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Cardiac Taurine Levels during Endotoxemia (41579)

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Abstract. Three hours after dogs were given an intravenous injection of *Escherichia coli* endotoxin significant decreases in cardiac taurine levels were observed in the apex and epicardial regions of the right ventricle and in all regions sampled from the left ventricle. A decrease in taurine was seen in all regions of the heart (including the atria) by 90 min after endotoxin treatment but the results were not statistically significant. Echocardiography and left ventricular cannulation were used in a separate group to confirm that the dose of endotoxin used was adequate to produce depression of cardiac output and force of contraction.

Taurine (2-aminoethanesulfonic acid) is one of the most abundant free amino acids in cardiac tissue (1). However, the role of taurine in the myocardium is unknown. Recent evidence suggests that taurine is involved with the regulation of calcium fluxes across the cardiac sarcolemma (2, 3). In this context McBroom and Welty (4) and Azari *et al.* (5) have demonstrated that taurine protects the cardiomyopathic hamster from calcium overload. In addition Kramer *et al.* (6) have shown that taurine protects the rat heart against loss of mechanical function in the calcium-paradox model.

It has been demonstrated in a variety of species including man that taurine is released from the damaged myocardium. Our laboratory has demonstrated that cardiac insults such as a coronary artery occlusion in the dog (7), anoxic perfusion of rat ventricles (8), drug-produced myocardial necrosis in rats (9), and acute myocardial infarction in man (10, 11) have all produced depletions in the taurine content of cardiac tissue. It has also been reported that rat hearts subjected to the calcium-paradox and global ischemia heart failure models (6) and turkey poults treated with furazolidone to produce a cardiomyopathy (12) had significantly reduced cardiac taurine levels.

Since endotoxin shock has been shown to produce myocardial damage (13) the present study was designed to explore the release of endogenous taurine from cardiac muscle after an intravenous injection of endotoxin.

Materials and Methods. *Release of endogenous taurine from cardiac tissue.* Adult mongrel dogs of either sex, weighing 12 to 20 kg, were anesthetized with pentobarbital sodium (30 mg/kg). A tracheotomy was performed and catheters were inserted into the left femoral artery and vein. Arterial blood pressure was monitored via the femoral catheter using a Statham P23 Db pressure transducer and a Beckman R-411 recorder. The animals were respired with room air using a Harvard large animal respirator. The femoral vein catheter served as the injection site for the endotoxin and additional pentobarbital sodium as needed to maintain a light stage III level of anesthesia. *Escherichia coli* endotoxin (Difco Labs—*E. coli* 055.B5) was administered at a dose of 0.5 mg/kg in 10 ml of physiological saline to the 90-min ($n = 8$) and 180-min ($n = 8$) experimental groups.

At the designated time periods a left thoracotomy was performed and the heart was excised and chilled on ice. Dissection of the heart into the appropriate regions for taurine analysis was carried out immediately. A portion of each sample was weighed, dried, and reweighed to determine the dry/wet ratio. The remainder of the tissue sample was homogenized in two volumes of 2% perchloric acid and centrifuged for 15 min at 10,000*g*. Aliquots of the supernatants were analyzed for taurine content on a Beckman 121 amino acid analyzer using 0.2 *N* sodium citrate buffer, pH 2.4.

Measurement of cardiac dynamics. Echocardiography and left ventricular cannulation were used to monitor cardiac function in an additional group of dogs ($n = 8$) given endotoxin. Cardiac output was calculated from

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the echocardographic tracing using previously described techniques (14). More recently, studies in this laboratory have demonstrated a high degree of correlation ($r = 0.96$) between cardiac output determined by utilizing echocardiography and the dye dilution technique in the dog (15). Mean arterial blood pressure (MAP) was calculated from the femoral catheter pressure tracings.

Statistical method. Analysis of variance was used to compare the cardiac tissue taurine levels of both the 90-min and 180-min groups with control values. Duncan's multiple range test was used to evaluate the MAP, *dP/dt,* and cardiac output data. Significance was accepted at $P < 0.05$.

Results. Taurine levels in cardiac tissue samples autopsied from the left and right atria were not statistically different from control values at either of the times tested (Fig. 1). The average taurine content in both right and left atria appeared to decrease more rapidly during the 0- to 90-min time period than during the 90- to 180-min time period.

Taurine levels of right ventricular samples are depicted in Fig. 2. Again it can be observed that the decrease in taurine content appeared more rapid during the 0- to 90-min period in all tissues although none of the taurine values in the 90-min samples were found to be statistically different from the control values. A significant decrease in taurine content was found to exist in two (apex and epicardial transmural sections) of the five regions obtained from the right ventricle at the 180-min time period.

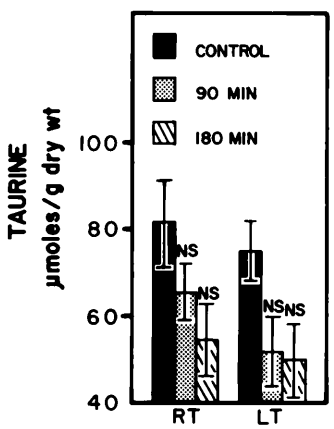

FIG. 1. Effect of endotoxin shock on taurine content in dog atria. Hearts were excised at the designated time periods after endotoxin injection, chilled on ice, and dissected. Hearts from control dogs were excised after 180 min of anesthesia following duplicate experimental conditions for the endotoxin-treated dogs. Each bar represents the mean taurine value from eight animals; SEM are represented by the brackets. (NS = nonsignificant; RT = right; LT = left.)

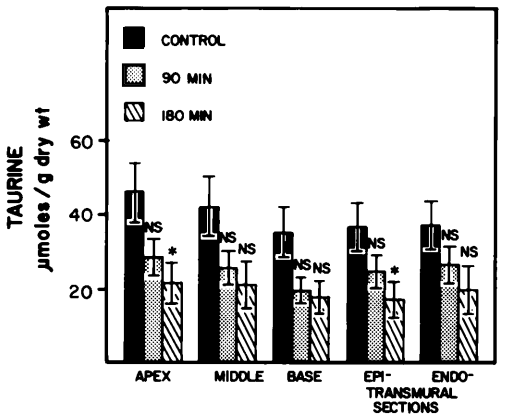

FIG. 2. Effect of endotoxin shock on taurine content in various regions of the dog right ventricle ($n = 8$). Experimental conditions are described in the legend to Fig. 1. Asterisk denotes significant difference from control value ($P < 0.05$). (EPI- = epicardial; ENDO- = endocardial.)

Taurine levels in all regions obtained from the left ventricle seemed to decrease more rapidly during the 0- to 90-min time period than during the 90- to 180-min time period (Fig. 3). However, at 90 min the only tissue showing a significant decrease in taurine content was the anterior papillary muscle. At the end of 180 min all of the regions demonstrated a significant decrease in taurine content.

A significant decrease in MAP (Fig. 4A), cardiac output (Fig. 4B), and *dP/dt* (Fig. 4C) occurred within 2 min after the endotoxin injection. A partial recovery in cardiac output occurred by 10 min post injection; however, a second, more pronounced decrease at 120 min depressed cardiac output to 35% of the control level. Left ventricular *dP/dt* improved slowly after the initial decrease. However, it remained significantly below control values for the entire 3-hr experimental period. The MAP response (Fig. 4A) seen in this group was not significantly different from that of the groups used for cardiac taurine determinations. A significant decrease in MAP occurred

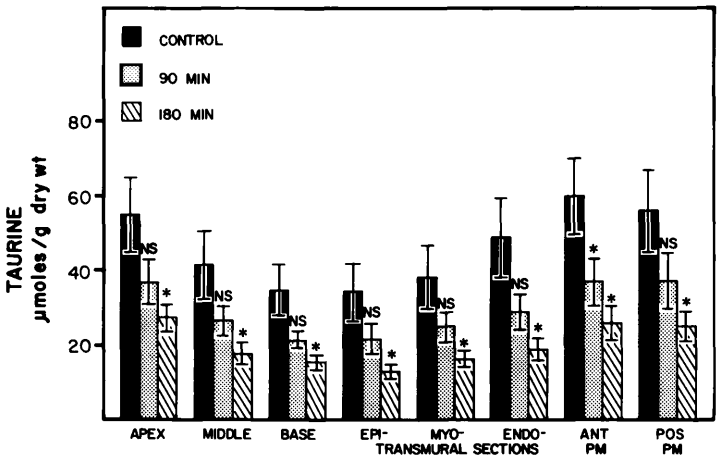

FIG. 3. Effect of endotoxin shock on taurine content in various regions of the dog left ventricle (n = 8). Experimental conditions are described in the legend to Fig. 1. Asterisk denotes significant difference from control value ($P < 0.05$). (EPI- = epicardial; MYO- = myocardial; ENDO- = endocardial; ANT PM = anterior papillary muscle; POS PM = posterior papillary muscle.)

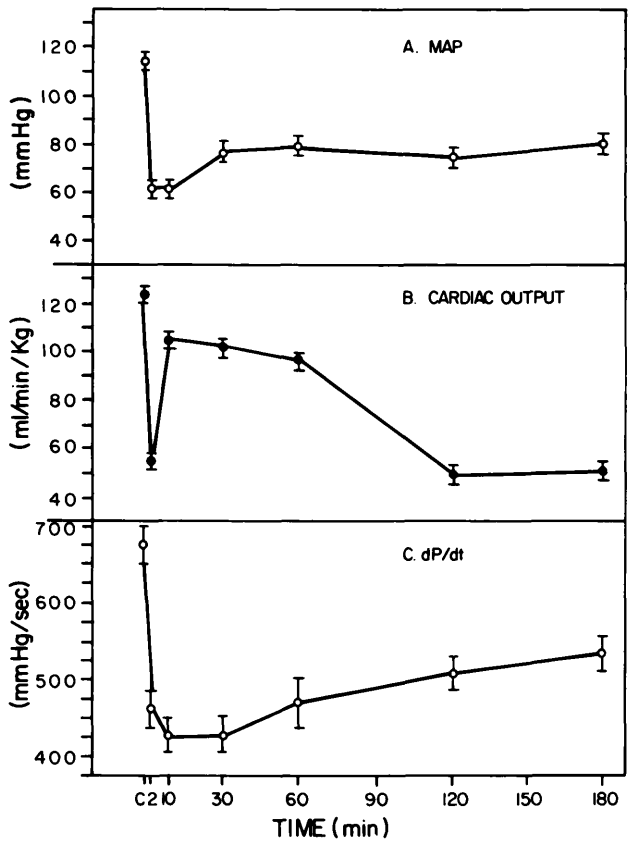

FIG. 4. Mean arterial pressure (A), cardiac output (B), and dP/dt (C) are shown for the group of animals monitored with echocardiography and left ventricular cannulation. This group was given endotoxin immediately following the control (c) period. The Duncan's multiple range test was utilized to determine significant differences between time points.

within 2 min after the endotoxin injection. Although a partial recovery of MAP occurred, it remained significantly below the control value for the 3-hr experimental period.

Discussion. It has been demonstrated that the intravenous injection of endotoxin produces myocardial damage (13), alterations in calcium flux (16), myocardial ischemia (17), and an increase in plasma taurine levels (18). The underlying mechanism of this damage remains unknown. Several possible explanations include damage produced by endogenously released compounds, a direct effect of endotoxin, or by an increased myocardial demand caused by elevated peripheral resistance in the presence of decreased ventricular filling.

Previous studies from our laboratory have suggested that if cardiac tissue is damaged by exposure to prolonged anoxia or ischemia, tissue taurine levels will be depleted (7–11). The apparent relationship between cardiac function and taurine remains unknown; however, stabilization of membranes (19) and involvement in calcium transport have been suggested as possible mechanisms (20–22).

The present study has shown that cardiac tissue levels of taurine are significantly depleted within 3 hr after administration of endotoxin. Although all of the regions of the cardiac muscle demonstrated a reduction in taurine levels, those areas showing a statistically significant decrease were found primarily in the left ventricle (eight of eight regions sampled—100%). In the right ventricle the number of regions demonstrating a significant decrease in taurine content was less (two of five regions sampled—40%). No significant difference could be found in either of the atria (zero of two regions sampled—0%). These results may reflect the stress placed upon the left ventricle as a result of increased peripheral resistance, which occurs in the latter stages of endotoxin shock (23).

A high taurine concentration is found in normally functioning cardiac tissue, and this taurine content is depleted when myocardial damage occurs (7–12). Based upon these previous studies it appears that taurine depletion can be used as an indicator of myocardial damage. Since in the current study the rate of taurine loss was substantially greater in the 0- to 90-min period than in the 90- to 180-min period, this would suggest that the onset of myocardial damage occurs shortly after the endotoxin bolus injection. Although only speculation, it is suggested that the onset of tissue taurine loss may signal the onset of myocardial damage and that the final tissue taurine content may be an indicator of the extent of myocardial damage.

The data in Fig. 4 illustrate that the dose of endotoxin used in these experiments was adequate to significantly alter MAP, cardiac output, and left ventricular *dP/dt*. Previous work in this laboratory has utilized the extrasystole potentiation technique (24) to investigate the extent of myocardial depression produced by endotoxin. A dose of endotoxin identical to that used in the current study was shown to decrease the augmentation of contractility, which remained at 25–35% of the control value throughout the 3-hr experimental period (25). This latter technique has been reported to measure myocardial performance independent of filling volumes (24). The decreasing taurine levels observed during endotoxemia would suggest that these depressed physiological parameters may be due to the loss of cellular integrity in the myocardium and/or altered calcium homeostasis.

The present study has shown that during the 3-hr period after an endotoxin bolus there is an insignificant decrease in the taurine concentration of atrial tissue. The taurine concentrations in all sampled regions of the left ventricle and in certain areas of the right ventricle demonstrated a significant decrease during the same time period. The rate of this taurine decrease appears to be more rapid during the early stages of the shock response; however, a significant decrease was not noted until after 90 min. If, as suggested by previous investigators (2, 3), taurine is important in the regulation of calcium fluxes, the rapid decrease in this amino acid may be involved in the calcium transport abnormalities that exist during endotoxin shock (16).

Although the dose of endotoxin utilized in these experiments is lower than that used in most other experimental shock studies (16, 18, 23), significant decreases in both the hemodynamic parameters and taurine content of the cardiac tissue were observed. It would be of future interest to determine if the observed changes in tissue taurine content or

depletion rate could be altered with different doses of endotoxin or attenuated with drugs reported to be beneficial in the treatment of endotoxin or septic shock.

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1. Jacobsen JG, Smith LH Jr. Biochemistry and physiology of taurine and taurine derivatives. Physiol Rev **48**:424–511, 1968.
2. Chovan JP, Kulakowski EC, Benson BW, Schaffer SW. Taurine enhancement of calcium binding to rat heart sarcolemma. Biochim Biophys Acta **551**:129–136, 1979.
3. Chovan JP, Kulakowski EC, Sheakowski S, Schaffer SW. Calcium regulation by the low-affinity taurine binding sites of cardiac sarcolemma. Mol Pharmacol **17**:295–300, 1980.
4. McBroom MJ, Welty JD. Effects of taurine on heart calcium in the cardiomyopathic hamster. J Mol Cell Cardiol **9**:853–858, 1977.
5. Azari J, Brumbaugh P, Huxtable R. Prophylaxis by taurine in the hearts of cardiomyopathic hamsters. J Mol Cell Cardiol **12**:1353–1355, 1980.
6. Kramer JH, Chovan JP, Schaffer SW. Effect of taurine on calcium paradox and ischemic heart failure. *Amer J Physiol* **240**:H238–H246, 1981.
7. Crass MF III, Lombardini JB. Loss of cardiac muscle taurine after acute left ventricular ischemia. Life Sci **21**:951–958, 1977.
8. Crass MF III, Lombardini JB. Release of tissue taurine from the oxygen-deficient perfused rat heart. Proc Soc Exp Biol Med **157**:486–488, 1978.
9. Lombardini JB. Effects of isoproterenol and methoxamine on the contents of taurine in rat tissues. J Pharmacol Exp Ther **213**:399–405, 1980.
10. Lombardini JB, Cooper MW. Elevated blood taurine levels in acute and evolving myocardial infarction. J Lab Clin Med **98**:849–859, 1981.
11. Balch JA, Lombardini JB. Taurine concentration in acute myocardial infarction. Pharmacologist **23**:220, 1981.
12. Schaffer SW, Czarnecki CM, Cawthray M, Chovan JP. Cardiac taurine levels and sarcolemmal calcium binding activity in furazolidone-induced cardiomyopathy. Comp Biochem Physiol **69C**:149–151, 1981.
13. Mela L, Hinshaw LB, Coalson JJ. Correlation of cardiac performance, ultrastructural morphology, and mitochondrial function in endotoxemia in the dog. Circ Shock **1**(4):265–272, 1974.
14. Pombo JF, Troy BL, Russell RO Jr. Left ventricular volumes and fractions by echocardiography. Circulation **43**:480–490, 1971.
15. Lust RM, Cooper MW, Lutherer LO. Applicability of m-mode echocardiography in the measurement of cardiac output in experimental animals. Fed Proc **40**(3):524, 1981.
16. Soulsby ME, Bennett CL, Hess ML. Canine arterial calcium transport during endotoxin shock. Circ Shock **7**:139–148, 1980.
17. Krause SM, Kleinman W, Hess ML. Cardiogenic endotoxin shock: coronary flow and contractile protein dysfunction as determinants of depressed cardiac contractility. Adv Shock Res **3**:105, 1980.
18. Walker RI, French JE, Walden DA, MacVittie TJ, Parker GA, Sobocinski PZ, Appelbaum FR. Protection of dogs from lethal consequences of endotoxemia with plasma or leukocyte transfusions. Adv Shock Res **4**:89–101, 1980.
19. Huxtable R, Bressler R. Effect of taurine on a muscle intracellular membrane. Biochim Biophys Acta **323**:573–583, 1973.
20. Iwata H, Fujimoto K. Potentiation by taurine of the inotropic effect of ouabain and the content of intracellular Ca^{++} and taurine in the heart. Experientia **32**:1559–1561, 1976.
21. Dolora P, Ledda F, Mugelli A, Mantelli L, Zilletti L, Franconi F, Giotti A. Effect of taurine on calcium inotropism, and electrical activity of the heart. In: Barbeau A, Huxtable RJ, eds. Taurine and Neurological Disorders. New York, Raven Press, p151, 1978.
22. Welty JD, Welty MC. Effects of taurine on subcellular calcium dynamics in the normal and cardiomyopathy hamster heart. In: Schaffer SW, Baskin SI, Kocsis JJ, eds. The effects of Taurine on Excitable Tissues. New York, Spectrum, p295, 1981.
23. Priano LL, Wilson RD, Traber DL. Cardiorespiratory alterations in unanesthetized dogs due to gram-negative bacterial endotoxin. Amer J Physiol **220**:705–711, 1971.
24. Cranefield PF. The force of contraction of extrasystoles and the potentiation of the postextrasystolic contraction. Bull NY Acad Med **41**:419–427, 1965.
25. Lust RM, Janssen HF, Lutherer LO, Cooper MW. Early effects of endotoxemia on myocardial contractile function *in vivo*. Circ Shock **8**:194, 1981.

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