CHANGES IN THE BLOOD NH_3-N LEVEL AFTER LIGATION OF THE HEPATIC ARTERY, WITH CHANGES IN THE HEMATOCRIT RATIO

by

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(Received for publication Sept. 26, 1959)

INTRODUCTION

Herrick and others[10),24)] pointed out that in annular nodular liver cirrhosis the causes of portal hypertension followed by accumulation of ascitic fluid were (1) that hepatic vein branches were compressed by regenerating liver nodules and (2) that the elevated hepatic arterial pressure was transferred to the portal pressure through free presinusoidal communications between hepatic arteries and portal veins. They also made it clear that in cirrhotic liver the rate of blood flow in the artery had a relative increase as compared with the normal liver.[5)] On the other hand, Markowitz[15)] et al. found that postoperative penicillin could lower the mortality of dogs deprived of hepatic arterial blood supply by preventing massive liver necrosis. In 1951, Rienhoff,[28)] Berman and others[1)] reported that the ligation of the hepatic artery in a case of cirrhosis with ascites caused a decrease in ascites and improvements of other clinical findings.

In our clinic too, in order to investigate clinical application of the ligation of the hepatic artery, a series of experimental studies have been performed on changes following the ligation, especially on liver necrosis, in both normal dogs and those ascitic ones which, hemodynamically, are similar to those with cirrhosis.[26)] In parallel with these studies, I tried to investigate the ammonia metabolism after the ligation of the hepatic artery.

Some investigators[13),16),18)] have reported that coma, which occurs in liver cirrhosis or in Eck fistula, is often accompanied by an elevation of blood ammonia level, and that administration of nitrogenous substance to patients with hepatic disorders may cause neurological and mental disturbances similar to hepatic coma.[12),14)] The blood ammonia level has come to play an important role in the development of neurological and mental disorders in hepatic diseases.

Experience tells us about the difficulty of drawing blood from peripheral vessels after the ligation of the hepatic artery. Tanturi and others[29)] also reported that the ligation of the hepatic artery caused hemoconcentration, the degree of which was of some help in predicting the prognosis of the dog. According to Moon and others,[17)] this hemoconcentration means a decrease in the circulating blood volume. Thus in the present study, the determination of hematocrit ratios was performed together with that of blood ammonia levels, so as to observe hemoconcentration after the

ligation of the hepatic artery and an aspect of the systemic or hepatic circulation. (In the following page hematocrit will be referred to as Ht.)

Moon defined "shock" as a type of circulatory failure, not central but peripheral in origin, characterized by decreased blood volume, and decreased volume flow and by hemoconcentration, and thought that hemoconcentration was caused by leakage of plasma due to abnormal permeability of systemic capillaries. In recent years, however, concerning the cause of hemoconcentration in "shock", there has been a tendency to put much emphasis on local leakage of plasma in the damaged part.[9),19),31)]

EXPERIMENT ON ANIMALS

I Materials and Methods

(1) Animals. Adult mongrel dogs, weighing about 10 kg, were used. They were fed chiefly on carbonhydrate diets, but given nothing but water within 12 hours before the experiment.

(2) Where and how to take blood samples. Peripheral vein blood was drawn from the hind limb, arterial blood from the femoral artery, and portal and renal vein blood from the portal trunk and the renal vein respectively by laparotomy. As the anticoagulant, 2 to 3 mg of potassium oxalate per 1 cc of blood was used. The potassium oxalate had been made NH_3 free by recrystalizing in an alkaline solvent with 10 to 11 pH.

(3) Method of measurement of blood ammonia. A modification of Conway's microdiffusion technique[2),4)] was employed. Ammonia was indicated by the amount of nitrogen it contained. A Conway's microdiffusion unit was prepared with the central absorbing chamber containing 0.7 cc 0.0002 or 0.001 N HCl, with Tashiro's reagent as indicator. The outer chamber contained 1 cc blood, to which 1 cc saturated boiled K_2CO_3 was added. After covering it with a glass lid, the unit was rotated 20 times for 20 seconds to mix up the solutions, then put aside to incubate for 10 to 20 minutes at room temperature. Barium hydroxide ($Ba(OH)_2$), 0.0004 or 0.002 N, delivered from a horizontal pyrex, micrometer driven burette, was used to titrate excess acid. The end point was set at 5.4 to 5.6 pH where the indicator became colourless. Using this technique titrations of controls and blanks were made simultaneously. For the control, 1 cc of $(NH_4)_2SO_4$ solution, containing 2r/dl or 10 r/dl of nitrogen, was put into the outer chamber, and for the blank, 1 cc of distilled water was used. To these solutions 1 cc saturated boiled K_2CO_3 was added to get end points of the control and the blank. With figures thus obtained from titrations of bloods, controls and blanks, the nitrogen content of the blood ammonia was calculated according to proportion. The results were then corrected by Conway's correction table, refering to the room temperatures and periods of incubation. The figures thus corrected were taken as the blood ammonia levels.

(4) Measurement of Ht. ratios. As the anticoagulant, 2 to 3 mg of potassium oxalate per 1 cc blood was used. The blood to be measured was put into a Wintrobe's tube till it rose to 100 in graduation, then centrifuged at a frequency of 3,000 r. p. m. for 30 minutes.

(5) Operative technique. Intravenous nembutal anesthesia (0.3 to 0.5 cc per kg) was used. By means of the technique established in our clinic, the common hepatic artery, right gastric artery, and gastroduodenal artery were doubly ligated and divided. Investigating the arterial collateral pathways to the liver, URABE and ISHIGURO made it clear that the ligation of these three arteries interrupts most of arterial blood supply to the liver, leaving no effective arteries enough to prevent liver necrosis.

In order to produce ascitic dogs McKEE's method was employed. Under closed inhalation anesthesia thoracotomy was performed to reach for the thoracic inferior vena cava, which was slightly constricted by a cellophane band. 2 to 3 weeks later those dogs in which ascites was produced were further subjected to the ligation of the hepatic artery briefly mentioned in the above.

II Results

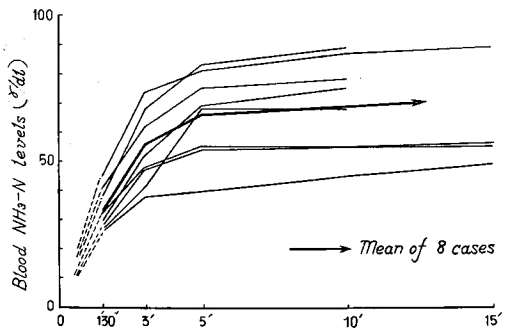
(1) Changes in the blood ammonia level after drawing of blood (Fig. 1, Table 1).

When blood is drawn and left in air, the ammonia level shows a rapid increase until 3 to 5 minutes after the drawing. Then it continued to rise progressively but very gradually. So in the following measurements performed on animals and clinical cases, care was taken to complete the procedure of adding saturated K_2CO_3 to the blood drawn and put into the outer chamber of the incubator between 5 and 10 minutes after drawing blood. The ammonia concentrations in peripheral vein blood obtained from 32 control dogs lay between 36 to 84 r/dl, averaging 51 r/dl.

(2) Physiological changes in the ammonia level in peripheral venous blood (Fig. 2, Table 2).

For investigation of diurnal variations

Fig. 1. Demonstration of changes in the blood NH_3-N levels after shedding blood.



Note: Blood was taken from peripheral vein

Table. 1a. Changes in blood NH_3-N levels after sheading blood.

Note: Blood was taken from peripheral vein.
※ indicates minutes after sheadding.

Dog No	wt. (Kg)	Sex	NH3-N levels (γ/dl) ※1'30"	3'	5'	10'	15'
1	10	♀	32	48	55	55	56
2	11	♂	26	38	40	45	49
3	7	♂	28	48	54	55	55
4	12	♀	41	62	75	78	
5	7	♂	38	68	83	89	
6	7	♂	45	74	81	87	89
7	11	♂	26	41	68	68	
8	6	♀	29	52	69	75	
	Mean		33	55	66	69	

Table. 1b. Peripheral blood NH_3-N levels (γ/dl) in 32 normal dogs.

46	79	69	61	40	50	62	47
56	43	76	53	50	70	62	46
52	44	61	45	38	66	84	58
42	38	62	45	72	67	58	64

Mean of 32 dogs. 56 ± 12

Table 2. Physiological changes in blood NH_3-N levels.

(1) Diurnal changes.

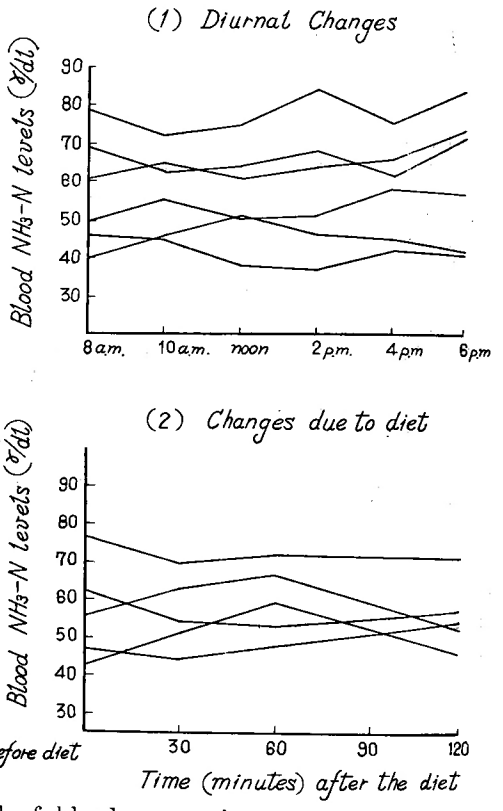
Dog No.	Wt (Kg)	Sex	8a.m.	10a.m.	Noon	2p.m.	4p.m.	6p.m.	Range of variations.
1	10	♂	46	45	38	37	42	41	37~46
2	14	♂	79	72	75	84	75	84	72~84
3	7	♂	69	63	64	68	62	72	63~72
4	11	♀	61	65	61	64	66	74	61~74
5	7	♀	40	45	51	46	45	42	40~51
6	8	♂	50	55	50	51	58	57	50~58

(2) Changes due to diet.

Dog No.	Wt (Kg)	Sex	Before the diet	※30′	60′	120′	Range of variations.
1	7	♂	62	54	53	56	53~62
2	10	♂	47	44	48	54	44~54
3	14	♀	56	63	63	52	52~66
4	8	♀	43	51	59	45	43~59
5	9	♂	76	70	72	71	70~76

Note : Blood was taken from peripheral vein.
※ indicates minutes after the diet.

Fig. 2. Physiological changes in blood NH_3-N levels.



in the level, the measurement was performed at an interval of 2 hours from 8 a. m. to 6 p. m. The range of the variations turned out to be 8 to 13 r/dl, with 10 r/dl on the average. In order to observe if eating would influence the level of blood ammonia, measurements were made before diet, and 30, 60 and 120 minutes after it. The result was that the variation was within the range of 6 to 16 r/dl, with an average of 11 r/dl. Thus physiological variations in the ammonia level in peripheral venous blood were around 10 r/dl, a little exceeding the analytical error of ± 3 r/dl.

(3) The ammonia levels in main blood vessels (Fig. 3, Table 3).

The ammonia levels in peripheral arterial and venous blood were much the same, ranging from 40 to 80 r/dl, the former being slightly higher than the latter. As for the portal vein blood, the level stood between 232 and 607 r/dl, 194 to 541 r/dl higher than the level in peripheral venous blood. The values for renal vein blood were 120 to 266 r/dl, exceeding those in peripheral vein blood by 58 to 221 r/dl.

SUMMARY OF PRELIMINARY EXPERIMENTS (1), (2) AND (3)

According to CONWAY,[2),3)] free ammonia in normal circulating blood is below the analytical level, and the ammonia concentration in the blood drawn in air rises rapidly for 3 to 5 minutes after drawing, after that the rise is very much slowed

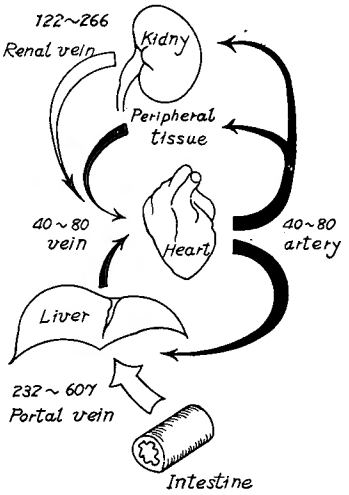
Table. 3. Blood NH_3-N levels (γ/dl) in main blood vessels.

Dog. No.	Wt. (Kg)	Sex	Peripheral artery	Peripheral vein	Difference
1	12	♂	60	53	7
2	14	♂	58	50	8
3	9	♂	76	70	6
4	7	♀	80	62	18
5	10	♂	51	46	5
6	9	♀	56	52	4
7	9	♀	53	44	9
8	11	♂	65	61	4

Dog. No.	Wt. (Kg)	Sex	Portal vein	Peripheral vein	Difference
1	7	♀	426	45	381
2	9	♀	232	38	194
3	10	♂	607	66	541
4	12	♀	411	84	327
5	10	♂	356	58	298

Dog. No.	Wt. (Kg)	Sex	Renal vein	Peripheral vein	Difference
1	10	♂	170	42	128
2	9	♀	147	38	109
3	7	♂	120	62	58
4	11	♂	266	45	221
5	14	♀	206	72	134

Fig. 3. Blood NH_3-N levels (γ/dl) in main blood vessels.



down. The cause of the ascent, as he asserts, is that adenosine in blood is decomposed by the function of adenosinedeaminase. To investigate this point many experiments[30),32),37)] have been made, in all of which the blood ammonia level as it stood 5 to 10 minutes after drawing was considered as the ammonia level of the blood samples.

KOPROWSKI and UNINSKI[21)] reported that the blood ammonia level and revelation of ammonia in blood after drawing were much the same with dogs as with man. My present experiment has come to a similar conclusion. Accordingly, the amount of N in ammonia determined between 5 and 10 minutes after drawing blood was taken as the ammonia level of the blood sample.

WHITE, MOTONAGA[23),37)] and others reported on very little physiological variations in the ammonia level of peripheral vein blood in healthy adult persons. This report has been supported by the result of my present experiment. It has been known that between the ammonia content of peripheral arterial and venous blood there is so little difference that it barely exceeds the analytical error. Some reported, however, that a rise in the ammonia level in arterial blood was followed by an appreciable arterio-venous difference in the ammonia content.[30),39)] Therefore, in this study, about 30 hours after ligation of the hepatic artery measurements were performed on arterial blood.

Compared with the values of peripheral blood ammonia, those of portal and renal venous blood ammonia come out remarkably high. This is proof that the intestines and kidney are the chief sources of blood ammonia. On the other hand, there are many reports that the ammonia content of peripheral blood is much the

same as that of hepatic vein blood.[23),30)] All these join to support the existing knowledge that the liver is the chief organ which dispose of ammonia.[11),16)]

(4) Changes in the blood ammonia levels and Ht. ratios in normal dogs after the ligation of the hepatic artery (Fig. 4, Table 4).

Ligation of the hepatic artery was performed on 13 normal dogs, all of which died within 30 hours postoperatively, revealing necrosis in the liver at autopsy. In 7 of them, the blood ammonia levels were followed up till their death. About 60 minutes after ligation, no marked changes in the level were observed. However, after 3 hours it began to rise gradually until at the 6th to 10th hour it reached a level 40 to 70 r/dl higher than the initial level. Then it went on with slight variations, and about 60 minutes before death a sudden, rapid increase took place. In 11 dogs the values of blood ammonia went up as high as 170 to 470 r/dl just before death (Fig. 11).

Serial determinations of Ht. ratios were performed on 3 dogs. The Ht. ratios began to rise immediately after the ligation, and 60 minutes later showed a 10% increase over the initial level. In Nos. 5 and 7 dogs it further increased by 4 to 5% between 3 and 10 hours following the ligation, after which it disclosed a slight tendency to decline. In No. 6 dog, however, the level did not show such a tendency, but only a slight variation upward or down.

Fig. 4 Changes in blood NH_3-N levels in normal dogs after the interruption of hepatic artery with changes in hematocrit ratios.

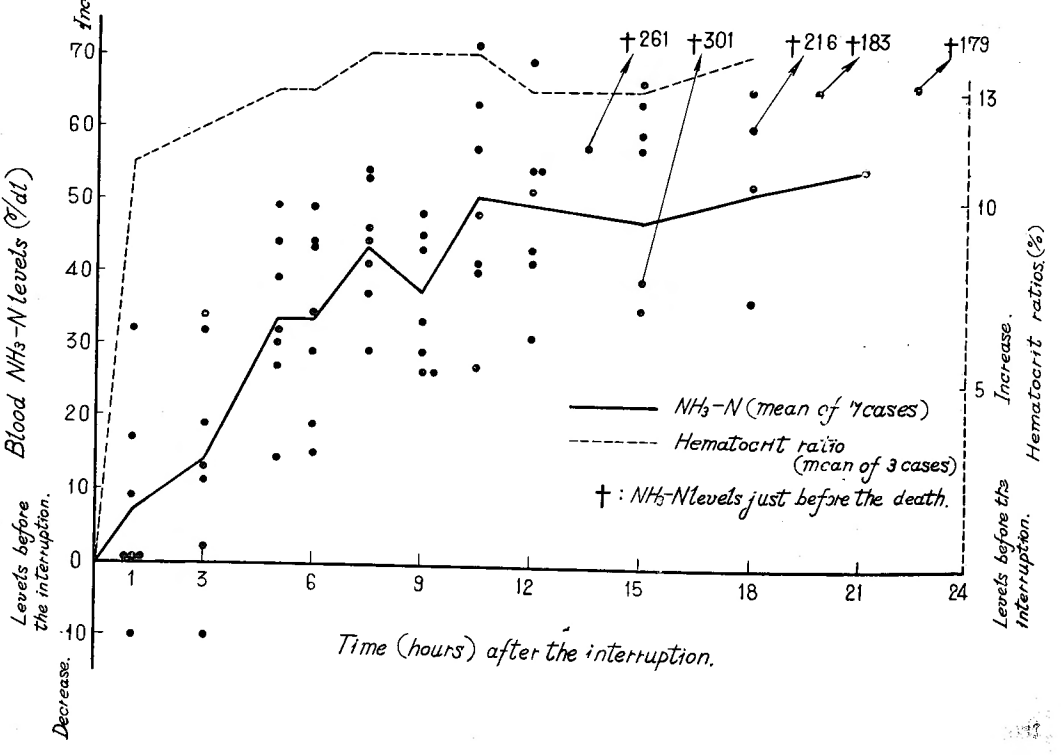


Table. 4. Changes in blood NH_3-N levels in normal dogs after the interruption of hepatic artery with changes in hematocrit ratios.

Dog. No.	wt. (Kg)	Sex	Before the interruption	NH_3-N levels (γ/dl) after interruption. ※ 1	3	5	6	7½	9	10½	12	15	18	21	22
1	9	♂	47	47	49	61	62	93	90	110	98	104	99	101	179†
2	7	♀	67	67	57	94	101	121	112	138	136	126	127	216 (1930†)	
3	7	♂	72	104	85	102	115	116	105	129	126	135 (1700†)			
4	10	♀	84	74	103	116	113	121	110	111	127	123	301 (1630†)		
5	8	♀	64	73	98	108	113	117	112	112	116	261†			
6	11	♀	38	55	70	79	59	69	67	79	79	104	103	183†	
7	7	♀	40	40	51	89	84	81	66	80	71	75	76 (1830†)		
				Mean of increase or decrease											

Dog. No.	wt. (Kg)	Sex	Before the interruption	Increase or decrease after the interruption. ※ 1	3	5	6	7½	9	10½	12	15	18	21	24
1	9	♂	47	0	2	14	15	46	43	63	51	57	52	54	132†
2	7	♀	67	0	-10	27	34	54	45	71	69	59	60	149†	
3	7	♂	72	32	13	30	43	44	33	57	54	63	†		
4	10	♀	84	-10	19	32	29	37	26	27	43	39	217†		
5	8	♀	64	9	34	44	49	53	48	48	52	197†			
6	11	♀	38	17	32	41	21	31	29	41	41	66	65	145†	
7	7	♀	40	0	11	49	44	41	26	40	31	35	36	†	
				Mean of increase or decrease	7	14	33	33	43	37	50	49	47	51	54

Dog. No.	wt. (Kg)	Sex	Before the interruption	Hematocrit ratios (%) after the interruption. ※ 1	3	5	6	7½	9	10½	12	15	18	21	24
5	8	♀	39	48	52	51	50	52	51	51	48	50			
6	11	♀	24	42	42	38	41	42	41	44	46	43	43	45	
7	7	♀	42	49	54	54	54	54	54	52	50	50	51		
				Mean of increase or decrease											

Dog. No.	wt. (Kg)	Sex	Before the interruption	Increase or decrease after the interruption ※ 1	3	5	6	7½	9	10½	12	15	18	21	24
5	8	♀	39	9	13	12	11	13	12	12	9	11			
6	11	♀	24	18	18	14	17	18	17	20	22	19	19	21	
7	7	♀	42	7	12	12	12	12	12	10	8	8	9		
				Mean of increase or decrease	11	12	13	13	14	14	14	13	13	14	

Note : ※ : Time after the interruption.
† Time of death.
Blood was taken from the femoral artery.

(5) Changes in the blood ammonia levels and Ht. ratios after constricttion of the hepatic veins (Fig. 5, Table 5).

URABE and HOSONO made it clear that in normal dogs interruption of the hepatic arterial blood flow caused the liver to be congested and to change its colour, inducing the reduction of the rate of portal blood flow. Then, what influence do congestion of the liver and reduction in the rate of hepatic blood flow exercise over the blood ammonia levels and Ht. ratios? To answer this question, 2 normal dogs were subjected to constriction of the hepatic veins by the method of TSUCHIYA in our clinic, and the values of ammonia and Ht. ratios were checked up. In 3 hours after the constriction the ammonia levels clearly ascended, showing 335 r/dl in one case and 225 r/dl in another, and the Ht. ratios showed a 14% increase in the former and a 12% increase in the latter.

(6) Changes in the blood ammonia levels and Ht. ratios in the case of fluid transfusion given after the ligation of the hepatic artery (Fig. 6, Table 6).

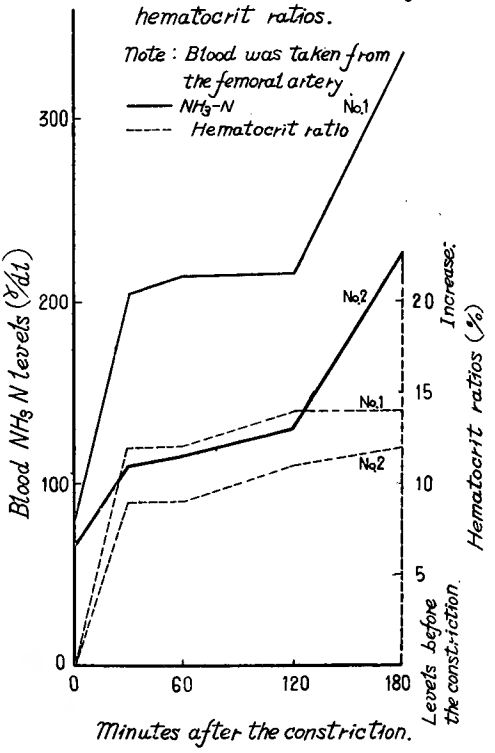
WAKIM, UEDA and others[33),34)] pointed out that fluid transfusion increased the amount of blood of not only systemic but also hepatic circulation. So in the present study, in order to observe how such transfusion would influence the blood ammonia levels and Ht. ratios, 600 cc of 5% glucose or balanced electrolyte solution were injected drip by drip into the femoral vein in 5 dogs for a period of 2 to 3 hours, beginning

Table. 5. Changes in blood NH_3-N levels after the constriction of the hepatic veins with changes in hematocrit ratios.

Dog. wt. Sex No. (Kg)		Before the constriction	Minutes after the constriction 30′	60′	120′	180′
1 8 ♀	NH_3-N (γ/dl)	83	204	214	215	335
	Increase	0	121	131	132	252
	Hematocrit-R (%)	34	46	46	48	48
	Increase	0	12	12	14	14
2 10 ♂	NH_3-N (γ/dl)	68	110	115	130	225
	Increase	0	42	47	62	157
	Hematocrit-R (%)	39	48	48	50	51
	Increase	0	9	9	11	12

Note : Blood was taken from the femoral artery.

Fig. 5. Changes in blood NH_3-N levels after the constriction of the hepatic veins with changes in hematocrit ratios.



from 10 to 17 hours after interruption of the hepatic artery when the Ht. ratios might well have increased and both the systemic and the hepatic blood flow decreased.

About 30 to 60 minutes after the commencement of the transfusion, the ammonia level in all cases declined nearly to the initial level. When the transfusion was finished, however, the level was restored almost to what it had been before the transfusion. Then, no more changes were observed in Nos. 2 and 5 dogs until their death. On the contrary, between 1 and 4 hours after the fluid transfusion had been finished, the ammonia levels in Nos. 1, 3 and 4 dogs again decreased nearly to the initial level maintaining these values until just before their death. Nos. 1, 2, 3 and 4 dogs died with blood ammonia levels of about 50 r/dl higher, and No. 5 dog with ammonia level of 150 r/dl higher, than their initial levels. In other words, the blood ammonia levels they disclosed just before the death ranged from 86 to 200 r/dl ··· much lower than normal 170 to 470 r/dl.

The Ht. ratios dropped during the transfusion in all cases. After it, however, they behaved erratically ··· some went on with much the same levels as before the ligation, some held the levels had shown during the transfusion, and others were restored to as high levels as before the transfusion.

(7) Changes in blood ammonia levels and Ht. ratios in penicillin-treated dogs

Fig. 6. a. Changes in blood NH_3-N levels in dogs receiving fluid transfusion after the interruption of the hepatic artery.
Note : Blood was taken from the femoral artery.
† : levels just before the death.

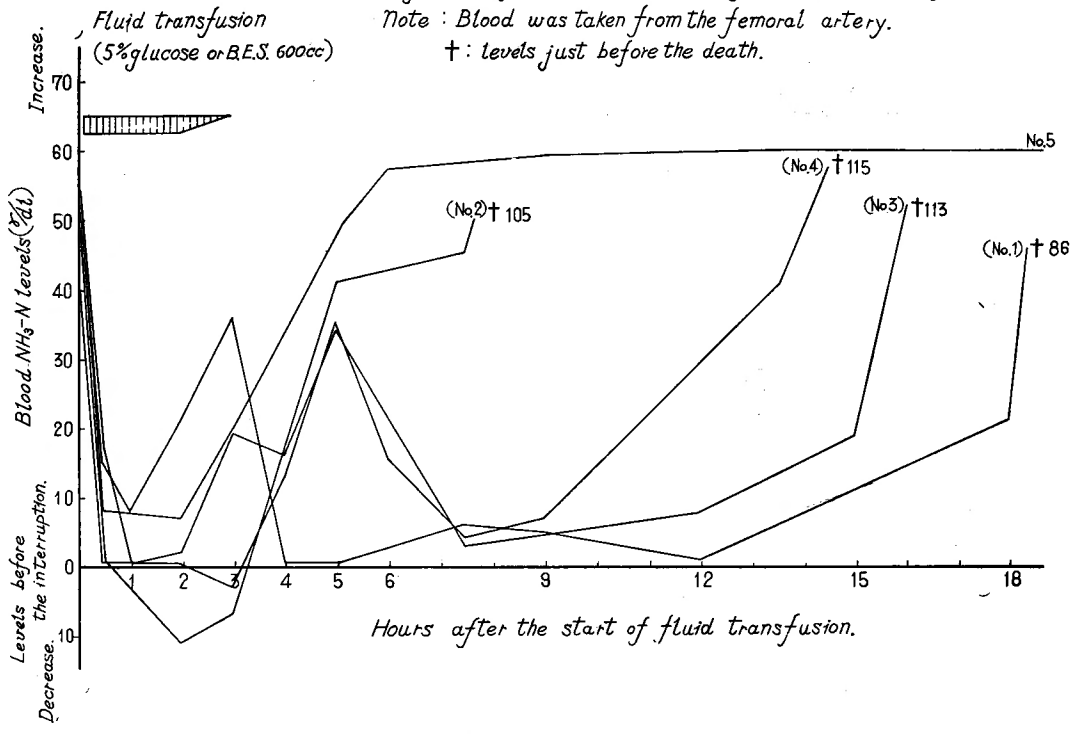


Fig. 6. b. Changes in hematocrit ratios in receiving fluid transfusion after the interruption of the hepatic artery.
Note : Blood was taken from the femoral artery.

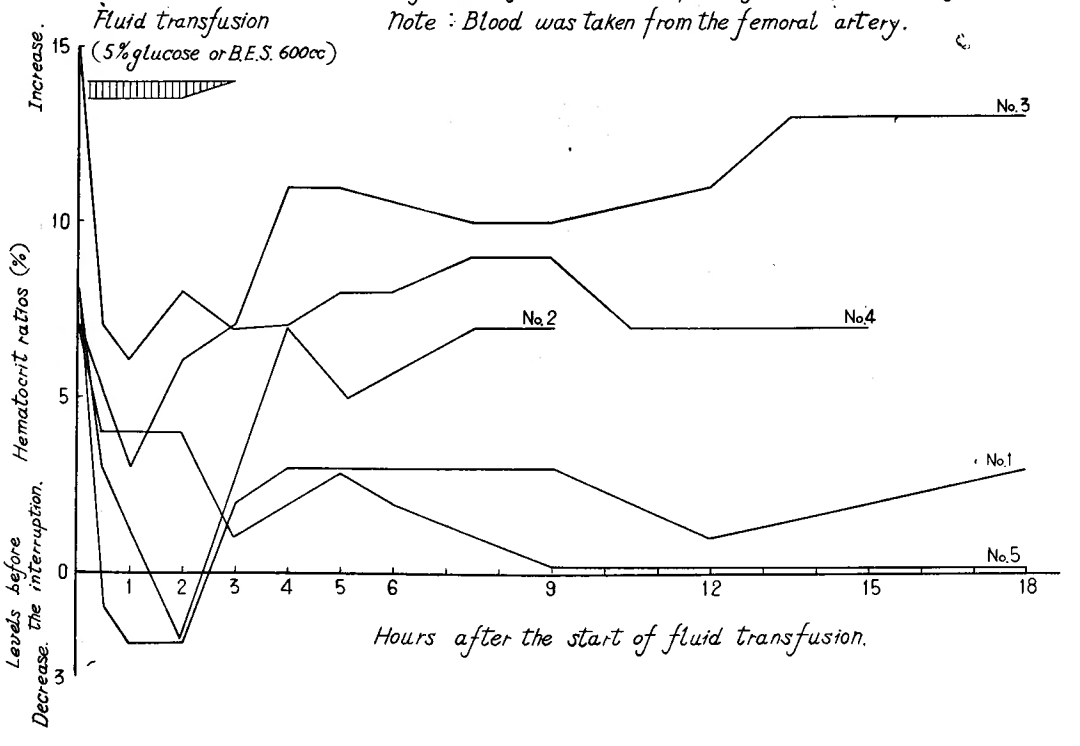


Table. 6. Changes in blood NH_3-N levels and hematocrit ratios in the case of fluid transfusion 10 to 17 hours after the interruption of the hepatic artery. Note: Blood was taken from the femoral artery

Dog. No.	Wt. (Kg)	Sex	※		Before the interruption	Before the transfusion	30′	1	2	3	4	5	6	7.30	9	10.30	12	13.30	15	18	21	24
1	11.5	♀	15.10′~17.10′ 5% glucose 600cc	NH_3-N(γ/dl)	40	91		55	48	61	76	40	40		46	45		41			61	86 † 18.20
				Increase or decrease	0	51		15	8	21	36	0	0		6	5		1			21	46
2	8.3	♀	13.45′~16.30′ 5% glucose 600cc	NH_3-N(γ/dl)	55	103		55	44	48	72	96		100	105 † 7.45							
				Increase or decrease	0	48		0	−11	−7	17	41		45	50							
3	10.5	♀	10.20′~12.50′ 5% glucose 600cc	NH_3-N(γ/dl)	61	115		78	61	63	80	77	95		64	66		69	74	90	113 † 16.00	
				Increase or decrease	0	54		17	0	2	19	16	34		3	5		8	13	19	52	
4	9	♂	17.00′~20.00′ B.E.S. 600cc	NH_3-N(γ/dl)	57	102		57	57	57	54	70	92	72	61	64	75		98	115 † 14.30		
				Increase or decrease	0	47		0	0	0	−3	13	35	15	4	7	18		41	58		
5	7.5	♂	14.25′~16.25′ 5% glucose 600cc	NH_3-N(γ/dl)	46	86		54		53	65		95	103		105			106		106	200 † 22.00
				Increase or decrease	0	40		8		7	19		49	57		59			60		60	154
1	11.5		15.10′~17.10′	Hematocrit (%)	42	50		41	39	39		44	45		45	45		43			45	44
				Increase or decrease	0	8		−1	−2	−2		2	3		3	3		1			3	1
2	8.3		13.45′~16.30′	Hematocrit (%)	36	44		39		34		43	41		43	43						
				Increase or decrease	0	8		3		−2		7	5		7	7						
3	10.5		10.20′~12.50′	Hematocrit (%)	33	40		38	36	39	40	44	44		43	43		44	46	46	46	
				Increase or decrease	0	7		5	3	6	7	11	11		10	10		11	13	13	13	
4	9		17.00′~20.00′	Hematocrit (%)	38	53		45	44	46	45	45	46	46	47	47	45		45	45		
				Increase or decrease	0	15		7	6	8	7	7	8	8	9	9	7		7	7		
5	7.5		14.25′~16.25′	Hematocrit (%)	33	40		37		37	34		36	35		33			33		33	
				Increase or decrease	0	7		4		4	1		3	2		0			0		0	

※ Hours from the interruption to the start and the end of fluid transfusion; injected fluid and its volume.
† indicates time of death.

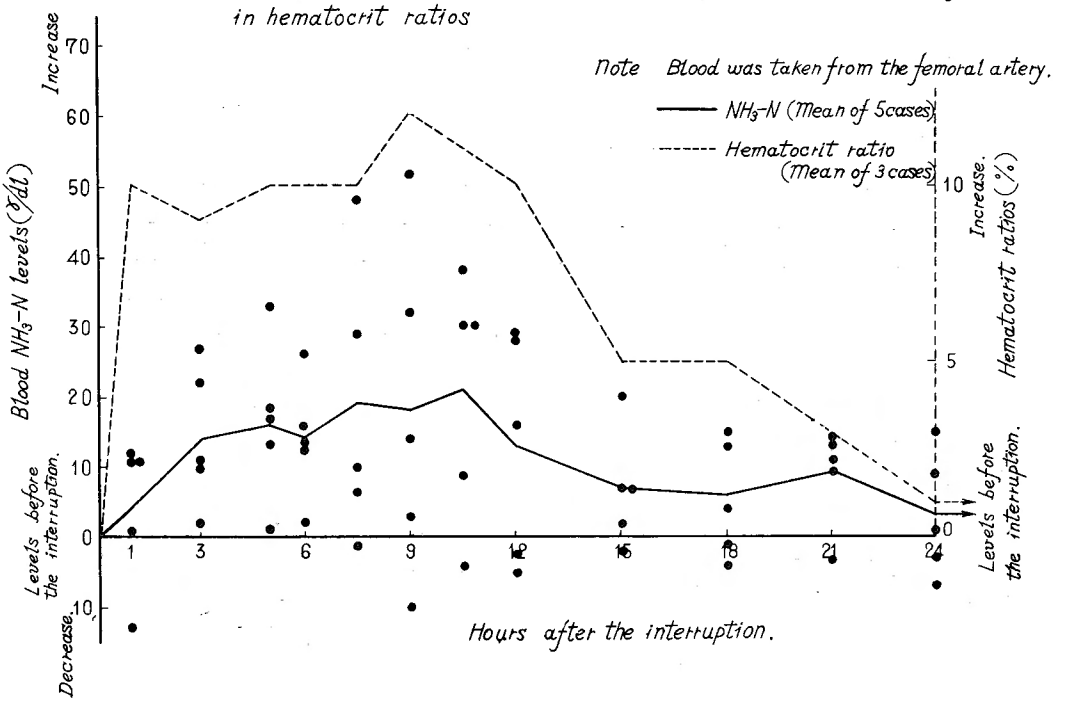
Table. 7. a. Changes in blood NH_3-N levels and hematocrit ratios in dogs which survived the interruption of the hepatic artery by the administration of penicillin.

Dog. No.	Wt. (Kg)	Sex	Before the interruption	NH_3-N levels (γ/dl) after the interruption. ※1	3	5	6	7.30	9	10.30	12	15	18	21	24	Increase or decrease after the interruption. ※1	3	5	6	7.30	9	10.30	12	15	18	21	24
1	12	♂	51	63	73	52	65	50	54	47	46	58	47	64	60	12	22	1	14	1	3	4	5	7	4	13	9
2	15	♂	70	81	97	7	72	77	60	79	68	77	85	84	85	11	27	17	2	7	10	9	2	7	15	14	15
3	8	♀	44	44	54	57	57	54	58	74	72	46	48	55	44	0	10	13	13	10	14	30	28	2	4	11	3
4	10	♀	53	64	64	71	69	82	85	91	69	51	52	50	52	11	11	18	16	29	32	38	16	2	1	3	1
5	11	♂	58	45	60	91	84	106	109	88	87	78	71	67	50	13	2	33	26	48	51	30	29	20	13	9	8
															Mean of increase or decrease.	4	12	16	14	19	18	21	13	7	6	9	3

Dog. No.	Wt. (Kg)	Sex	Before the interruption	Hematocrit ratios (%) after the interruption ※1	3	5	6	7.30	9	10.30	12	15	18	21	24	Increase or decrease after the interruption. ※1	3	5	6	7.30	9	10.30	12	15	18	21	24
3	8	♀	31	40	40	44	43	45	48	45	45	38	36	33	32	9	9	13	12	14	17	14	14	7	5	2	1
4	10	♀	32	43	42	41	42	41	40	40	38	37	35	33	32	11	10	9	10	9	8	8	6	5	3	1	0
5	11	♂	30	40	37	37	37	36	42	40	40	34	36	35	33	10	7	7	7	6	12	10	10	4	6	5	3
															Mean of increase or decrease	10	9	10	10	10	12	11	10	5	5	3	1

Note ※ indicates hours after the interruption.
Blood was taken

Fig. 7a. Changes in blood NH_3-N levels in dogs which survived the interruption of the hepatic artery by the administration of penicillin with changes in hematocrit ratios



which survived the ligation of the hepatic artery (Fig. 7a, Table 7a).

It has been well noticed since the time of MARKOWITZ[15] that penicillin is effective in saving animals with the hepatic artery ligated. There have also been many reports that penicillin therapy done only for a short duration after the ligation can diminish the mortality of animals operated on. URABE, for example, says that only a dose of 100,000 units is sufficient for the purpose. Moreover, the penicillin thus given can stay in the blood or the liver for no more than several hours.[25] On the other hand, VOGEL[35] reported on a function of penicillin to suppress the oxygen consumption in the tissue, and NAKASE has made it clear that this drug is effective in preventing the release of hepatic ferritin from an anoxic liver after the ligation of the hepatic artery.

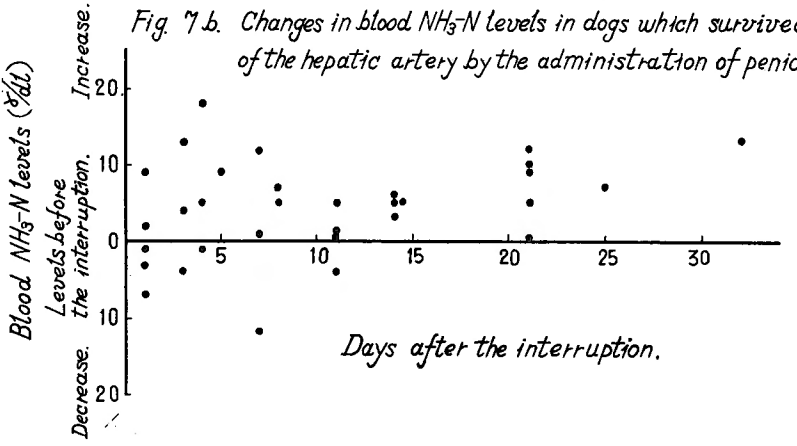
Now, in my present experiment, 10 dogs were subjected to the ligation of the hepatic artery. Just after the operation, they received 100,000 units of crystalline potassium penicillin G intraperitoneally and 300,000 units of penicillin in aqueous suspension intramuscularly. Six of them survived. In 5 surviving dogs, the blood ammonia levels were followed up till the 24th postoperative hour. All of them showed a temporary increase of 20 to 50 r/dl between the 3rd and the 12th hour after the ligation, after that the levels again reduced nearly to the initial ones. In 3 dogs, the Ht. ratios were checked up. About 60 minutes following the ligation, they increased by some 10%, and from then on they showed slight changes up to

Table. 7b. Changes in blood NH_3-N levels in dogs wich survived the interruption of the hepatic artery by the administration of penicillin.

Dog No.	wt. (Kg)	Sex	Before the interruption	Blood NH_3-N levels (γ/dl) after the interruption. ※ 1	3	4	5	7	8	11	14	21	25	32
1	12	♂	58	60	62	57		70	65		63	63		
2	15	♂	75	84	88	93		76		75	80	87		
3	8	♀	44	41			53		49	45	50	44		
4	10	♀	53	52		58				58		62	60	66
6	11	♂	55	48	51					51	58	65		

Note: ※ indicates days after the interruption.
Blood was taken from peripheral vein.

Fig. 7b. Changes in blood NH_3-N levels in dogs which survived the interruption of the hepatic artery by the administration of penicillin.



the 12th hour, after which, gradually declining, they returned to the preoperative levels at about the 24th hour after the ligation. Furthermore, in 5 dogs the levels were observed after the 24th postoperative hour (Fig. 7b, Table 7b). The blood ammonia levels showed a slight increase in some cases from the 1st till the 5th postoperative day. The increase, however, was a little over the range of physiological changes. In comparison with the fate of control dogs (which died within 30 hours after the ligation), that of most dogs operated on may well have been decided before the 15th postoperative hour. This makes us realize that penicillin is necessary only for a short period after the ligation of the hepatic artery.

(8) The ammonia tolerance tests by means of intravenous injection of ammonium chloride (NH_4Cl) in penicillin treated dogs that survived the ligation of the hepatic artery (Fig. 8a, b, Table 8a, b).

After intravenous injection of 2.5 cc per kg of 1% NH_4Cl solution, the ammonia levels in peripheral blood were followed up to observe the function of disposing of ammonia. In 11 normal dogs the levels returned nearly to the initial ones 20 minutes after the injection. On 4 of the 6 penicillin-treated surviving dogs NH_4Cl tolerance tests were performed. From the 1st till the 5th day after ligation the return to

Table. 8 a. Ammonium tolerance tests in normal dogs through intravenous NH_4Cl injection.

Dog No.	Wt. (Kg)	Sex	Before the injection.	NH_3-N levels (γ/dl) after the injection. ※ 5′	10′	15′	20′	Increase or decrease after the injection. 5′	10′	15′	20′
1	15	♂	59	248	129	93	65	189	70	34	6
2	8	♂	60	199	108	82	56	139	48	22	−4
3	15	♀	45	219	130	86	43	174	85	41	−2
4	9	♂	66	222	126	101	77	156	60	35	11
5	15	♂	53	267	170	115	62	214	117	62	9
6	10	♂	89	266	131	88	89	177	42	−1	0
7	12	♂	47	198	92	51	54	151	45	4	7
8	12	♂	73	294	139	85	75	221	66	12	2
9	15	♂	85	400	196	136	110	315	111	51	25
10	8	♀	75	214	106	92	69	139	31	17	−6
11	10	♂	74	252	151	105	69	178	77	31	−5
Mean of 11 cases			66	253	134	85	70	187	69	28	4

Note: 1. 2.5 cc/kg of 1% NH_4Cl was intravenously injected.
2. ※ indicates minutes after the injection.
3. Blood was taken from peripheral vein.

Table. 8 b Ammonium tolerance tests in dogs which survived the interruption of the hepatic artery by the administration of penicillin.

No 1 dog.

Days after the interruption	Before the injection.	NH_3-N levels (γ/dl) after the injection. ※ 5′	10′	15′	20′	Increase or decrease after the injection. ※ 5′	10′	15′	20′
Before the interruption	58	278	124	70	55	220	66	12	3
1	60	297	135	95	73	237	75	35	13
2	64	258	143	110	88	194	79	46	24
3	62	253	136	89	74	191	74	27	12
4	57	201	115	85	79	144	58	28	22
6	76	263	142	98	84	187	66	22	8
8	65	246	111	86	61	181	46	21	4

No.2. dog.

Days after the interruption	Before the injection	NH_3-N levels (γ/dl) after the injection. ※ 5′	10′	15′	20′	Increase or decrease after the infection. 5′	10′	15′	20′
Before the interruption	75	390	186	126	100	315	111	51	25
1	84	519	285	219	156	435	201	135	72
2	88	385	238	178	128	297	150	90	40
3	78	395	247	189	145	317	169	111	67
4	93	496	247	212	161	403	154	119	68
7	76	386	227	166	117	310	151	90	41
9	83	337	192	148	108	254	109	65	25

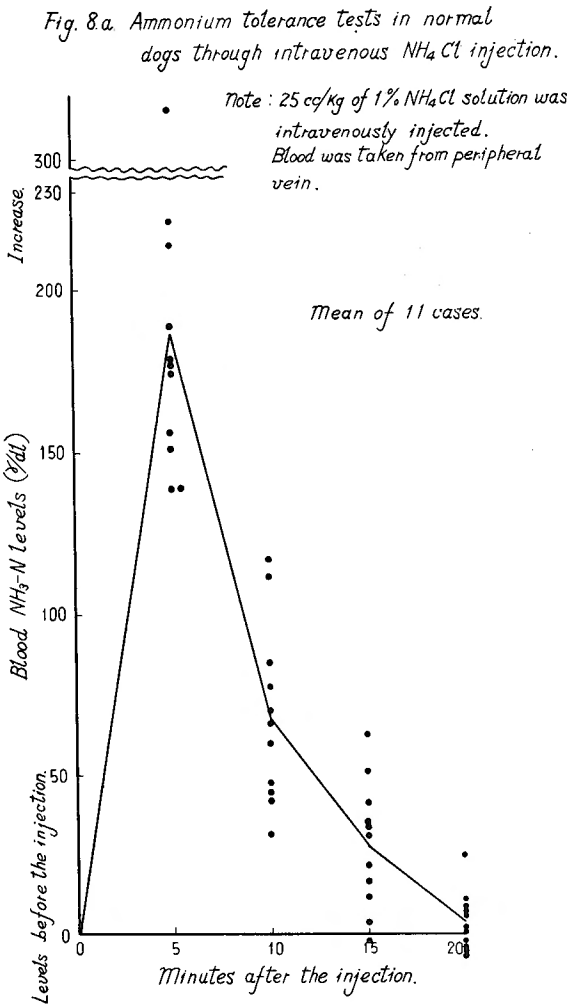
No. 3. dog.

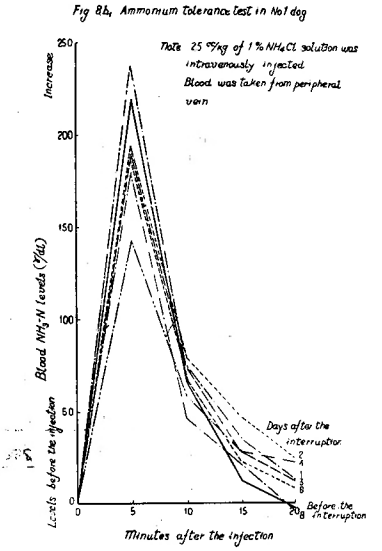
Days after the interruption	Before the injection.	NH_3-N levels (γ/dl) after the injection. ※ 5′	10′	15′	20′	Increase or decrease after the injection. ※ 5′	10′	15′	20′
5	53	239	154	105	78	186	101	52	25
11	45	210	112	71	43	165	67	26	2
14	50	223	106	74	59	173	56	24	9
21	44	224	88	64	49	180	44	20	5

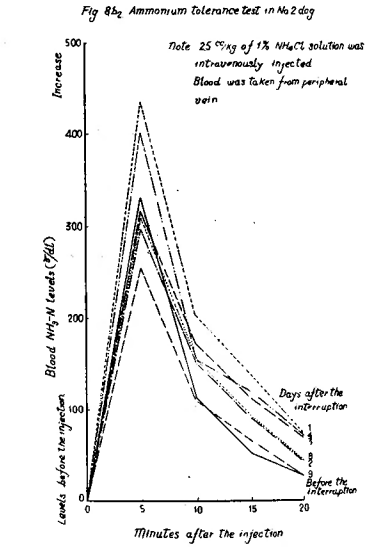
No.4. dog.

Days after the interruption	Before the injection	NH_3-N levels (γ/dl) after the injection 5′	10′	15′	20′	Increase or decrease after the injection. 5′	10′	15′	20′
1	52	213	139	99	95	161	87	47	43
4	58	232	153	104	89	174	95	46	31
11	58	147	103	81	67	89	45	23	9
25	60	157	89	77	70	97	29	17	10
32	66	201	108	84	72	135	42	18	6

Note: 1. 2.5 cc/kg of 1% NH_4Cl solution was intravenously injected.
2. ※ indicates minutes after the injection.
3. Blood was taken from peripheral vein.





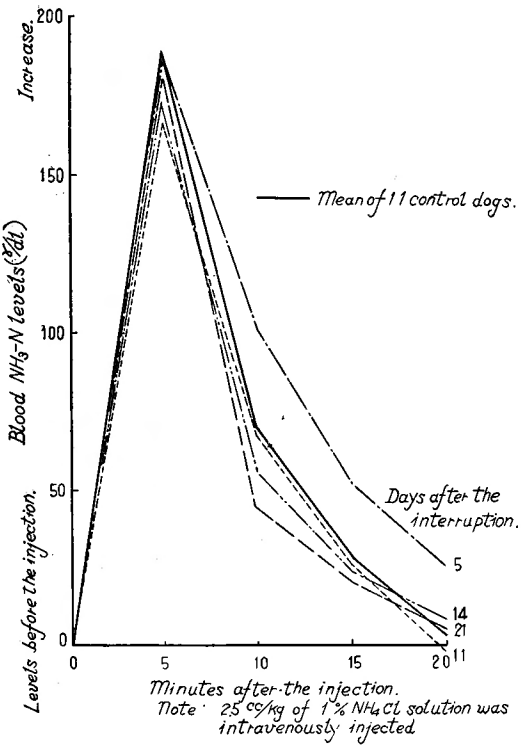


the initial levels was delayed. A week after the ligation, however, it was observed to have been restored to the preoperative state.

(9) Changes in the blood ammonia levels and Ht. ratios in dogs which survived the ligation of hepatic artery by the application of acetylcholine and atropine at the operation (Fig. 9, Table 9).

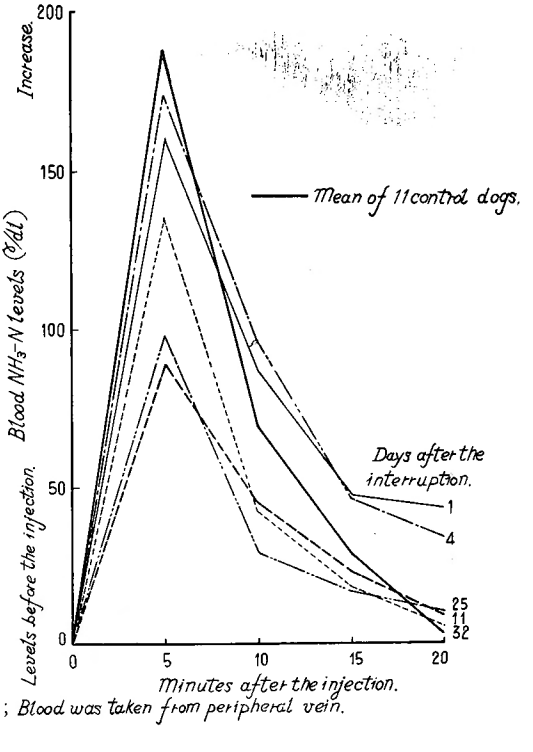
NAKASE in our clinic made this experiment for the first time. An acetylcholine shock was produced before the ligation of the hepatic artery so as to decrease hepatic ferritin beforehand. After the ligation atropine was given to suppress the release of hepatic ferritin from the anoxic liver caused by ligation. The occurrence of liver necrosis was prevented and the mortality diminished. In this way he made it clear

Fig. 8.b3. Ammonium tolerance test in No.3 dog.



Note: 25 cc/kg of 1% NH_4Cl solution was intravenously injected

Fig. 8.b4. Ammonium tolerance test in No.4 dog.



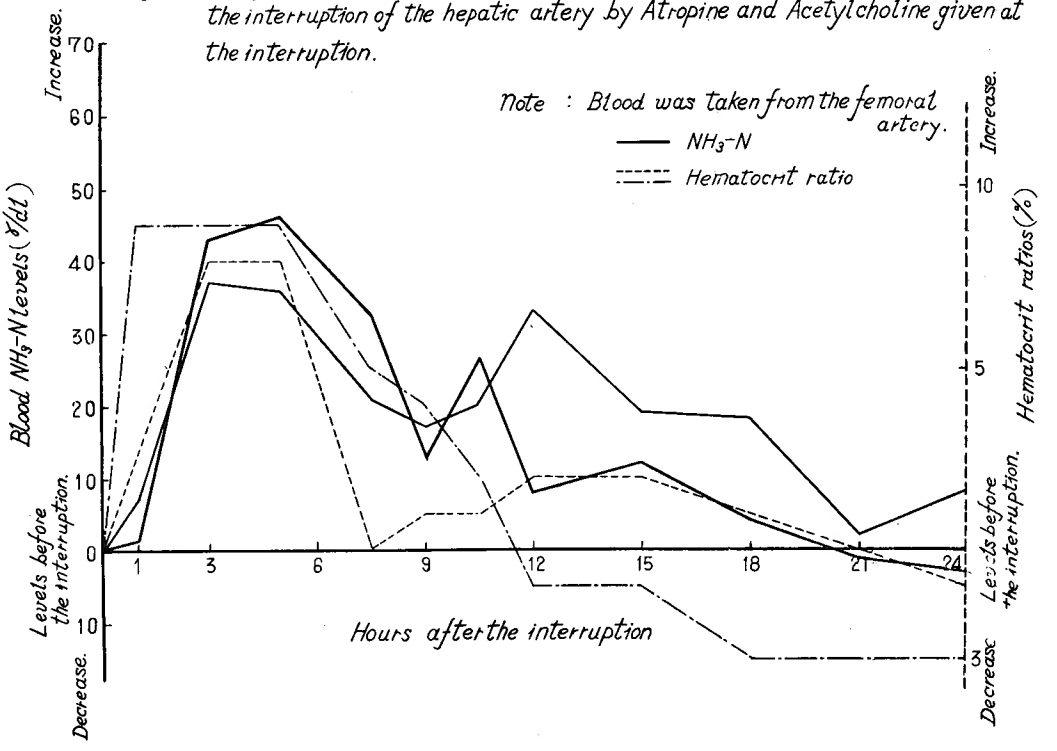
; Blood was taken from peripheral vein.

Table. 9. Changes in blood NH_3-N levels and hematocrit ratios in dogs wich survived the interruption of the hepatic artery by Atropine and Acetylcholine given at the interruption.

Dog. wt. No (Kg) Sex		Before the interruption	Hours after the interruption. 1	3	5	7.30	9	10.30	12	15	18	21	24
1 . 8 ♀	NH_3-N (γ/dl)	73	74	116	119	108	86	100	81	85	77	72	70
	Increase or decrease	0	1	43	46	35	13	27	8	12	4	−1	−3
	Hematocrit-R(%)	37	46	46	46	42	41	39	36	36	34	34	34
	Increase or decrease	0	9	9	9	5	4	2	−1	−1	−3	−3	−3
2 9 ♂	NH_3-N (γ/dl)	51	58	88	87	72	68	71	84	70	69	53	59
	Increase or decrease	0	7	37	36	21	17	20	33	19	18	2	8
	Hematocrit-R(%)	30	33	38	38	30	31	31	32	32	31	30	29
	Increase or decrease	0	3	8	8	0	1	1	2	2	1	0	−1

Note : Blood was taken from the femoral artery.

Fig. 9. Changes in blood NH_3-N levels and hematocrit ratios in dogs wich survived the interruption of the hepatic artery by Atropine and Acetylcholine given at the interruption.



that hepatic ferritin plays some role in the development of necrosis.

Thus in the present study, 100 mg acetylcholine was intravenously injected on the previous night and 3 hours before the ligation of the hepatic artery. After the ligation, 0.5 mg atropine was given every hour for 10 hours. Two of the 4 dogs survived the above mentioned experiment. In these two, the blood ammonia levels were observed up to the 24th hour following the ligation. During the period from the 3rd till the 8th hour and towards the 12th hour, the levels showed an increase of 33 to 46 r/dl and then returned to the initial levels.

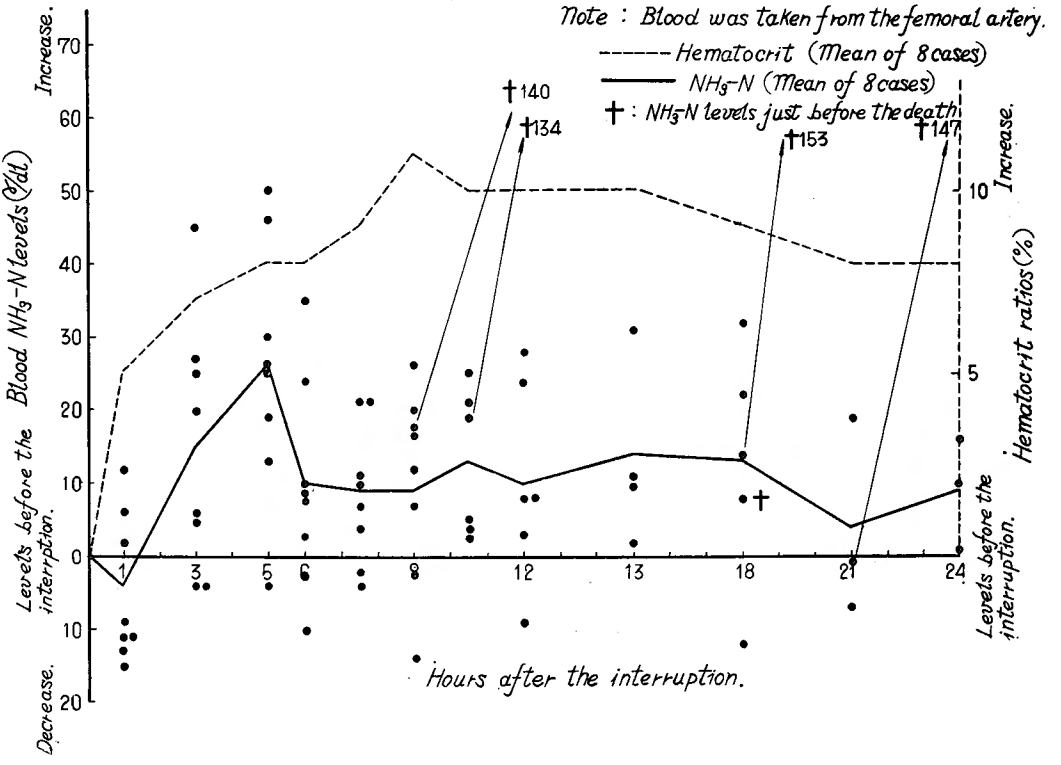
The Ht. ratios rose 8 to 9% from the 60th minute till the 5th hour following the ligation, after which they gradually dropped to the initial levels where they remained after the 12th hour in all cases but one in which the level went farther down.

From the above, it seems clear that the fate of dogs operated on had already been decided before the 15th postoperative hour.

(10) Changes in the blood ammonia levels and Ht. ratios in ascitic dogs after the ligation of the hepatic artery (Fig. 10, Table 10a, b).

In Nos. 1 and 2 dogs the ascites was removed by paracentisis on both the day and the previous night of the ligation in order to lessen the changes in abdominal pressure which would be caused by the intraabdominal operative procedure at the

Fig. 10. Changes in blood NH_3-N levels and hematocrit ratios in ascitic dogs after the interruption of the hepatic artery.



time of the ligation. The blood ammonia levels increased by 20 to 40 r/dl from the 9th till the 21st postopeative hour, but were restored to the preoperative levels after 24 or 30 hours. No. 1 dog died accidentally on the 3rd day and No. 2 dog died of peritonitis on the 7th day postoperatively. In Nos. 3 to 8 dogs the ascites was not removed before the ligation. In most of them the blood ammonia levels showed a temporary increase of 20 to 50 r/dl during the period from the 3rd till the 6th postoperative hour, after which they declined nearly to the initial levels. And just before the death, as high a level as about 150 r/dl was suddenly reached in all cases except No. 6 dog. No. 7 dog was a little different. Although it showed a preoperative level of 90 r/dl, a little higher than in the other ascitic and the normal dogs, it remained within the range of 90 ± 10 r/dl until its death.

As in normal dogs, the Ht. ratios rose in all the 8 cases after the ligation, though in Nos. 1, 5 and 7 dogs the rise was of a slight degree. In Nos. 1 and 2 dogs, the levels decreased nearly to the initial ones on the 2nd day, after which no changes were observed till they died. In Nos. 3 to 8 dogs the increase in the Ht. ratio was not followed by a decrease in it.

On gross examination at autopsy, no case revealed liver necrosis, but most of them showed congestion of high degree.

(11) Comparison between the blood ammonia levels reached just before the

Table. 10. a. Changes in blood NH_3-N levels in ascitic dogs after the interruption of the hepatic artery.

Dog No.	Wt. (Kg)	Ascites (l)	Sex	Before the interruption.	NH₃-N levels (γ/dl) after the interruption. ※1	3	5	6	7.30	9	10.30	12	15	18	21	24	Increase or decrease after the interruption. ※1	3	5	6	7.30	9	10.30	12	15	18	21	24
1	10.5	3	♂	52	41	58	77	62	73	78		76		84		68	-9	6	25	10	21	26		24		32		16
2	14	4	♂	40	29	45	36	48	47	60	61	68	71	62	59	50	-11	5	-4	8	7	20	21	28	31	22	19	10
3	12.5	3.5	♂	53	65	98	83	77	64	71		140†					12	45	30	24	11	18		87†				
4	8	1	♂	41	28	37	60	39	45	48	46	49	52	55	1900† 153		-13	-4	19	-2	4	7	5	8	11	14	112†	
5	8.5	2	♀	68	53	88	118	103	89	80	93	71		56	61	68	-15	20	50	35	21	12	25	3		-12	-7	0
6	17	7	♀	52	54	79	78	55	48	50	56	43	54	60†			2	27	26	3	-4	-2	4	-9	2	8†		
7	7.5	1.5	♀	90	79	86	103	80	100	76	93	98	100		89	147†	-11	-4	13	-10	10	-14	3	8	10		-1	57†
8	10	2	♂	72	88	97	118	81	70	89	91	134†					6	25	46	9	-2	17	19	62†				
																	Mean of increase or decrease.											
																	-4	15	26	10	9	9	13	10	14	13	4	9

Note: ※ indicates hours after the interruption.
† means time of death.
Blood was taken from the femoral artery.

Addendum: No. 1 dog: 51 at the 30th hour, 65 on the 2nd, 48 on the 3rd day. (γ/dl) died in accident on the 3rd postoperative day.
No. 2 dog: 38 on the 2nd, 38 on the 3rd, 49 on the 4th, 54 on the 5th, 45 on the 6th day. (γ/dl) died of peritonitis on the 7th day.
No. 3 dog: died with acute enlargement of the stomach.
No. 5 dog: died at the 30th hour.

Table. 10. b. Change in hematocrit ratios in ascitic dogs after the interruption of the hepatic artery.

Dog No.	Wt. (Kg)	Ascites (l)	Sex	Before the interruption.	Hematocrit ratios (%) after the interruption. ※1	3	5	6	7.30	9	10.30	12	15	18	21	24	Increase or decrease after the interruption. ※1	3	5	6	7.30	9	10.30	12	15	18	21	24
1	10.5	3	♂	25	29	29	28	32	32	29		31		30		32	4	4	3	7	7	4		6		5		7
2	14	4	♂	26	33	35	35	36	37	39	36	36	35	35	34	33	7	9	9	10	11	13	10	10	9	9	8	7
3	12.5	3.5	♂	35	39	47	50	48	48	52		52				28 on 2nd day	4	12	15	13	13	17		17				
4	8	1	♂	28	36	36	37	39	37	40	41	39	40	42			8	8	9	11	9	12	13	11	12	14		2
5	8.5	2	♀	40	44	44	44	45	45	45	45	45		45	45	45	4	4	4	5	5	5	5	5		5	5	5
6	17	7	♀	33	38	45	46	47	45	47	47	47		47	45		5	12	13	14	12	14	14	14		12		
7	7.5	1.5	♀	21	24	24	24	26	25	27	27	27	27		32	33	3	3	3	5	4	6	6	6	6		11	12
8	10	2	♂	34	38	39	40	42	45	48	48	48					4	5	6	8	11	14	14	14				
																	Mean of increase.											
																	5	7	8	8	9	11	10	10	10		8	8

Note: ※ means hours after the interruption.
† indicates the time of the death.
Blood was taken from the femoral artery.

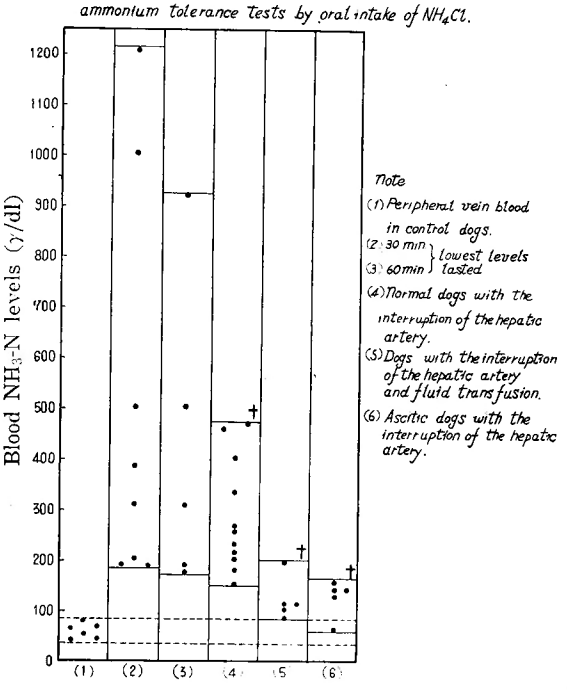
death in dogs with the ligation of hepatic artery and the lowest levels remaining for 30 or 60 minutes during ammonia tolerance tests by means of NH_4Cl through mouth (Fig. 11).

Six dogs which survived ligation of the hepatic artery by postoperative penicillin were orally given 0.5 to 1 g per kg of NH_4Cl about 10 days after the ligation.

The lowest levels which could be maintained for 30 or 60 minutes without endangering their lives were 200 to 1200 r/dl or 200 to 900 r/dl respectively.

On the other hand, in normal dogs the ammonia level after the ligation of the hepatic artery is at highest 100 to 150 r/dl. About 60 minutes before the death, it showed a sudden, rapid increase as high as 170 to 470 r/dl. It is similarly observed in the ascitic dogs and the dogs which received fluid transfusion after the ligation of the hepatic artery: the ammonia levels showed a rapid rise, reaching 60 to 150 r/dl and 86 to 200 r/dl respectively, but not so high as in normal dogs. It may be properly assumed that the rapid rise in the ammonia level were not due to a rapid advance in degeneration of liver parenchyma but due to a high degree of hepatic as well as systemic circulation impediment at the agonal stage,[6] for it would be unreasonable to suppose such a rapid advance of the degeneration in such a short time.

Fig. 11. Blood NH_3-N levels just before the death in dogs with the interruption of the hepatic artery and the lowest levels lasted for 30 or 60 minutes in ammonium tolerance tests by oral intake of NH_4Cl.



The dogs (with ligation of the hepatic artery under the administration of penicillin) could endure as high levels as 200 to 1200 r/dl for 30 to 60 minutes, whereas the normal dogs died as a result of the ligation with ammonia levels of 170 to 470 r/dl, and the dogs receiving fluid transfusion and the ascitic ones died with lower ammonia levels. From this, it is clear that the increase in blood ammonia level and its duration after the ligation of the hepatic artery is not the direct cause of their death.

CLINICAL CASES

In all cases, measurements of ammonia levels and Ht. ratios were performed on antecubital vein blood drawn at a fasting state early in the morning by exactly the same method as in the animal experiment.

In 7 healthy adult persons, the blood ammonia levels were 33 to 68 r/dl, with the average of 50 r/dl. This accords with the results of CONWAY and others. The Ht. ratios were 38 to 45%. In 8 cases with liver cirrhosis or BANTI's syndrom, the levels of blood ammonia were 51 to 193 r/dl, averaging 98 r/dl, with the Ht. ratios ranging from 22 to 52% (Fig. 12, Table 12).

Preoperative treatment with 1 g per day of streptomycin caused the blood

Table. 12. Peripheral blood NH_3-N levels and hematocrit ratios in healthy adult persons and patients with liver diseases.

(1) Healthy adult persons.

Name	Age	Sex	NH_3-N(γ/dl)	Hematocrit (%)
M. T.	30	♂	48	43
T. S.	54	♀	43	41
M. O.	55	♂	33	38
W. K.	22	♀	40	39
T. O.	51	♂	53	48
Y. K.	23	♀	68	38
M. H.	35	♂	62	45
Mean			50	

(2) Patients with liver disease.

Name	Age	Sex	Disease	NH_3-N(γ/dl)	Hematocrit (%)
K. T.	46	♀	Liver cirrhosis	193	34
F. M.	42	♀	〃	92	22
T. N.	28	♀	〃	84	37
K. R.	51	♂	〃	83	52
N. O.	32	♂	〃	76	
K. O.	33	♂	〃	53	52
H. M.	31	♂	〃	51	37
N. S.	20	♂	Banti's syndrom	152	40
N. O.	48	♂	Malignant liver tumor	210	

Note : Antecubital vein blood was drawn at a fasting state in the early morning

Fig. 12. Peripheral blood NH_3-N levels in healthy adult persons and patients with liver diseases.

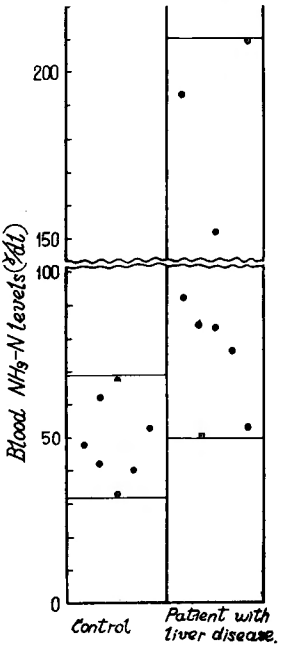
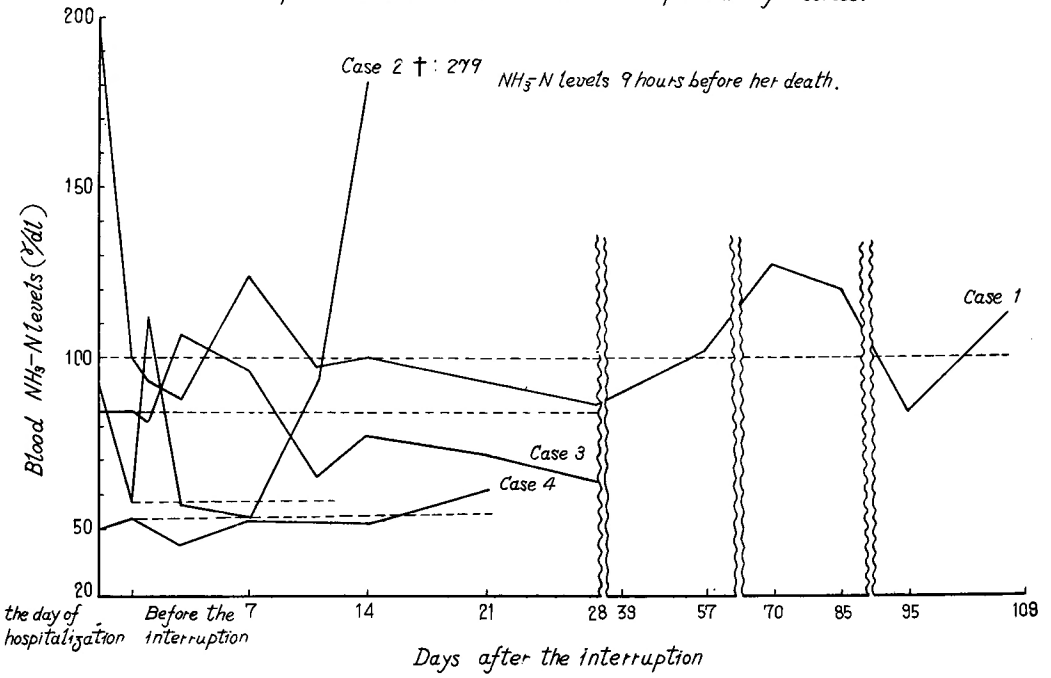


Table 13. Changes in blood NH_3-N levels and Hematocrit ratios after the interruption of the hepatic artery in patients with liver cirrhosis accompanied by ascites.

Patient	Sex	age		the day of hospitalization.	Before the interruption.	Days after the interruption. 1	3	7	11	14	21	28	39	57	70	85	95	109
Case 1	♀		NH_3-N(γ/dl)	193	99	93	88	124	97	100		86	90	102	127	120	84	112
K. T.		46	Hemato-crit-R(%)	34	34	40	38	44	40	37		32	33		32	32	33	
Case 2	♀		NH_3-N(γ/dl)	92	58	112	56	53	92	279 †								
F. M.		42	Hemato-crit-R(%)	22	25	25	26	29	25	43								
Case 3	♀		NH_3-N(γ/dl)	84	84	81	107	96	65	77								
T. N		28	Hemato-crit-R(%)	37	35	35	36	32	38	33								
Case 4	♂		NH_3-N(γ/dl)	50	53	50	45	52		51								
K. O.		33	Hemato-crit-R(%)	52	61	54	53	52		44								

Note Antecubital vein blood was drawn at a fasting state in the early morning all the cases except Case 2. are alive for the present, April 1 1959
Case 2 died of Uremia on 14th day.

Fig. 13 Changes in blood NH_3-N levels after the interruption of the hepatic artery in patients with liver cirrhosis accompanied by ascites.



ammonia level to decrease from 193 to 99 r/dl in Case 1, and from 92 to 58 r/dl in Case 2. In Cases 3 and 4, however, no such decrease were observed (Fig. 13, Table 13).

Cases 1, 3 and 4 were followed up until April 1, 1959, that is 109, 28 and 21 days after the ligation of the hepatic artery respectively. The blood ammonia levels showed a slight increase over or decrease against the preoperative level. The fluctuation was at most within ± 25 r/dl in Cases 1 and 3. They once showed a temporary increase of 25 and 23 r/dl respectively within a week after the ligation, after which they remained no higher or a little lower than the initial level. Case 1, however, again disclosed an increase of 27 to 20 r/dl from the 70th through 85th postoperative day. In Case 4 the blood ammonia level remained within the range of 50 ± 10 r/dl, with little changes.

The Ht. ratios showed a very little fluctuation except in Case 1, which revealed a 3 to 10% increase about 2 weeks after the ligation.

In Case 2 the blood ammonia level rose to 112 r/dl 18 hours postoperatively, but later dropped to 53 to 56 r/dl where it stayed from the 3rd till the 7th day. On the 11th day it again began to rise and on the 14th day, 9 hours before her death as high a level as 279 r/dl. About 2 days before the death, this case disclosed such symptoms as oliguria, a marked increase in blood N. P. N. and mental confusion. At autopsy, histological examinations revealed that there were no necrotic changes in the liver but hemoglobinuric nephritis in the kidney.

DISCUSSION

In normal dogs, the ligation of the hepatic artery bring about a high anoxic liver not only by interrupting arterial supply of oxygen to the liver but also by producing portal circulatory disturbance. From this anoxic liver develops a necrotic liver through a high degree of parenchymal degeneration, as URABE, ISHIGURO, NAKASE, MIYAWAKI and others have experimentally shown. On the other hand, NELSON, EISEMAN[7),22),23),27)] and others pointed out that the hepatic function to dispose of ammonia is failed with a general anoxic state of the liver and with a high degree of parenchymal degeneration, not to mention with a decrease in the hepatic blood flow.

In normal dogs, a ligation of the hepatic artery cause a 40 to 70 r/dl increase in peripheral blood ammonia 6 to 10 hours after ligation. Symptoms which immediately follow the ligation are congestion of the liver and, as to the quality of blood, hemoconcentration, that is, a rise in the Ht. ratio, as a forerunner to an increase in blood ammonia. If the view is to be held that hemoconcentration which soon occurs is due to local leakage of plasma into the damaged part, the hemoconcentration which immediately followed the ligation is considered as a result of hepatic circulatory failure, that is, stasis.

If there was no bleeding worthy of mention during the operative procedure, an increase from 30 to 40% in the Ht. ratio without any bleeding will have made the amount of plasma equal to 25% of the total blood volume leak out of the systemic circulation system. This condition may well be called "shock". And such an increase in the Ht. ratio was observed in all cases after the ligation. In some cases the blood ammonia level showed a temporary decrease about 60 minutes after the ligation. This phenomenon can be explained by the report that there is a temporary spurt in the activity of urea synthesis (in the liver) at an early stage of shock.[36)]

In order to see how rapidly the disturbance of the hepatic circulation would affect the blood ammonia levels and Ht. ratios, congestion was experimentally produced by constricting the hepatic veins. The blood ammonia levels and Ht. ratios increased rapidly. On the contrary, when fluid transfusion, which would increase the blood volume of systemic and consequently hepatic circulation, was applied after the hepatic arterial ligation, the ammonia levels and Ht. ratios in peripheral blood declined for a while. These facts prove that the hepatic circulatory disturbance is one of the cause of the increase in the blood ammonia levels and Ht. ratios occurring after the ligation of the hepatic artery.

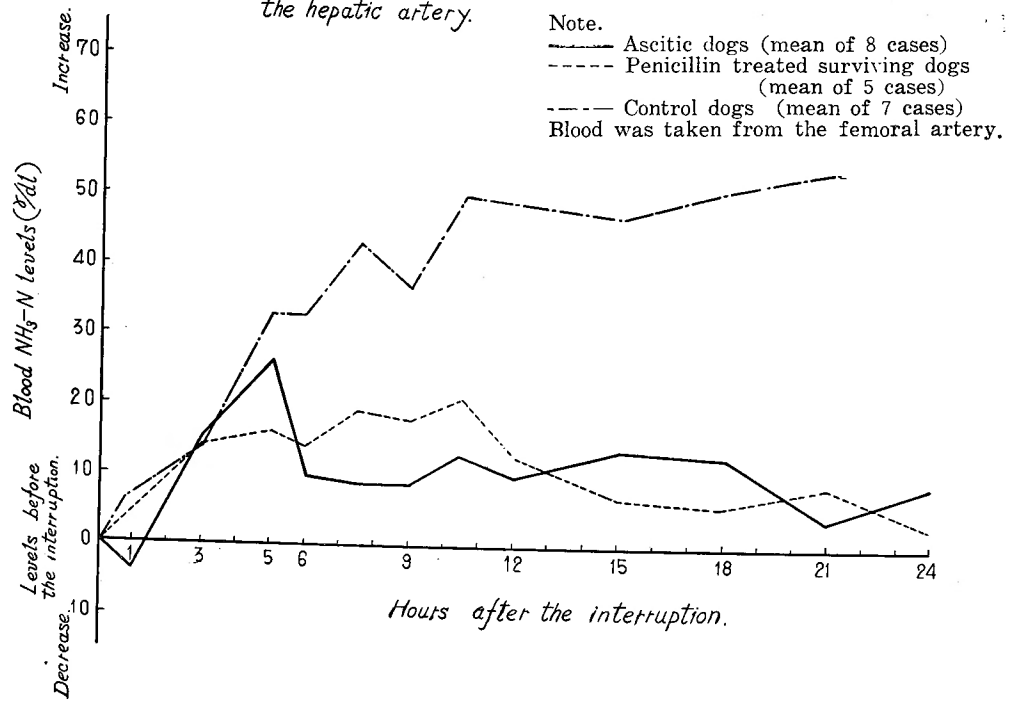
The decrease in the blood ammonia levels and Ht. ratios caused by the fluid transfusion did not last. This may be partly because the transfused 5% glucose solution, balanced electrolyte solution swiftly leaked out of the circulation system, and partly because parenchymal degeneration was ever advancing in the liver.

In those dogs which survived the ligation by penicillin or atropine and acetylcholine, the blood ammonia levels and Ht. ratios in the blood once increased following the ligation, but after 9 to 12 hours returned nearly to the initial levels. The return to the initial levels may be explained in this way: in these cases, a hepatic

circulation impediment, temorary as it was, occurred, depriving the liver of its oxygen supply; but parenchymal degeneration was not advanced to a very high degree, so that the circulatory disturbance recovered before long, when the liver had adapted itself to the hepatic circulation by the portal blood only. If the liver well tolerated the ligation of the hepatic artery, it does not necessarily mean that is would immediately recover its normal function to dispose of ammonia. Ammonia tolerance tests performed on dogs which survived the ligation revealed some degree of impediment to the function which lasted for about a week after the ligation. FRASER and others[8] obtained similar results from thymol turbidity tests, bromsulphalein clearance tests etc.

In ascitic dogs, from 3 through 6, or from 9 through 21 hours following the ligation, the blood ammonia levels showed a temporary increase. Otherwise, however, they stood around the initial levels. This proves that these dogs have the same adaptability to the hepatic circulation by the portal blood only as had those dogs which survived the ligation by the administration of penicillin etc. This findings also accords with the following observations which the investigators in our clinic have reported. NAKASE found that the liver of ascitic dogs already showed a marked decrease in hepatic ferritin before the ligation of the hepatic artery, so that little was left to be released after the ligation. HOSONO observed a lasting increase in the rate of the portal blood flow, and ADACHI an increase in the oxygen content of the portal blood after the ligation.

Fig. 14 Changes in blood NH_3-N levels after the interruption of the hepatic artery.



Autopsy disclosed no necrotic changes in the liver. In only two of the 8 dogs, however, the Ht. ratios returned to the initial levels after a while, with the other six holding the increased levels. This may be explained in the light of the autopsy finding that all the 8 cases had congestion in the liver.

In Fig. 14 a comparison is made between the postoperative blood ammonia curves in normal dogs and in ascitic dogs or the dogs which survived ligation of the hepatic artery with the help of penicillin. In normal ones, the rise is not followed by a decline until the death, while in the others, a temporary rise is brought about by a hepatic circulatory failure due to the ligation. This circulatory failure, however, does not develop into parenchymal degeneration, but the liver comes to adapt itself to the circulation by the portal blood only, with the result that the ammonia levels soon return to normal.

On Clinical Cases

In most cases with liver cirrhosis or BANTI's syndrom, the ammonia levels in peripheral blood are generally higher than in healthy adults, but in some cases they remain within normal range. The difference may have occurred because the ammonia levels varied according not only to the degree of hepatic parenchymal damage the patients suffered, but also to the anatomy of the shunt between the systemic and the portal circulation.

In cases with cirrhosis accompanied by ascites, ligation of the hepatic artery cause little changes in the ammonia content of blood, just as had been expected from the results of the experiments on ascitic dogs. KITANI and others[14] pointed out that in cirrhotic cases with high blood ammonia levels, antibiotics, such as streptomycin which are effective in suppressing intestinal bacteria, prevented the production of ammonia in the intestines, lowering the ammonia level in peripheral blood. This was proved by Case 1 in the present study. The patient had an ammonia level of 193 r/dl when hospitalized, but administration of streptomycin brought it down to 99 r/dl just before the ligation of the hepatic artery. After the ligation, it remained within the range of 99 ± 25 r/dl ··· much the same level as before the ligation ··· even though no antibiotics were given. This means that the hepatic functions once impeded had been recovered by the ligation. In this case the ligation had been performed for the purpose of decreasing the ascites. The purpose was attained; both the circumference of the belly and the serum bilirubin decreased.

In Case 2, 18 hours after the ligation of the hepatic artery the blood ammonia increased from the preoperative 58 r/dl to 112 r/dl. The increase, however, may be explained by the fact that a large amount of preserved blood, containing 320 to 500 r/dl of ammonia, was transfused during the operation. This case had been unconscious for 2 days until she died, showing a blood ammonia level as high as 279 r/dl 9 hours before her death. These findings may give the impression that she died of hepatic coma. From the results of various clinical examinations and autopsy findings it is obvious that the cause of the death was uremia. And the increase in the blood ammonia level may be explained in this way : the increase in blood N. P. N. caused an increase in amine, which was detected as ammonia

according to the method of measurement of ammonia employed in the experiment.

SUMMARY AND CONCLUSION

The results of the above experiments and tests and findings of the clinical cases are summed up as follows:

(1) In normal dogs, the ligation of the hepatic artery cause a 40 to 70 γ/dl increase in the blood ammonia level. About 60 minutes before death, the level went high up to 170 to 470 r/dl. The Ht. ratio also showed an increase of 10 to 20%. If fluid transfusion was performed, the blood ammonia levels and Ht. ratios decreased for a longer or shorter time, and moreover the blood ammonia level just before death showed a decrease, with 86 to 200 r/dl.

(2) In those dogs which survived the ligation of the hepatic artery by the help of penicillin or atropine and acetylcholine, the blood ammonia levels and Ht. ratios revealed a temporary rise after the ligation, but soon returned to the initial levels. In the dogs which survived with the help of penicillin, the function to dispose of ammonia was restored to the preoperative state one week after the ligation.

(3) In the ascitic dogs, the ligation of the hepatic artery caused a temporary increase in the blood ammonia level, which was soon restored to the initial level. But it again increased rapidly just before the death. The Ht. ratios were not different on the average from those in normal dogs. Two of the 8 dogs returned to the initial levels on the 2nd days after ligation, and two others showed a slight increase in the Ht. ratio.

(4) In all dogs, the increase in blood ammonia level observed after the ligation of the hepatic artery was not the direct cause of their death.

(5) In the 4 cases with liver cirrhosis accompanied by ascites, the blood ammonia levels remained much the same after the ligation of the hepatic artery as before, with the exception of one case which died of nephritis.

From the above mentioned facts it may be concluded that when normal dogs died after the ligation of the hepatic artery, the increase in the blood ammonia level which is not followed by a decrease is not the direct cause of their death. The increase has been brought about by the hepatic circulatory disturbance combined with parenchymal degeneration. On the other hand, when the dogs survived the interruption of arterial blood flow to the liver, the increase in the ammonia content of blood lasted only for a while after the ligation, in parallel with hepatic circulatory disturbance. A similar tendency was observed in the ascitic dogs. The clinical cases with liver cirrhosis accompanied by ascites tolerated well the ligation of the hepatic artery, unless liver cirrhosis was accompanied by a high degree of renal insufficiency. These cases faced no danger of hepatic coma due to the interruption, maintaining their preoperative levels of the blood ammonia.

I was very fortunate to have the enthusiastic guidance and support of Prof. Dr. CHISATO ARAKI and Assistant Prof. Dr. ICHIO HOSHO throughout this study.

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和文抄録

肝動脈遮断後の血中アンモニア値の変動

(附，ヘマトクリット値の変動)

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Rienhoff 等により創始された，腹水を主訴とする肝硬変症に対する肝動脈遮断術については，賛否両論が相対立している．当教室に於ても，その臨床的応用を検討する為に，各方面より検索が行われており，その研究の一環として，肝に於て特異的に尿素合成により処理され，他方最近肝性昏睡の原因として重要視される血中アンモニア(以下「ア」と記す)を指標として，肝動脈遮断後の肝機能の変動を，実験犬並びに臨床例について追究した．尚遮断後の全身循環状態の一面を窺う為に，ヘマトクリット値(以下「ヘ」値と記す)を同時に測定した．

実験方法：血中「ア」値はConway 氏微量拡散法により，「ヘ」値は Wintrobe 氏法により，実験犬では主として股動脈血，臨床例では肘静脈血について測定した．尚血中「ア」値の採血直後3～5分間の急激な増加の傾向を考慮して，採血後5分より10分の間に検出した「ア」の窒素量をもつて，血中「ア」値とみなした．肝動脈遮断は教室の術式により，総肝動脈，右胃動脈，胃十二指腸動脈を結紮，切断した．腹水犬作製は McKee 氏法により，胸部下空静脈を狭窄した．

実験並びに臨床成績

(1) 正常犬及び健康成人の末梢血の「ア」値は，夫々40～80γ/dl，40～70γ/dlで，その生理的変動は10γ/dl内外で，測定誤差(±3γ/dl)を僅かにこえる程度であつた．

(2) 正常犬で無処置の場合，血中「ア」値は，遮断後6～10時間後より，持続的に40～70γ/dl増加し，死亡直前には更に急激に増加して170～470γ/dlに達した．この死亡直前の血中「ア」値は，NH_4Cl経口負荷試験中30分又は60分間の最低維持値 200～1200γ/dl に比し遙かに低値が含まれており，遮断後にみられる血中「ア」の増量そのものが直接の死因でないことは明らかである．

「ヘ」値は遮断直後より持続的に10%内外増加した．尚全例30時間以内に死亡し，種々の程度の肝の鬱血及び壊死が認められた．

(3) 肝動脈遮断直後より門脈の血行が停滞することが確められている．他方，正常犬で肝静脈を狭窄し，肝を鬱血させると，血中「ア」値，「ヘ」値共に急激に増加する．そこで，肝動脈遮断後輸液を行つて，肝流血量を増加させると，持続時間の長短はあるにしても，血中「ア」値，「ヘ」値共に術前値近くまで減少する．このことは，遮断後の血中「ア」値及び「ヘ」値の増加の原因の一つは肝の血行障害であることを示している．

(4) 肝動脈遮断に際して，乏酸素肝よりの肝 Ferritin の遊離を抑制し，血行の障害された肝が壊死に陥ることを防ぐPenicillin又はAtropineを投与して生存した犬では，血中「ア」値，「ヘ」値共に遮断後3時間よ

り12時間に亘り，一時的に20～50γ/dl増加するが，その後は略々術前値に復帰し経過した．Penicillin 投与により生存した犬の「ア」処理機能を，NH_4Cl 経静脈負荷試験により検すると，遮断後1週間前後は，血中「ア」曲線の負荷前値への復帰は遅延するが，その後は術前と全く差異はなくなる．

(5) 腹水犬では，血中「ア」値は，遮断後3時間より6時間に亘り，又は9時間より21時間に亘り，一時的に20～50γ/dl 増加するが，その他は略々術前値にとゞまる．これらの腹水犬は肝動脈遮断後一時生存するが，肝壊死発生以外の原因でその後死亡し，その際血中「ア」値は死亡直前に急激に増加する．「ヘ」値の変動は対照犬と同様の傾向を示すが，その増加の程度は対照犬に比し軽度のものもあり，中にはPenicillin投与下で生存した犬の如く，術前値に復帰したものもある．

(6) 臨床例4例に於ては，術前より合併した腎炎の悪化のため死亡した1例を除き，術後3～15週間を経た今日まで，術前値の50～120γ/dlを維持しており，特に第1症例は，術前の S. M. 投与により減少した血中「ア」値を，術後は S. M. の投与によらずとも維持している．

以上の成績を肝動脈遮断後の肝壊死発生の機序に関する教室の知見――即ち，肝動脈を遮断すると，門脈の血行障害を来し，肝は極度の乏酸素状態に陥り，この乏酸素肝が肝壊死発生の基盤となる――と併せ考えると，犬が肝動脈遮断により死亡する場合の血中「ア」値の持続的増加は，遮断直後より起る肝血行障害に続く肝実質障害によるものと考えられる．

他方，犬がPenicillin又はAtropine投与により肝動脈遮断に耐えて生存する場合，血中「ア」値は遮断後短時間一時的に増加するが，その後は術前値に復帰して経過し，門脈のみによる肝循環に順応したことを示している．遮断に際してPenicillin 等の投与により生存した犬におけると同様の傾向は腹水犬に於ても認められ，更に臨床的に，腹水を主訴とする肝硬変症は，高度の腎不全を伴わざる限り，肝動脈遮断によく耐え，血中「ア」値も術前値を維持し得て，肝性昏睡の危険はない．