiness" (17, 18), and following intracisternal administration of ACTH (19). More research into the etiology of this response pattern is certainly warranted.

Summary. The acute response of the unanesthetized monkey to occlusion of the anterior descending coronary artery consists of a pressor response, tachycardia and increase in d(LVP)/dt. This response resembles that reported for comparable situations in man, and may be due to elevated cardiac sympathetic tone. Anatomical and physiological

characteristics of the non-human primate suggest that these animals provide attractive models for experimental study of anterior MI.

1. McNamara, J. J., Smith, G. T., Suehiro, G. T., Soeter, J. R., Anema, R. J., Morgan, A. L., Jr., and Liao, S. K., J. Thoracic Cardiovasc. Sur. **68,** 248 (1974).
2. Chase, R. E., Amer. J. Phys. Anthrop. **23,** 299 (1938).
3. Webb, S. W., Adgey, A. A. J., and Pantridge, J. F., Br. Med. J. **3,** 89 (1972).
4. Strauss, H. W., Harrison, K., Langan, J. K., Lebowitz, E., and Pitt, B., Circulation **51,** 641 (1975).
5. Hill, J., Malinow, M., McNulty, W., and Ochsner, A., Amer. Heart J. **84,** 82 (1972).
6. Lapin, B. A., Kechker, M. I., Kantaria, P. M., and Cherkovich, G. M., Exp. Pathol. **10,** 199 (1975).
7. Hill, J. D., Lees, M. H., Malinow, M. R., Oschsner, A. J., III, and Thomas, C., Cardiovasc. Res. **7,** 375 (1973).
8. Hood, W. B., Jr., Joison, J., Kuman, R., Katayama, I., Neiman, R. S., and Norman, J. C., Cardiovasc. Res. **4,** 73 (1970).
9. Heyndrickx, G. R., Millard, R. W., McRitchie, R. J., Maroko, P. R., and Vatner, S. F., J. Clin. Inv. **56,** 978 (1975).
10. Theroux, P., Ross, J., Jr., Franklin, D., Kemper, W., and Sasayama, S., Circulation **53,** 302 (1976).
11. Bakshandch, K., Kaiser, G. A., Bolooki, H., Surgical Forum **23,** 152 (1972).
12. Skinner, J. E., Lie, J. T., and Entman, M. L., Circulation **51,** 656 (1975).
13. Harris, A. S., Circulation **1,** 1318 (1950).
14. Corr, P. B., Pearle, D. L., Hinton, J. R., Roberts, W. C., and Gillis, R. A., Circ. Res. **39,** 840 (1976).
15. Mallinani, A., Schwartz, P. J., and Zanchetti, A., Amer. J. Physiol. **217,** 703 (1969).
16. Peterson, D. F., Kaspar, R. L., and Bishop, V. S., Circ. Res. **32,** 652 (1973).
17. Hall, K. R., Nature 196, 1258 (1962).
18. Hinde, R. A., and Rowell, T. E., Proc. Zool. Soc. Lond. **138,** 1 (1962).
19. Gessa, G. L., Vargiu, L., and Ferrari, W., Nature (London) **211,** 426 (1966).
20. Randall, D. C., Hasson, D. M., and Brady, J. V., Physiologist **18,** 361 (1975).

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