

INFLUENCE OF EXPERIMENTAL LOCAL MEDULLARY HYPOXIA ON THE NUMBER OF RED BLOOD CORPUSCLES

By:

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We based our experiments on the assumption that the best method for the study of medullary function would consist in the direct application, through the nutritive artery of the bone-marrow, of the stimuli influencing medullary function, and in the observation of the effects on the venous blood issuing from the medulla.

According to data available in pertinent literature puncture of the nutritive vein was performed first by *Ascoli*, and subsequently by *F. Müller*. Later *Schoen* and *Berchtold* (1) studied in a small number of experiments the effect of adrenalin, administered to the vein, on the qualitative composition of the blood taken from the vena nutritia tibiae; *Imai* (2) examined the qualitative blood reaction in the venous blood leaving the medulla femoralis to the effect of adrenalin, histamine and thyroxin. *Drinker* (3), (4) reported the appearance of normoblasts as the effect of anoxia in connection with perfusion of the bone-marrow. *Bock* (5) observed the cell-producing activity of the medulla of bones pertaining to the thorax in the isolated circulatory system of this area.

We studied the effect of experimental *local* medullary hypoxia on the dogs *fermur*. Hypoxia was produced in two different ways: 1. by *ligation of the nutritive artery*: a method which will be described in our next publication; and by injection of potassium cyanide.

Ad 1. In order to enable the numerical changes of the red blood corpuscles to be properly evaluated, we determined the daily oscillation in the number of red blood corpuscles of a healthy dog and found a peak value of 500.000 (See Diagram 1.). Ligation of a muscle branch instead of a medullary artery of the same size did not cause any change in the number of red blood corpuscles (See Diagram 2.). The intervention itself either had no effect on this number. Neither the injection of physiological salt solution into the medullary artery nor ligation of the efferent *vein* of the medulla had any effect

on the number of red blood corpuscles. The external circumstances were, of course, the same in each experiment.

Table I. shows the results obtained after *transitory* ligation (of 5, 10, 15, 30 minutes' duration) of the artery. During ligation of the medullary artery, samples for the determinations were taken from the corresponding medullary and the auricular veins simultaneously. *Development of considerable erythrocytosis was observed in both places ; it took place somewhat later, though, in the auricular vein.*

Diagram I.

Daily oscillation in the number of the red blood corpuscles of normal dogs

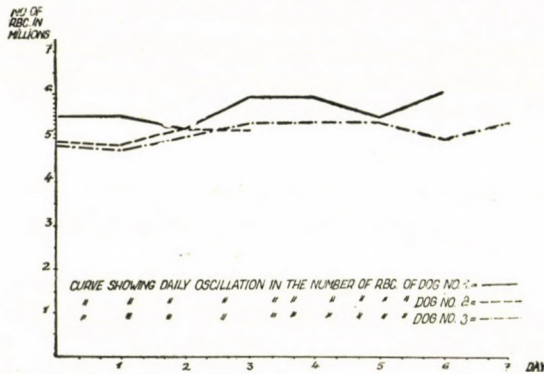
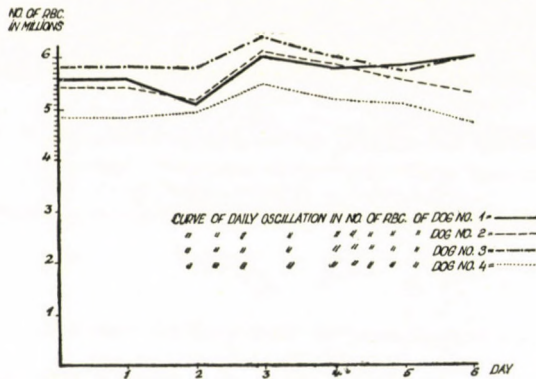


Diagram II.

Daily Oscillation in the No. of red blood corpuscles when an artery leading to a muscle is ligated



In the next group of experiments (Table I/a) we exposed the nutritive arteries on both extremities ; however, *the artery was ligated on one side only.* Then we determined the number of red blood corpuscles a) in the medullary vein corresponding to the ligated artery ; b) in the medullary vein of the unaffected side and c) in the auricular vein. We found that the *increase in the number of red blood corpuscles occurred almost at the same time in all three places.* Very often, this reaction set in already after 10 minutes ; sometimes, however, — especially in the peripheries — it was somewhat retarded. This

brought us to the conclusion that not only the outflowing stores of the medulla but other red blood corpuscles stores of the organism collaborated likewise in the development of erythrocytosis; consequently the latter was not of local, but of general origin.

The degree of change in the number of red blood corpuscles is indicated not only by figures but by a biological type of markings in the form of crosses. The table reveals that the increase reached in most of the cases the degree of 4 crosses which is equivalent to an increase of at least 800.000 red blood corpuscles per cubic mm.

Table II. charts a group of experiments in which the medullary artery was *definitely* ligated for the study of the duration of the erythrocytosis developed. Determination of the number of red blood corpuscles was performed on the blood samples taken from the corresponding medullary and the auricular veins, respectively.

Samples of blood were taken at intervals of $\frac{1}{2}$, 1 hour, and subsequently at intervals of 1—2 days. *Increase in the number of red blood corpuscles was observable already during the first hour, the peak value was generally reached after 24 hours and maintained for 3—4 days. Then it began to drop.* In Experiment No. 5. the presence of increase could be proved for an entire week.

Table III. charts experiments performed in a similar way: erythrocytosis was evoked by ligation of the artery. 8—10 days later, when the effects had already abated, the medullary artery *on the other thigh* of the same animal was ligated. Increase in the number of red blood corpuscles took place in the usual way, which proved that it was possible to evoke *this reaction twice on the same animal.*

Ad 2. In another group of experiments (*Table IV.*), local hypoxia was called forth by the injection of a 2% solution of *potassium cyanide* at 7.4 pH. 2 cc. were injected in each of two cases and 10 cc. in each of 5 cases into the medullary artery. The number of red blood corpuscles was checked in the corresponding medullary and the auricular vein, respectively, 5 and 10 minutes after the first, and 10 and 15 minutes after the second injection. With the exception of the first two animals which received insufficient quantities of potassium cyanide, *we observed in each case considerable erythrocytosis maintained for weeks.*

These results seem to indicate that medullary hypoxia evoked on a very small area causes a considerable general increase in the number of red blood corpuscles.

In order to enable this phenomenon to be interpreted in the proper light, we had to decide the question whether the erythrocytosis evoked was genuine or merely a blood concentration. Absence of increased plasma protein and leukocytosis in spite of the increase in the number of red blood corpuscles contradicts any supposition of haemoconcentration due to exsiccosis. The following arguments can be brought up against haemoconcentration due

to outflowing plasmal albumin (operative shock): 1. the operation itself — without ligation — does not cause any increase in the number of red blood corpuscles. 2. The number of white blood corpuscles does not change parallel with the increase in the number of red blood corpuscles; on the contrary, it often shifts in the opposite direction. 3. The fact that the blood samples used for control tests repeated after the passing of some days were taken from the auricular veins of perfectly healthy, non-anesthetized animals definitely refutes the theory of exsiccational haemoconcentration; the number of red blood corpuscles remained at the same high level.

Consequently, the erythrocytosis provoked was genuine and was the result of medullary hypoxia. Systematically performed determinations of the number of reticulocytes revealed the absence of reticulocytosis (the degree of increase was no more than 1—2‰), indicating that the increase in the number of red blood corpuscles was not the result of increased production of red blood corpuscles, but came from the outflowing contents of the red blood corpuscle stores. Examination of 11 histological slides prepared from the medulla revealed no morphologically recognizable disturbances in nutrition (as degeneration or necrosis). Portions of hypoxic medulla and healthy medulla were taken in 7 cases to control the cell-producing activity of the bone-marrow. Comparative analysis of the medullary areas showed no mentionable regenerative symptoms in the production of red blood corpuscles. Nevertheless, differentiation between early and later stages of medullary reaction necessitates further studies.

The mechanism of erythrocytosis activated by hypoxia suggested the hypothesis that *some kind of humoral substance might be released in the medulla, by which substance the ripe red blood corpuscles stored in the medulla, and in the blood-corpuscle storing places are mobilized and driven into the circulation.*

The theory of this humoral mechanism is supported by the data shown on *Table V*. In this group of experiments we ligated the medullary artery and the corresponding vein as well, to prevent any escape of this assumed substance into the general circulation. In 4 out of a total of 9 experiments no increase of red blood corpuscles was, in fact, observable, but in the other 5 cases venous ligation failed to prevent the development of erythrocytosis. This led us to the conclusion that the substance in question must have reached the blood stream through other blood vessels. (Extensive collateral circulation of this area makes this perfectly understandable, nor can the transporting role of the lymphatic circulation be entirely excluded either.)

Consequently, we performed another series of experiments, the results of which are registered on *Table VI*., in which not only the medullary vein corresponding to the ligated medullary artery, but all the other efferent veins which had to be taken into consideration were ligated. Erythrocytosis *did not set in*. Only in one of the cases was some increase observable after 3 days.

The data summarized in *Table VII* supplement these experiments. In the course of these no increase of the number of red blood corpuscles was observable after ligation of the medullary artery and of all efferent veins, while *ligation of the medullary artery of the other extremity with the veins left intact resulted in the customary appearance of erythrocytosis.*

The above results seem to indicate that erythrocytosis due to hypoxia of the bone-marrow is of *humoral origin*. Further evidence of this and details of its actual mechanism will be the subject of our next publication.

Literature

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Table I.
TEMPORARY LIGATION OF NUTRITIVE MEDULLARY ARTERY IN PHASES
(5'—10'—15'—30') IN ACUTE EXPERIMENT.

Serial No.	Place of puncture	Values of RBC at the beginning	No. of RBC after 5'	No. of RBC after 10'	No. of RBC after 15'	No. of RBC after 30'
I.	Peripheral (auricular) vein	5·70	6·08 +	6·50 ++++	6·98 ++++	5·90 ∅
	Medullary vein	6·08	++++	7·20 ++++	7·48 ++++	6·82 ++++
II.	Peripheral (auricular) vein	6·14	6·40 +	6·20 ∅	7·94 ++++	7·10 ++++
	Medullary vein	6·56	6·80 +	11·30 ++++	9·46 ++++	7·34 ++++
III.	Peripheral (auricular) vein	6·32	—	—	6·94 + +	7·02 + + +
	Medullary vein	5·82	—	—	6·86 ++++	7·70 ++++
IV.	Peripheral (auricular) vein	6·28	7·28 ++++	—	8·32 ++++	7·80 ++++
	Medullary vein	6·10	8·38 ++++	—	8·74 ++++	8·94 ++++
V.	Peripheral (auricular) vein	4·08	4·32 +	—	4·86 +++	5·26 ++++
	Medullary — vein	3·80	4·70 ++++	—	5·10 ++++	5·76 ++++

Marking of the number of RBC. =

0 — 200.000 ∅ | > 400.000 ++ | > 800.000 ++++
> 200.000 + | > 600.000 +++

Table I'a
LIGATION OF THE MEDULLARY ARTERY.
CHANGES IN THE NUMBER OF RED BLOOD CORPUSCLES IN THE VARIOUS
VASCULAR AREAS.

Serial No.	Place of puncture	Values of RBC at the beginning	No. of RBC 10' after ligation of art. nutr.	No. of RBC 20' after ligation of art. nutr.	No. of RBC 30' after ligation of art. nutr.	No. of RBC 40' after ligation of art. nutr.	No. of RBC 50' after ligation of art. nutr.	No. of RBC 60' after ligation of art. nutr.
I.	Peripheral (auricular) vein	5.02	5.98 ++++	5.20 ∅	6.20 ++++	6.08 ++++	5.40 ++	5.36 ++
	Right-side* medullary vein	3.80	4.68 ++++	4.16 +	4.42 +++	5.04 ++++	4.88 ++++	4.28 ++
	Left-side medullary vein	5.12	5.28 ∅	6.00 ++++	5.32 ∅	5.20 ∅	4.18 ∅	6.52 ++++
II.	Peripheral (auricular) vein	5.70	5.30 ∅	4.68 ∅	5.82 ∅	6.34 +++	5.68 ∅	5.78 ∅
	Right-side medullary vein	5.72	4.20 ∅	5.06 ∅	5.70 ∅	—	5.70 ∅	5.58 ∅
	Left-side* medullary vein	4.80	6.54 ++++	5.40 +++	4.82 ∅	6.60 ++++	—	6.12 ++++
III.	Peripheral (auricular) vein	4.74	5.26 ++	—	5.08 +	—	6.26 ++++	6.36 ++++
	Right-side medullary vein	4.56	5.74 ++++	—	5.48 ++++	5.52 ++++	6.08 ++++	6.14 ++++
	Left-side* medullary vein	5.06	6.00 ++++	—	5.86 +++	6.22 ++++	5.90 ++++	5.98 ++++
IV.	Peripheral (auricular) vein	6.28	7.28 ++++	8.32 ++++	7.80 ++++			
	Right-side* medullary vein	6.10	8.38 ++++	8.74 ++++	8.94 ++++			
	Left-side medullary vein	6.48	6.59	6.74 +	7.48 ++++			
V.	Peripheral (auricular) vein	4.08	4.32 +	4.86 +++	5.26 ++++			
	Right-side* medullary vein	3.80	4.76 ++++	5.10 ++++	5.76 ++++			
	Left-side medullary vein	4.10	4.60 ++	4.64 ++	5.10 ++++			

* Ligation of medullary artery was performed *only* on this side.

Table II.

DEFINITIVE LIGATION OF MEDULLARY NUTRITIVE ARTERY IN CHRONIC EXPERIMENT.

Serial No.	Place of puncture	Rbc. values at the beginning	Rbc. values after 30'	Rbc. values after 60'	Rbc. values after 3 h	Rbc. values after 24 h	Rbc. values after 48 h	Rbc. values after 72 h	Rbc. values after 96 h	Rbc. values after 120 h	Rbc. values at later dates
I.	Peripheral (auricular) vein	5.34	—	5.84 ++	—	—	—	—	—	—	—
	Medullary vein	4.82	6.50 ++++	6.23 ++++	—	—	—	—	—	—	—
II.	Peripheral (auricular) vein	5.32	—	—	—	7.90 ++++	—	7.12 ++++	—	—	—
	Medullary vein	7.60	6.16 ∅	6.64 ∅	8.04 ++++	10.28 ++++	8.40 ++++	—	—	—	—
III.	Peripheral (auricular) vein	—	—	—	—	—	—	—	—	—	—
	Medullary vein	4.02	—	—	—	4.86 +++	5.30 ++++	—	—	6.06 ++++	—
IV.	Peripheral (auricular) vein	4.90	—	5.80 ++++	—	5.90 ++++	5.90 ++++	6.04 ++++	6.00 ++++	—	—
	Medullary vein	5.28	—	5.60 +	—	5.70 +	5.70 +	—	—	5.70 +	—
V.	Peripheral (auricular) vein	4.48	6.90 ++++	—	—	5.90 ++++	—	—	5.85 ++++	—	After 144 h 5.38 ++++
	Medullary vein	—	—	—	—	—	—	—	—	—	—

VI.	Peripheral (auricular) vein	5.04	6.90 ++++	—	—	7.60 ++++	7.00 ++++	—	5.32 +	—	After 240 h 4.68 ∅
	Medullary vein	—	—	—	—	—	—	—	—	—	—
VII.	Peripheral (auricular) vein	5.30	—	7.30 ++++	—	7.50 ++++	7.10 ++++	—	5.24 ∅	—	After 240 h 4.92 ∅
	Medullary vein	—	—	—	—	—	—	—	—	—	—
VIII.	Peripheral (auricular) vein	4.90	5.54 ++	—	—	6.10 ++++	—	6.00 ++++	—	—	—
	Medullary vein	—	—	—	—	—	—	—	—	—	—
IX.	Peripheral (auricular) vein	4.82	—	5.36 ++	—	6.94 ++++	6.62 ++++	6.60 ++++	—	—	—
	Medullary vein	—	—	—	—	—	—	—	—	—	—
X.	Peripheral (auricular) vein	4.30	—	—	—	—	3.60 ∅	3.14 ∅	3.00 ∅	—	* After 288 h 2.78 ∅
	Medullary vein	4.16	—	—	—	—	—	—	3.28 ∅	—	—

Markings for the increase of RBC :

- 0—200.000 ∅
- > 200.000 +
- > 400.000 + +
- > 600.000 + + +
- > 800.000 + + + +

* Sepsis.

Table III.
I. UNILATERAL LIGATION OF MEDULLARY NUTRITIVE ARTERY II. LIGATION OF THE OPPOSITE ONE AFTER ABATEMENT OF ERYTHROCYTOSIS.

Serial No.	Place of punction	Rbc. values at beginning	Rbc. values after 30'	Rbc. values after 60'	Rbc. values after 24 h	Rbc. values after 48 h	Rbc. values after 72 h	Rbc. values after 96 h	Rbc. values after 120 h	Rbc. values after 144 h	Rbc. values after 168 h	Rbc. values at later dates	
1.	I. experiment	Peripheral (auricular) vein	3·36	—	—	3·30 ∅	4·30 ++++	4·20 ++++	4·10 +++	4·10 +++	—	—	—
		Medullary vein	3·26	—	—	3·70 ++	—	—	4·38 ∅	3·70 ++	—	—	—
	II. experiment	Peripheral (auricular) vein	4·10	—	—	4·14 ∅	—	4·40 +	4·10 ∅	—	4·32 +	4·32 +	—
		Medullary vein	3·70	—	—	—	—	4·40 ++++	—	—	—	4·40 ++++	—
2.	I. experiment	Peripheral (auricular) vein	5·70	6·40 +++	—	—	6·70 ++++	—	—	5·40 ∅	—	—	—
		Medullary vein	6·10	7·00 ++++	—	—	7·40 ++++	—	—	5·40 ∅	—	4·80 ∅	—
	II. experiment	Peripheral (auricular) vein	4·70	—	6·14 ++++	5·90 ++++	—	—	4·20 ∅	—	—	—	—
		Medullary vein	5·08	6·44 ++++	6·86 ++++	6·30 ++++	—	—	4·54 ∅	—	—	—	—
3.	I. experiment	Peripheral (auricular) vein	5·98	6·50 ++	6·94 ++++	—	6·46 ++	—	—	—	6·24 +	—	After 192 ^h 6·56 ++ After 244 ^h 6·6 ∅
		Medullary vein	5·80	7·30 ++++	7·06 ++++	—	6·36 ++	—	—	—	6·38 ++	—	6·86 ++++

3.	II. experiment	Peripheral (auricular) vein	6.08	—	7.00 ++++	7.00 ++++	—	—	—	5.98 ∅	4.70 ∅	—	—
		Medullary vein	6.12	6.62 ++	7.28 ++++	7.10 ++++	—	—	—	6.46 +	—	—	—
4.	I. experiment	Peripheral (auricular) vein	6.10	—	7.06 ++++	—	6.06 ∅	—	—	—	—	6.54 ++	After 244 ^h 5.16 ∅
		Medullary vein	6.04	6.88 +++	7.84 ++++	—	6.76 +++	—	—	—	—	—	—
	II. experiment	Peripheral (auricular) vein	4.96	7.36 ++++	—	6.78 ++++	7.08 ++++	—	—	—	—	—	—
		Medullary vein	5.16	6.94 ++++	—	—	—	—	—	—	—	—	—
5.	I. experiment	Peripheral (auricular) vein	5.30	—	6.10 +++	—	4.70 ∅	—	—	—	—	—	—
		Medullary vein	5.30	6.20 ++++	6.20 ++++	—	4.30 ∅	—	—	—	—	—	—
	II. experiment	Peripheral (auricular) vein	5.26	—	6.02 +++	—	—	—	—	—	—	—	—
		Medullary vein	5.54	6.11 ++	6.40 ++++	—	—	—	—	—	—	—	—

Markings for the increase in RBC :

0—200.000	∅
> 200.000	+
>> 400.000	++
>>> 600.000	+++
>>>> 800.000	++++

Experiment II. began after the effects of Experiment I. had subsided.

Table IV.
INJECTION OF POTASSIUM CYANIDE IN 2% SOLUTION INTO THE MEDULLARY ARTERY.

Serial No.	Place of puncture	Values of RBC at beginning (in millions)	I.				II.				Values obtained at later dates				
			Values 5' after injection of 10 cc of potassium cyanide in 2% sol.	Values 10' after injection of 10 cc of potassium cyanide in 2% sol.	Values 10' after renewed injection of 10 cc of pot. cyanide in 2% sol.	Values 15' after renewed injection of 10 cc of pot. cyanide in 2% sol.	Values 5' after injection of 10 cc of potassium cyanide in 2% sol.	Values 10' after injection of 10 cc of potassium cyanide in 2% sol.	Values 10' after renewed injection of 10 cc of pot. cyanide in 2% sol.	Values 15' after renewed injection of 10 cc of pot. cyanide in 2% sol.					
1.	Peripheral (auricular) vein	5.00	* —	4.70 ∅	* 5.20 ∅	—	28' 4.34 ∅	—	—	—	—	—	—	—	
	Medullary vein	4.80	—	3.80 ∅	—	4.04 ∅	20' 5.40 ++	40' 5.36 ++	—	—	—	—	—		
2.	Peripheral (auricular) vein	6.62	* 6.74 ∅	6.18 ∅	* —	—	23' 6.60 ∅	—	—	—	—	—	—		
	Medullary vein	6.44	5.18 ∅	6.00 ∅	—	—	29' 4.50 ∅	41' 6.00 ∅	—	—	—	—	—		
3.	Peripheral (auricular) vein	5.30	—	—	—	—	7.34 ++++	4 day 6.50 ++++	11 day 4.46 ∅	—	—	—	—		
	Medullary vein	6.08	7.38 ++++	7.80 ++++	—	—	—	—	—	—	—	—	—		
4.	Peripheral (auricular) vein	5.16	—	—	—	5.44 +	1 day exitus	—	—	—	—	—	—		
	Medullary vein	4.70	7.20 ++++	6.10 ++++	6.14 ++++	—	—	—	—	—	—	—	—		
5.	Peripheral (auricular) vein	5.92	—	7.26 ++++	—	—	40' 7.18 ++++	1 day 6.96 ++++	3 day 6.70 +++	13 day 6.76 +++	—	—	—		
	Medullary vein	5.72	—	7.86 ++++	—	—	20' 8.42 ++++	—	—	—	—	—	—		
6.	Peripheral (auricular) vein	4.84	—	—	5.20 +	—	1 day 6.16	2 day 5.11 +	19 day 5.12 +	23 day 5.86 ++++	31 day 5.84 ++++	—	—		
	Medullary vein	4.96	4.90 ∅	5.24 +	6.70 ++++	—	—	—	—	—	—	—	—		
7.	Peripheral (auricular) vein	4.82	—	—	—	7.36 ++++	2 day 7.32 ++++	5 day 5.42	12 day 5.78 ++++	16 day 5.02 ∅	24 day 5.02 ∅	—	—		
	Medullary vein	5.60	7.62 ++++	7.06 ++++	7.60 ++++	—	—	—	—	—	—	—	—		

Markings: 0—200 ∅, 200 < +, 400 < ++, 600 < +++, 800 < ++++.

* In experiment I. and II. we gave 2 cc of potassium cyanide in 2% sol. into the art. nutr.

Table V.

SIMULTANEOUS LIGATION OF MEDULLARY NUTRITIVE ARTERY AND EFFERENT VEIN.

Serial No.	Place of puncton	Rbc. values at the beginning	Rbc. values 30' after	Rbc. values 60' after	Rbc. values 24 h after	Rbc. values 48 h after	Rbc. values 72 h after	Rbc. values 96 h after	Rbc. values 120 h after	Rbc. values 168 h after	Rbc. values 192 h after	Rbc. values 216 h after	Rbc. values at later dates
I.	Peripheral (auricular) vein	4.34	5.14 +++	—	4.98 ++	4.94 ++	4.84 ++	—	—	—	—	—	—
II.	Peripheral (auricular) vein	* 4.30	4.10 ∅	—	4.30 ∅	3.90 ∅	4.40 ∅	—	—	—	—	—	—
III.	Peripheral (auricular) vein	5.10	7.32 ++++	7.56 ++++	—	7.02 ++++	—	6.60 ++++	—	5.26 ∅	—	5.96 ++++	336h 6.96 ++++ 528h 7.96 ++++
IV.	Peripheral (auricular) vein	4.42	4.28 ∅	4.92 ++	5.56 ++++	—	4.87 ++	—	—	—	4.80 ++	4.80 ++	—
V.	Peripheral (auricular) vein	* 5.56	6.02 ++	5.38 ∅	6.02 ++	—	5.58 ∅	—	—	6.06 ++	—	—	240h 5.26∅ 336h 5.25∅ 408h 5.90+ 504h 600h 5.03∅ 5.07∅
VI.	Peripheral (auricular) vein	* 5.98	5.72 ∅	5.98 ∅	5.24 ∅	—	6.38 +	—	5.96 ∅	—	6.74 +++	—	240h 6.52++ 288h 6.80+++ 360h 6.96h 5.14∅ 6.46++
VII.	Peripheral (auricular) vein	5.48	6.56 ++++	—	6.44 ++++	6.28 ++++	—	—	—	—	—	—	—
VIII.	Peripheral (auricular) vein	* 5.54	5.10 ∅	—	6.02 ++	5.44 ∅	—	—	—	—	—	—	—
IX.	Peripheral (auricular) vein	5.24	6.94 ++++	—	7.18 ++++	—	5.96 +++	—	—	—	—	—	—

Markings for the increase in RBC :

○—200.000 ∅ > 400.000 ++
 > 200.000 + > 600.000 +++
 > 800.000 +++++

Table VI.
SIMULTANEOUS LIGATION OF THE MEDULLARY ARTERY AND OF ALL EFFERENT VEINS.

Serial No.	Place of puncture	Values of RBC at the beginning	No. of RBC after 30'	No. of RBC after 24 h	No. of RBC after 48 h	No. of RBC after 72 h	No. of RBC after 120 h	No. of RBC after 144 h	No. of RBC after 168 h	No. of RBC after 192 h
I.	Peripheral (auricular) vein	6·86	6·48 ∅	6·56 ∅	6·50 ∅	6·60 ∅	—	6·80 ∅	—	6·70 ∅
II.	Peripheral (auricular) vein	5·40	5·16 ∅	5·30 ∅	5·74 +	5·92 ++	—	7·38 ++++	—	6·60 ++++
III.	Peripheral (auricular) vein	5·54	5·66 ∅	5·76 +	—	5·40 ∅	4·90 ∅	—	5·00 ∅	—
IV.	Peripheral (auricular) vein	6·16	5·82 ∅	6·26 ∅	—	6·48 +	5·90 ∅	6·10 ∅	—	—
V.	Peripheral (auricular) vein	5·18	5·38 ∅	5·56 +	—	5·30 ∅	5·10 ∅	—	—	—

Markings for the increase of RBC :

0—200.000	∅
> 200.000	+
> 400.000	++
> 600.000	+++
> 800.000	++++

Table VII.

LIGATION OF THE MEDULLARY ARTERY AND OF THE EFFERENT VEINS ON ONE SIDE (EXPERIMENT I.) AND LIGATION OF THE MEDULLARY ARTERY ON THE OTHER SIDE AT A LATER DATE.

Serial No.	Place of puncture	Values of RBC at beginning	No. of RBC after 60'	No. of RBC after 24 h	No. of RBC after 48 h	No. of RBC after 72 h	No. of RBC after 120 h	No. of RBC after 144 h	No. of RBC after 168 h	No. of RBC after 240 h
1.	I. experiment	Peripheral (auricular) vein 5.28	5.24 ∅	—	4.90 ∅	—	5.42 ∅	—	5.04 ∅	—
	II. experiment	Peripheral (auricular) vein 5.04	6.00 ++++	5.44 +	—	5.80 +++	—	6.36 ++++	—	5.30 +
2.	I. experiment	Peripheral (auricular) vein 4.98	4.58 ∅	—	4.60 ∅	—	—	5.84 ++++	5.40 +	—
	II. experiment	Peripheral (auricular) vein 5.40	7.00 ++++	7.20 ++++	—	5.24 ∅	—	5.32 ∅	—	5.64 +
3.	I. experiment	Peripheral (auricular) vein 7.32	6.88 ∅	—	7.80 ++	—	—	6.72 ∅	—	—
	II. experiment	Peripheral (auricular) vein 6.72	7.36 +++	6.70 ∅	—	7.24 ++	—	6.84 ∅	—	—

Markings for the increase in the number of RBC :

0—200.000	∅
> 200.000	+
> 400.000	++
> 600.000	+++
> 800.000	++++