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Protection of Functional and Vascular Integrity of the Spleen in Traumatic Shock by Denervation.* (29584)

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Coeliac ganglionectomy prevents the damage to the splanchnic tissues that leads to death in traumatic shock(1). Since the damaged innervated tissues in such a preparation are less sensitive than the denervated tissues to circulating catecholamines, the latter cannot be responsible for the damage. Moreover, whereas the innervated tissues show substantial depletion of their stores of norepinephrine(2,3,4,5), the denervated tissues show no depletion. Hence protection of the latter and survival of the animal may be the result of preventing the release of norepinephrine from its stores in the splanchnic tissues.

This report presents data from studies of the spleen which explore this hypothesis and support its validity.

Method. Under sterile technic and nembutal anesthesia the pedicle of half the spleen in 12 dogs was completely divided transversely, sparing only the large vessels, which were carefully stripped of their nerve supply. Shock was produced, as described below, immediately thereafter in 5 of these dogs, but not until 3 weeks later in the other 7. In 6 additional dogs the pedicle to half the spleen was denervated with xylocaine or nupercain-in-oil injected immediately before inducing shock.

In every experiment the spleen was exteriorized before shock was induced. The flank incision made for this purpose was partly closed around the splenic pedicle. The spleen was covered with a plastic sheet, and kept continuously moistened with saline at 37°C. Care was taken not to induce the shock state until the anesthesia employed to perform the laparotomy had largely worn off.

Hemorrhagic shock was induced by the elevated reservoir technic. The dogs were bled to a mean systolic pressure of 35 mm Hg, and kept at this pressure until they took back 40% of their maximal bleed-out volume, following which normovolemia was restored by transfusion of all the shed blood.

Endotoxic shock was produced by a minimal lethal dose of *E. coli* 0111 B4 endotoxin (1 mg/kg body weight) given intravenously.

Linear contraction of the spleen was measured in millimeters in response to shock, and to intravenous epinephrine or norepinephrine injected before and at various times during shock.

All animals were observed until impending death, at which time they were killed by an overdose of nembutal. Immediately thereafter samples were taken from the 2 halves of the spleen for histological study, for assay

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TABLE I. Effect of Denervation on Contractile Response of Spleen.*

Time after onset of bleeding	Bleeding vol	Non-denervated	Denervated
A. In hemorrhagic shock (blood pressure 35 mm Hg)			
1-2 min	500	++	—
5 min	680	++	—
1 hr	980	++	—
2 hr	900	+	—
3 hr	800	+	—
5 hr	600	+	—
6½ hr (1½ hr after transfusion)	0	—†	—
B. In endotoxic shock			
Time after inj of one MLD endotoxin	Bleeding vol	Non-denervated	Denervated
5 min	0	++	—
1 hr	0	++	(+)‡
2 hr	0	++	—
3 hr	0	+	—
4 hr	0	(+)	—
5 hr	0	(+)	—
5½ hr	0	—	—

* Contractile response is graded from — to +++. +++ is equivalent to maximal response to 100 μg of norepinephrine intravenously.
† Spleen swollen and congested.
‡ (+) = contraction was slight to absent.

of catecholamine content(6,7), and for assay of endotoxin-detoxifying potency(8,14).

Results. The contractile response in hemorrhagic shock was studied in 8 dogs, 4 of which had been denervated 3 weeks previously, and 4 just before shock (Table I).

In all 8 dogs the non-denervated half of the spleen contracted strongly within 60 seconds after bleeding was started. It remained contracted throughout the hypotensive period. When the shed blood was returned to the dog after some 3-5 hours in shock, this half of the spleen regained its normal size. Later, when the dog relapsed into shock, this half swelled and exceeded its original size. *The denervated half of these spleens did not change in size or appearance at any time.*

The contractile response to intravenous catecholamine was studied before and during shock (Table II).

A. Observations before shock. That half of the spleen which had been denervated several weeks previously contracted in response to some 8% of the amount of intravenous epinephrine or norepinephrine (100 micrograms) required to induce contraction of the non-denervated half. When half the spleen was blocked by a local anesthetic, both halves contracted equally in time and intensity in response to 100 μg of intravenous epinephrine or norepinephrine. When half the spleen was denervated surgically just prior to this injection, the non-denervated half contracted, but the denervated half did not contract. After 90 minutes the denervated half responded like the non-denervated half. We have no explanation for this transient refractory behavior.

B. Observations during shock. The denervated half of the spleen continued to respond to intravenous norepinephrine even in the late stage of hemorrhagic and endotoxic shock, just as it did before shock. The non-denervated half responded to intravenous norepinephrine early in shock as it did before shock, *but it did not respond at all or very slightly in the late stage of hemorrhagic and endotoxic shock.*

The denervated half of the spleen showed no significant histopathological changes. The non-denervated half showed varying degrees of necrosis and hemorrhage. In several instances structural disorganization was widespread.

TABLE II. Effect of Denervation on Contractile Response of Spleen to Intravenous Epinephrine or Norepinephrine.*

Dose in μg	Non-denervated	Denervated	Time in sec between injections & contraction
Before shock			
5	—	+	35
10	—	+	19
20	—	++	13
40	—	+++	12
60	+	+++	10
100	+++	+++	9
1 Hr after transfusion for shock of 5 hr duration (blood pressure = 100 mm Hg)			
100	—	+++	
4 Hr after transfusion for shock of 5 hr duration (blood pressure = 40 mm Hg)			
100	—	++	

* Half the spleen was denervated 3 wk prior to experiment.

No contraction is indicated by — and maximal contraction by +++.

TABLE III. Effect of Shock on Norepinephrine Content of Spleen.

Type of shock	No. of animals	Non-denervated		Denervated	
		Before shock‡	Late in shock	Before shock‡	Late in shock
Hemorrhagic*	3	1.64	.58	1.52	1.47
" †	2	1.42	.70	0	0
Endotoxic*	2	1.51	.48	1.37	1.28

Figures on norepinephrine content represent mean values in each group and are expressed as $\mu g/g$ tissue (wet wt).

* In these 2 groups denervation was done immediately prior to experiment.

† In this group denervation was done 3 wk prior to experiment.

‡ By use of special soft clamps, samples of spleen were taken without loss of blood.

The catecholamine content of the non-denervated half of the spleen in terminal shock was half or less than half the original value (Table III). In the chronically denervated half the content was zero. In the acutely denervated or blocked spleen, the content was the same as before shock.

The endotoxin-detoxifying activity present in an aliquot of a saline extract of a homogenate of the denervated half of the spleen late in shock was in every instance as potent (0.1 ml detoxifies one gamma of *E. coli* 0111 B4 by chick embryo test) as that from normal spleen. The same aliquot from the non-denervated half of the spleen of the same dog was in every instance inactive.

Discussion. In the literature on the catecholamines in traumatic shock the focus of interest has been on the role of the circulating titer(9,10,11), especially during the early stages of hemorrhagic hypotension, or shortly after injection of endotoxin. The data herein reported again demonstrate that the circulating catecholamines do not influence the course of the shock state. This is clear from the observation that the denervated spleen did not contract during shock. Since the denervated spleen was at least 10 times more sensitive to intravenous epinephrine or norepinephrine than the innervated spleen, the circulating titer clearly was not high enough to cause contraction of the innervated spleen. Hence the vigorous contraction of the latter, and the injury to its functional and vascular integrity must have been caused by norepinephrine released from its stores in the tissues. The denervated spleen does not contract, and escapes functional and vascular injury because norepinephrine is not released in denervated tissues(12).

Enlargement of the innervated half of the spleen beyond its normal size late in endotoxic shock and during the post-transfusion stage of decline in hemorrhagic shock is a manifestation of loss of tone in vascular muscle, concurrent with the loss of reactivity to norepinephrine.† The loss of tone accounts for impounding of blood in the splanchnic area late in shock and, in consequence, for the progressive decline in venous return to the heart, and the fall in cardiac output to a level incompatible with survival. Splanchnic denervation not only prevents injury to the tissues in this region, but also preserves vascular reactivity. The better hemodynamics of the systemic circulation in the denervated animal(1) indicate that without adequate flow in the splanchnic area the general circulation cannot be sustained.

It has been said that the norepinephrine content of tissue does not fall as a result of prolonged overactivity of the adrenergic nerves, because synthesis keeps pace with consumption(3,10). From our data(12) and those of others(4,13) the severe depletion of the tissue stores of norepinephrine late in shock indicates that the rate of synthesis is less than the rate of release.

It has been said that a deficiency of catecholamines accounts for the loss of vascular reactivity late in endotoxic shock(11). This view is untenable because the vascular tissue does not respond to large intravenous doses of catecholamines, whereas chronically de-

† In the dog this enlargement will not occur if there is a substantial continuing blood loss into the intestine.

nervated vascular tissues, which are devoid of norepinephrine, retain their tone and are highly reactive to intravenous catecholamines.

Preservation of the endotoxin-detoxifying power in the denervated half of the spleen, and its loss in the innervated half, indicate that the injury to function is the direct result of poor blood flow to the tissues owing to excessive activity of the locally released norepinephrine.

Summary. In dogs with part of the spleen denervated, the morphologic and functional injury that shock inflicts is restricted to the non-denervated part of the spleen. The denervated part escapes injury because denervation prevents release of norepinephrine from its stores. It is the norepinephrine released in the innervated tissue that accounts for the injury to this tissue, including its vascular muscle, and hence for the progressive failure of the circulation of the non-denervated animal. The circulating catecholamines do not play a significant role in production of this injury or in the vascular collapse and death.

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Infection of Suckling Cottontail Rabbits with Shope's Fibroma Virus.* (29585)

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Myxoma virus and the virus of Shope's fibroma are closely related(1). Both viruses produce localized, self-limiting tumors in their adult natural hosts(2,3), wild *Sylvilagus* rabbits. In domesticated and wild European rabbits *(Oryctolagus cuniculus)* each virus produces a very different disease. Myxoma virus produces a generalized, rapidly fatal disease(4). Low passage fibroma virus produces only localized proliferation in domestic rabbits(5). In neonatal domestic rabbits, however, fibroma virus can cause fatal disease of a rapidly acute or slower neoplastic type(6,7,8). The response of the neonatal natural host to fibroma virus has not been reported. In this paper are described some effects of fibroma virus on suckling eastern cottontail rabbits *(Sylvilagus floridanus)* and this response is compared to that reported for the young European rabbit.

Materials and methods. The virus used in this study was isolated from grossly typical

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