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Study of 5-Hydroxytryptamine (Serotonin) in Pericardial Fluid in Different Causes of Death (II.)

Experimental Study of 5-HT Levels in Two Types of Shocks (Hemorrhagic and Septic) in Dogs

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Summary. In the present work we studied the levels of 5-HT in the pericardial fluid of 160 cadavers according to cause of death; such as myocardial infarction, violent asphyxia, pulmonary embolism, infections, bronchopulmonary diseases, traumatic and hemorrhagic diseases in the CNS, and multiple traumatism. We did not find significant differences in the various causes of death.

A complementary study of 20 dead dogs having suffered induced shock, either hemorrhagic or septic, was made. In both series we studied the serum levels of 5-HT and the following parameters: systolic and diastolic arterial pressures, central venous pressure, hematocrit value, pH value, total proteins, albumin, PO_2, and PCO_2.

We found the significant correlation ($r = 0.828$, $P < 0.01$) only between 5-HT serum levels and the systolic arterial pressure in the hemorrhagic shock.

Key words: Serotonin, pericardial fluid – Hemorrhagic shock and septic shock, 5-hydroxytryptamine

Zusammenfassung. In der Pericardflüssigkeit von 160 Leichen mit unterschiedlichen Todesursachen (Herzinfarkt, Ersticken, Lungenembolie, Infektionen, Lungenentzündung, traumatisch-hämorrhagische Erkrankungen des ZNS, Polytrauma etc.) wurde die Konzentration von 5-Hydroxytryptamin bestimmt. Bezüglich der Todesursachen wurden keine signifikanten Konzentrationsunterschiede gefunden.

In einer experimentellen Untersuchung an 20 Hunden, die mittels hämorrhagischem oder septischem Schock getötet wurden, wurden neben 5-Hydroxytryptamin im Serum noch die folgenden Parameter gemessen: systolischer und diastolischer Blutdruck, zentraler Venendruck, Hämatokrit, pH, Gesamt-Serum-Protein, Albumin, PO_2 und PCO_2. Hierbei konnte nur eine signifikante

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Korrelation ($r=0.828$, $P<0.01$) zwischen der 5-Hydroxytryptamin-Serum-Konzentration und dem systolisch-arteriellen Blutdruck beim hämorrhagischen Schock festgestellt werden.

Schlüsselwörter: Schock – Pericardflüssigkeit, 5-Hydroxytryptamin – 5-Hydroxytryptamin, in Pericardflüssigkeit – Serotonin, in Pericardflüssigkeit

Introduction

In the forensic practice the quantification of serotonin (5-hydroxytryptamine) is used for the differential diagnostics between vital and postmortem injuries; more recently it has been used in the diagnostics of cerebral infarction and traumatism by its concentration in the brain tissue (Jellinger et al. 1979; Mohanty et al. 1978).

In a previous paper (Luna et al. 1981) we found significant differences ($P<0.001$) in the 5-hydroxytryptamine and 5-HIA acid levels measured in the pericardial fluid in relation to the length of agonal suffering.

In the present work we have studied the 5-HT pericardial fluid levels in relation to various causes of death. Moreover, we have studied the 5-HT serum levels in hemorrhagic and septic shocks induced experimentally in dogs, and the 5-HT relation to the following parameters: systolic arterial pressure, diastolic arterial pressure, central venous pressure, hematocrit value, arterial blood pH, arterial blood PO_2, arterial blood PCO_2, serum total protein and albumin.

Material and Methods

Postmortem Pericardial Fluid Series

We studied 160 cadavers at the Anatomic Forensic Institute of Granada. We obtained pericardial fluid during autopsy, sampling through a pericardial sac opening with a sterilized syringe. The samples were centrifuged at 2,000 *g* for 10 min and frozen at −20° C.

Serotonin was estimated by the method of Udenfriend (1962) using a Perkin-Elmer spectrofluorimeter, model MPF 45-A, set at 295 nm and 550 nm.

The samples were classified into several groups according to the causes of death, such as violent asphyxias, myocardial infarction, pulmonary embolism, pneumonia and other lung infections, multiple injuries, traumatic and hemorrhagic diseases of the CNS, and others.

The samples were grouped within each category, in relation to the length of agony as follows:

Group I. Less than 10 min (almost instantaneous death due to, e.g., massive pulmonary embolism, multiple trauma, and violent asphyxia).

Group II. Between 10 min and 6 h corresponding to acute processes with intense suffering and medium lengths.

Group III. Agony of long duration (more than 6).

Agony is defined as the period of time between the point where the irreversible decompensation of the vital processes begins and the moment of death.

Experimental Series

Hemorrhagic and septic shocks were induced in two groups of 10 dogs each. The animals weighed from 12 to 15 kg.

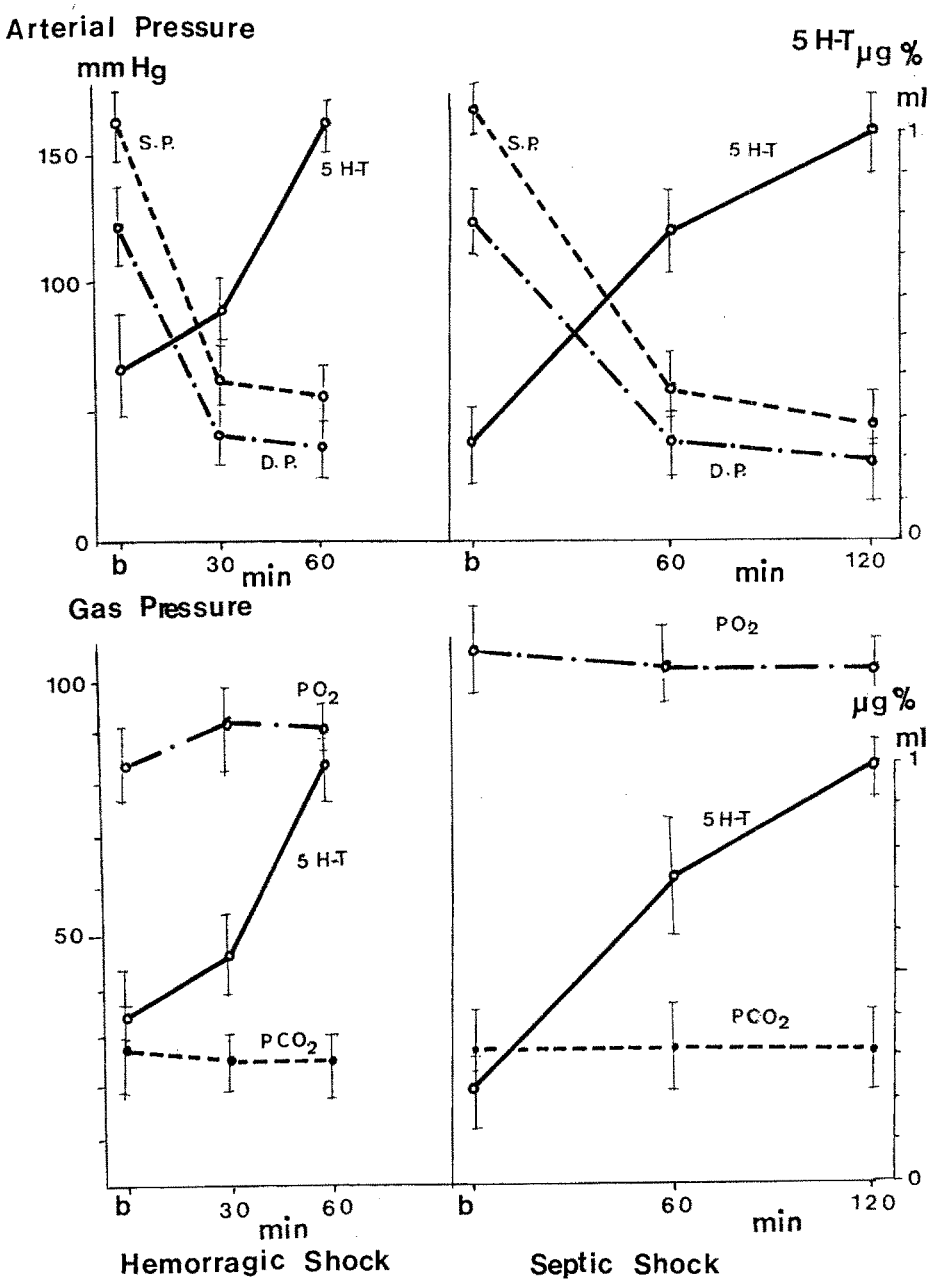

Fig. 1. 5-HT serum levels, arterial pressure, PO_2 and PCO_2 in hemorrhagic and septic shock

For the hemorrhagic shock, the dogs were treated in the following way: one catheter was placed within the superior cava to measure the central venous pressure, another in the aorta to measure the arterial pressure, and a third in the inferior cava for taking blood samples and for bleeding to produce haemorrhagic shock.

A vacuum pump was used for bleeding adjusting its speed to the arterial pressure and keeping an average arterial pressure of 4.5 ± 1. We took a basal sample and then two more samples after 30 and 60 min.

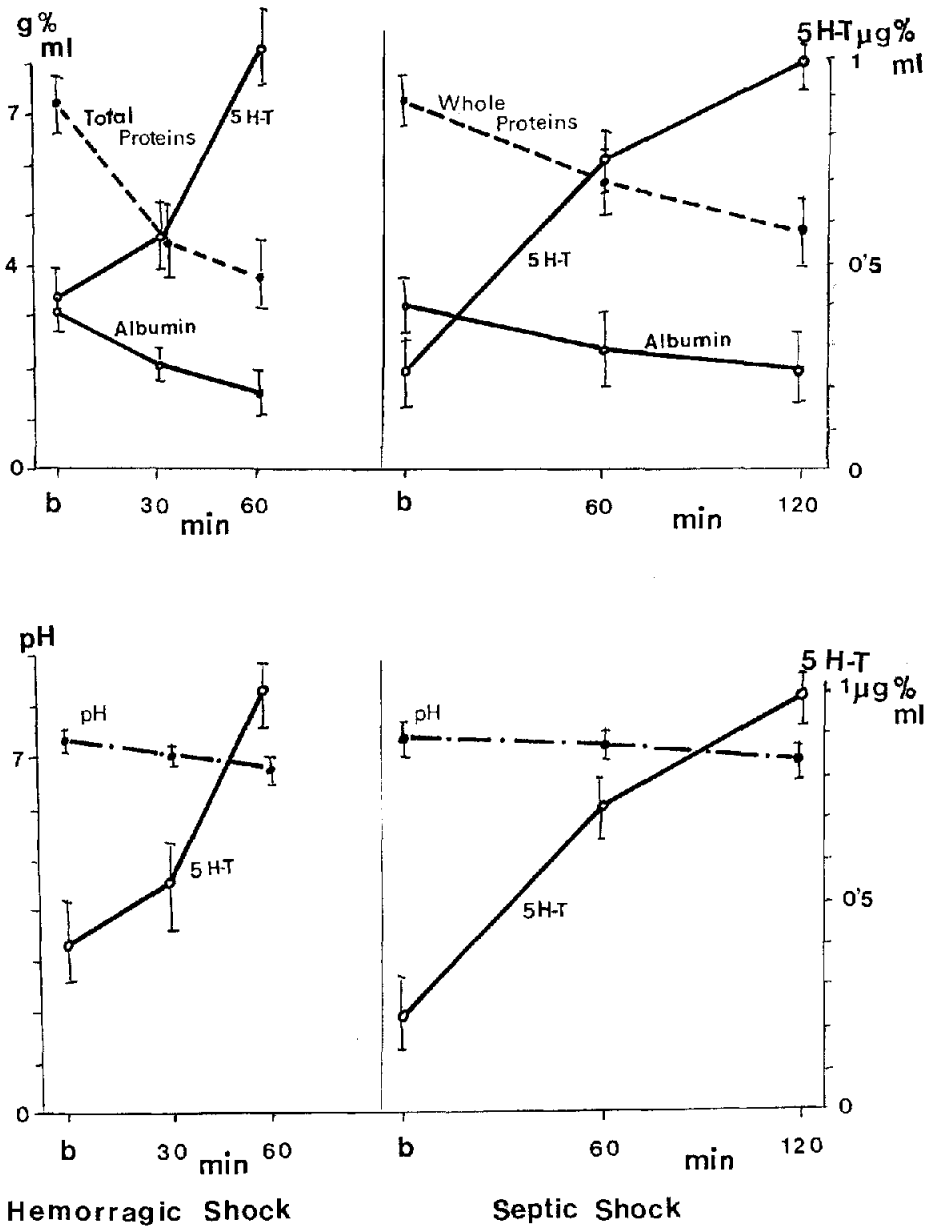

Fig. 2. 5-HT serum levels, total protein, albumin, and pH values in hemorrhagic and septic shock

The pH, the PCO_2, and the PO_2 were measured in arterial blood (using a gasometer AVL 937 for the last two). For measuring total protein, albumin, and 5-HT serum levels, venous blood was used. The hematocrit value was measured in venous blood (Figs. 1–3).

The CVP and the arterial pressure was monitored by an electromagnetic manometer (STATHAM) connected to above the mentioned catheters.

The dogs with septic shock which induced were prepared similarly. Shock was provoked by i.v. administration of purified *Escherichia coli* endotoxin (DISCO) in a dose of 1 mg/kg in isotonic solution. We measured the levels of the different parameters at zero time and after 60 and 120 min.

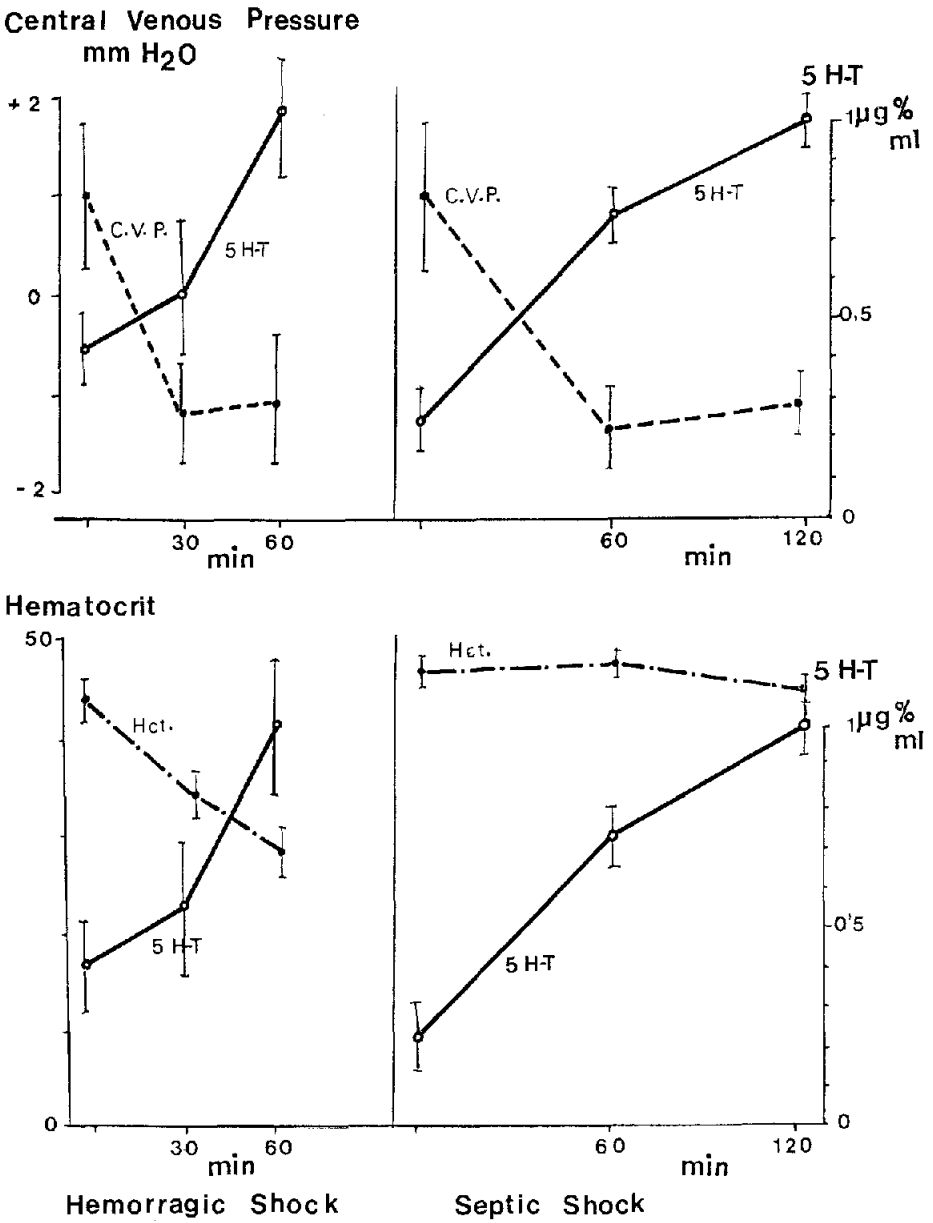

Fig. 3. 5-HT serum levels, hematocrit values, and the central venous pressure in hemorrhagic and septic shock

Results

The serotonin (5-HT) levels in pericardial fluid expressed in µg/ml in the various death cases (Table 1).

In the following tables the 5-HT levels are classified according to the different length of agony in the different death cases (Tables 2, 3).

Table 1

	No.	Mean	SD	SE
Myocardial infarction	25	0.60	0.299	0.059
Violent asphyxias	19	0.58	0.428	0.098
Pneumonic and others lung infections	24	0.52	0.409	0.083
Pulmonary embolism	20	0.49	0.351	0.078
Traumatic and hemorrhagic diseases in the CNS	18	0.37	0.148	0.034
Multiple injuries	33	0.70	0.483	0.084
Others	21	0.64	0.866	0.188

Table 2

			No.	Mean	SD	SE
Myocardial infarction	→	Group I	3	0.49	0.085	0.049
		Group II	19	0.66	0.310	0.071
		Group III	3	0.30	0.020	0.011
Violent asphyxias	→	Group I	19	0.58	0.428	0.098
Pulmonary embolism	→	Group I	3	0.348	0.164	0.094
		Group II	11	0.75	0.293	0.088
		Group III	6	0.17	0.043	0.017
Pulmonary and others lung infections	→	Group II	15	0.68	0.430	0.111
		Group III	9	0.25	0.159	0.053
Traumatic and hemorrhagic diseases in the CNS	→	Group I	15	0.328	0.120	0.031
		Group II	3	0.58	0.097	0.056
Multiple injuries	→	Group I	15	0.55	0.484	0.124
		Group II	14	0.94	0.436	0.116
		Group III	4	0.40	0.078	0.039
Others	→	Group I	3	0.47	0.240	0.138
		Group III	7	1.34	1.315	0.497
		Group III	11	0.24	0.037	0.011

Discussion

The rise of the 5-HT blood levels has been shown previously under various circumstances by several workers (Laves and Berg 1966) after carcinoid syndrome, traumatic shock, surgical intervention and trauma, CO intoxication, IMAO intoxications, burns, electrocution, anaphylactic shock.

Hoshino et al. (1979) found that after physical exercise (2 h of basket ball playing) serum serotonin levels showed a slight increase, which was not, however,

Table 3

A. Septic shock

	No.	SAP		DAP		CVP		Hematocrit		pH	
		Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
0 time	10	171.7	18.726	127.2	16.380	1	2.309	46.6	3.717	7.34	0.042
1 h	10	60.7	11.560	38.7	6.896	−1.4	1.429	47.1	5.877	7.18	0.065
2 h	10	46.7	6.030	31.6	5.891	−1.1	1.523	44.7	2.983	7.00	0.067
	No.	PO_2		PCO_2		Total Pr[a]		Albumin[a]		5-HT[b]	
		Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
0 time	10	105.3	12.710	26.6	3.272	7.27	0.601	3.26	0.294	0.39	0.095
1 h	10	102.0	10.593	7.0	2.581	5.53	1.106	2.45	0.245	0.75	0.307
2 h	10	102.8	7.390	26.0	1.885	4.73	0.926	1.91	0.448	1.04	0.318

B. Hemorrhagic shock

	No.	SAP		DAP		CVP		Hematocrit		pH	
		Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
0 time	10	168.3	18.577	124.4	16.015	0.9	0.730	44.4	5.211	7.34	0.069
30 min	10	63.0	8.819	41.0	7.086	−0.95	1.570	32.6	7.876	7.10	0.230
1 h	10	53.2	15.288	33.5	9.582	−0.90	1.370	28.1	6.590	6.83	0.278
	No.	PO_2		PCO_2		Total Pr[a]		Albumin[a]		5-HT[b]	
		Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
0 time	10	94.2	13.94	27.5	3.807	7.13	0.583	3.18	0.225	0.41	0.045
30 min	10	92.9	17.72	26.1	6.244	4.51	0.860	2.03	0.419	0.56	0.059
1 h	10	92.4	11.93	23.6	4.498	3.73	0.763	1.52	0.449	1.15	0.270

[a] The serum levels of total proteins and albumin are expressed in g/ml
[b] The serum levels of 5-HT are expressed in µg/ml

SAP = systolic arterial pressure; DAP = diastolic arterial pressure; CVP = central venous pressure; Total Pr = total proteins

statistically significant. Parbtani et al. (1980) showed that the intraplatelet serotonin levels were depressed and the plasma serotonin levels were raised in lupus nephritis. Kulinski and Nefedova (1976) found that hypoxia mobilizes 5-HT (rise of the serotonin blood levels) and stimulates its biosynthesis. Morris and Moon (1974) showed that there is an increased serotonin synthesis in the endotoxin shock in mice. Katsman (1973) found in 80 patients with myocardial infarction that the blood 5-HT level decreases within the firsts 24 h, and a rise is seen on days 8 and 10 and again days 18 an 23 of illness. Mrsulja et al. (1976) and Harrison et al. (1979) found a reduction of cerebral serotonin in the brain during experimental ischemia (the ischemia being produced by unilateral carotid ligation). Jellinger et al. (1979) found a total depletion of dopamine and 5-HT in the necrotic area in various patients who had died after acute and non-acute cerebral infarction, while the perifocal edema zone showed considerable accumulation of 5-HT and 5-HIAA. Mohanty et al. (1978) found similar results in patients with gross evidence of brain contusion and edema following trauma. Hirvonen et al. (1978) found a rise of 5-HT urinary excretion in severe cold exposure of normal and cold-acclimated guinea pigs (0.08–0.21 μg/h in normal guinea pigs and 0.08–0.17 μg/h in cold-acclimated animals).

The statistical treatment of 5-HT pericardial fluid levels in different causes of death confirms the non-specificity of the 5-HT liberation. We think that this is not valid for postmortem diagnosis among the different causes of death due to a nonspecific type of response of the parameter.

On the contrary, it is an excellent parameter to estimate the length of the agonic process, as we have already established in a previous work. We found that the simultaneous estimation of 5-HT and its metabolite 5-HIA acid in the pericardial fluid, is very appropriate for that purpose.

The myocardial interstitial fluid and the myocardial plasma are responsible for the 5-HT pericardial fluid levels, due to both an eventual overflow of the first and a passive utrafiltration of the latter.

According to the general characteristics of the cardiac function and to the structure of heart one or another mechanism seems to be more relevant. The ultrafiltration will predominate during the diastole, the overflow mechanism more during cardiac contraction, and in some cases (inflammatory processes) the visceral serose may play an important role. The serum rises of 5-HT post mortem by its liberation from blood cells through an autolytic process (Luna et al. 1981) may contaminate the pericardial fluid. Since an overflow mechanism does not exist after death, only the passive diffusion deserves some attention as an influencing mechanism on the rising of 5-HT post mortem pericardial fluid levels. However the 5-HT gradient difference through the pericardial serose during the post mortem period must be so high that the phenomenon is only noticeable near the large vessels or in the cardiac cavities. Nevertheless, we have not obtained experimental data.

To explain the eventual mechanism that could induce the 5-HT serum liberation, we have outlined an experimental study in which we compare the 5-HT serum levels with several other parameters in two kinds of experimentally induced shock in dogs.

Table 4

	F. Exp.			
	Septic shock		Hemorrhagic shock	
SAP	278.94	$P<0.001$	142.61	$P<0.001$
DAP	245.20	$P<0.001$	131.83	$P<0.001$
CVP	10.32	$P<0.01$	9.39	$P<0.01$
Total Pr	42.45	$P<0.001$	203.33	$P<0.001$
Albumin	47.35	$P<0.001$	80.70	$P<0.001$
pH	142.83	$P<0.001$	16.50	$P<0.001$
5-HT	20.23	$P<0.001$	65.06	$P<0.001$
Hematocrit	—	—	20.51	$P<0.001$

Table 5

	F. Exp.	
	Septic shock	Hemorrhagic shock
PO_2	0.55	2.54
PCO_2	0.29	1.84
Hematocrit	0.66	—

In these experiments we found significant differences in the following parameters in relation to different time (Table 4). We did not observe significant differences in PO_2, PCO_2, and Hematocrit (Table 5).

The study of correlation between the different parameters and the 5-HT serum levels showed a significant correlation ($r=0.828$, $P<0.01$) only concerning the systolic arterial pressure in the hemorrhagic shock.

Ramadan et al. (1973), studying the 5-HT blood levels in normal and pre-eclamptic patients, showed a highly significant increase (nearly twice the normal values) in mild and severe pre-eclamptic patients as compared to normal pregnant women. Jelen et al. (1979) gave no evidence that 5-HT is implied in hypertension during pregnancy. The direct "pressor" effect of 5-HT is mainly due to the increase of total peripheral resistance and cardiac output (Goodman and Gilman 1980).

We suggest that the liberation of 5-HT is a nonspecific type of physiologic response due to numerous circumstances as we cannot explain its specific function with precision.

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