

ALIMENTARY LEUCOCYTOSIS IN DOGS

M. Nikš, S. Cagán and I. Hulin

Department of Experimental Pathology and Pharmacology
(Head, Prof. G. Bardos) of the Medical Faculty of the Komenskii University, Bratislava

(Presented by Active Member AMN SSSR V. V. Parin)

Translated from *Byulleten' Éksperimental'noi Biologii i Meditsiny*, Vol. 51, No. 6, pp. 17-24, June, 1961

Original article submitted February 4, 1960

There are numerous references [1,2,4,6,8,10,12,13,15,17,18,21,22,23,25] to control by the brain of changes in the number of leucocytes in the circulating blood induced by food as the basic stimulus. The object of our investigation has been to study complex reflex alimentary leucocytosis, in which both unconditioned and conditioned reflex activity must be involved.

METHOD

We thought it most useful to study changes in the number of leucocytes and in the differential white cell count in an experiment on sham feeding. V. G. Chizhikov [19,20] in 1921-1926 and A. R. Makarova [9] in 1956 used the method to demonstrate the reflex influence of digestion on the blood system.

In three mongrels aged 1-2 years and weighing 14-18 kg, the daily variation in the number of leucocytes was determined while the stomach was empty. During the day (between 7 a.m. and 2 p.m.) the number of leucocytes varied in different animals between plus and minus 15%. For several months they were fed once per day between 8 and 9 a.m.

An esophagotomy was then performed. Under pentothal anesthesia, an incision was made along the mid-line of the neck. A tube was then introduced into the esophagus, which was then divided between the myohyoid muscles. A longitudinal incision was now made in the esophagus and a Belous cannula of perspex introduced into it. The wound was sewn up by layers. The fistula was arranged so that apart from the experiment the animal could take fluid normally, and sham feeding could be arranged by blocking the lead-out portion of the cannula.

After sewing up the wound, and when the normal leucocyte level had been restored in all the animals, a differential white cell count was made after the animals had fasted for 24 hours.

The first blood was collected at 7 a.m. and at four subsequent 15 minute intervals; then after one hour, a further five samples were collected at half-hour intervals. Thus, in all, ten samples were collected over a period of five hours, and always from the saphenous vein.

The blood count was mainly byurker chamber, and the cells were counted in 27 of the large squares. The differential white cell count was made in blood smears stained in Pappenheim's solution. The numbers were then given as a percentage [26].

On the following days, at 7 a.m., on each dog before feeding, sham feeding was given twice. It lasted for from 5 to 10 minutes, and the food consisted of minced meat, suet, and bread soaked in milk. During the next five hour period, the differential white count was repeated at the intervals described above.

RESULTS

In the dogs with an esophageal fistula tested while the stomach was empty, there was no change in the mean number of the leucocytes over the five hour period (the variation was between 94 and 106.7% of the original value. There was also no essential change in the differential white cell count (Table I and Fig. 1).

After sham feeding we could detect no increase in the number of leucocytes in any of the animals. When the experiment was repeated, the upper and lower limits of the count were found to be 3.8 and 8.2%, respectively (Table 2, and Fig. 1). There were no departures either in the differential white cell count from the values obtaining before feeding (see Table 2).

TABLE 1. The Number of Leucocytes and the Differential White Cell Count for Circulating Blood in Dogs Fitted with an Esophageal Fistula and Tested on an Empty Stomach (mean results of 3 studies)

Time blood was taken, min	Leucocytes per mm ³	Different cells, absolute units per mm ³ of blood					
		neutrophils		eosinophils	basophils	lymphocytes	monocytes
		lobed nuclei	rod-shaped nuclei				
0	10 483	7 862,3	765,2	41,9	0	1 048,3	765,3
15	9 917	6 386,5	694,2	29,8	29,8	1 914,0	862,7
30	11 183	7 190,7	816,4	78,3	190,1	1 711,0	1 196,5
45	9 850	6 835,9	689,5	29,5	0	1 477,5	817,7
60	10 917	7 314,4	687,8	76,4	76,4	1 965,1	796,9
90	10 317	6 943,3	897,6	72,2	30,9	1 475,3	897,7
120	10 483	6 845,4	880,6	0	31,4	1 991,8	733,8
180	10 150	6 394,5	629,2	30,5	30,5	2 233,0	832,3
240	9 850	5 782,0	591,0	98,5	29,5	2 492,1	856,9
300	11 023	6 503,6	992,1	77,2	33,0	1 907,0	1 510,1

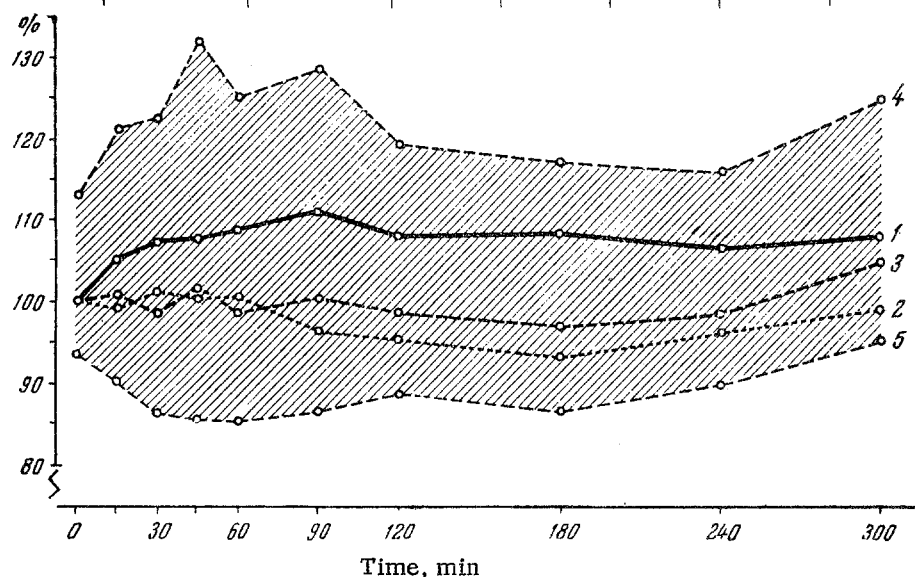


Fig. 1. Variation in the number of leucocytes in the circulating blood in dogs. 1) Mean number of leucocytes as percentage of the value before feeding (initial point of the curve) and during a 300-minute period after feeding; 2) the same before and after sham feeding; 3) changes in the number of leucocytes which were not fed during the experiment and not sham-fed; 4 and 5) mean deviations in the number of leucocytes in the above animals.

Next a further study was made on three dogs after they had been fed with 500 g of a powdered mixed food, in order to determine the extent of the leucocytic response. The maximum increase in the number of leucocytes was 12.5%, but there was no statistically significant increase (Table 3 and Fig. 1).

From Fig. 1 it can be seen that just as in the case of sham feeding, after genuine feeding there was no significant deviation of the quantities measured from those obtained when the stomach was empty. Changes in the differential white cell count do not differ essentially from those tested before feeding (see Table 3).

Statistical treatment of the results showed that there were no significant differences.

We were therefore unable to confirm the results reported in [9,20], where it was claimed that there was an alimentary leucocytosis following sham feeding in dogs.

The absence of any effect might be due to an unsuitable choice of animals. We therefore tested 12 other dogs aged from one to four years and weighing 9-23 kg. Tests were made after starving the animals for 24-48 hours, and at the same time intervals as in the previous experiments. Repeated tests were then made after feeding the

TABLE 2. Number of Leucocytes and Differential White Cell Count in the Circulating Blood of Dogs Fitted with an Esophageal Fistula Before and After Sham Feeding (mean results of 6 studies)

Time blood was taken, min	Leucocytes per mm ³	Different cells, absolute units per mm ³ of blood					
		neutrophils		eosinophils	basophils	lymphocytes	monocytes
		lobed nuclei	rod-shaped nuclei				
0	9 717	6 374,3	602,5	0	0	1 943,4	796,8
15	9 867	6 433,3	917,6	0	0	1 559,0	957,1
30	9 517	6 309,8	837,5	0	38,1	1 351,4	980,2
45	10 083	6 705,2	937,7	20,2	20,2	1 583,0	816,7
60	9 708	6 329,7	1 029,0	0	0	1 621,2	728,1
90	9 467	6 181,9	1 117,1	0	18,9	1 543,2	605,9
120	8 917	5 903,1	802,5	0	8,9	1 489,1	713,4
180	9 325	6 247,7	885,9	0	0	1 603,9	587,5
240	9 050	5 918,7	841,6	18,1	0	1 475,2	796,4
300	9 617	6 424,2	1 029,0	19,2	28,8	1 538,7	577,1

TABLE 3. Number of Leucocytes and Differential White Cell Count in the Circulating Blood of Dogs Fitted with an Esophageal Fistula Before and After Feeding (mean results of 7 studies)

Time blood was taken, min	Leucocytes per mm ³	Different cells, absolute units per mm ³ of blood					
		neutrophils		eosinophils	basophils	lymphocytes	monocytes
		lobed nuclei	rod-shaped nuclei				
0	9 993	6 365,5	689,5	30,0	10,0	2 158,5	739,5
15	10 928	6 120,0	896,0	43,7	10,9	2 939,5	917,9
30	10 571	6 554,0	909,2	52,8	0	2 071,9	983,1
45	10 528	6 874,8	852,8	31,6	0	2 063,5	705,3
60	11 093	7 332,5	809,8	11,1	11,1	1 930,2	998,3
90	10 907	6 882,3	730,8	65,4	32,7	2 290,5	905,3
120	11 243	7 071,8	798,3	33,7	0	2 282,3	1 056,9
180	10 186	6 590,3	611,2	61,1	0	2 027,0	896,4
240	10 986	6 888,2	802,0	76,9	0	2 230,2	988,7
300	10 790	6 743,7	766,1	10,8	0	2 298,3	971,1

animals with 500–1,000 g of a mixed food.

No effect was obtained in these animals, either on the number of white cells or on the differential count (Tables 4 and 5, Fig. 2).

The limits of variation in the number of leucocytes in dogs tested before they had taken food were + 2.4 and –7.7%, and in the experiments made on dogs after they had fed the limits were + 1.3 and –0.4%. From Fig. 2 it can be seen that these deviations lie within the limits for normal animals tested when the stomach was empty. In no single case was a statistically significant increase in the number of leucocytes found to occur after feeding.

According to some authors [2,3] the occurrence of an alimentary leucocytosis depends on the kind of food taken. In the last set of experiments, which were carried out on four mongrel dogs aged from one to three years and weighing 12–18 kg, we made blood tests before and after giving food consisting of protein (400 g of meat), fat (250 g of suet) and carbohydrate (250 g of glucose solution).

It can be seen from Figs. 3 and 4 that there was no statistically significant difference before and after feeding between either the numbers of leucocytes or the differential white cell counts.

There is disagreement in published reports on the time of appearance of the leucocytic reaction after feeding. Thus, A. R. Makarova [9] found an increase in the number of leucocytes to occur four hours after food had been taken.

TABLE 4. Number of Leucocytes and Differential White Cell Count in the Circulating Blood of Dogs Before and After Feeding (mean results of 22 studies)

Time blood was taken, min	Leucocytes per mm ³	Different cells, absolute units per mm ³ of blood					
		neutrophils		eosinophils	basophils	lymphocytes	monocytes
		lobed nuclei	rod-shaped nuclei				
0	13 886	8 692,6	791,5	69,4	27,8	3 068,8	1 235,9
15	13 943	8 700,4	822,6	125,5	41,8	3 025,7	1 227,0
30	14 060	9 082,7	815,5	253,1	42,2	2 460,5	1 406,0
45	13 829	8 988,8	885,0	152,2	41,5	2 586,0	1 175,5
60	14 043	8 931,4	940,9	126,4	28,1	2 822,6	1 193,6
90	13 857	8 937,9	859,1	180,1	13,8	2 674,4	1 191,7
120	13 891	8 876,3	833,5	125,0	27,8	2 792,1	1 236,3
180	14 064	8 818,1	857,9	84,4	14,1	2 897,2	1 392,3
240	13 961	8 809,4	949,3	83,8	27,9	2 931,8	1 158,8
300	13 848	8 765,9	872,4	138,4	27,7	2 811,1	1 232,5

TABLE 5. Number of Leucocytes and Differential White Cell Count in the Circulating Blood of Dogs Tested while the Stomach was Empty (mean results of 12 studies)

Time blood was taken, min	Leucocytes per mm ³	Different cell, absolute units per mm ³ of blood					
		neutrophils		eosinophils	basophils	lymphocytes	monocytes
		lobed nuclei	rod-shaped nuclei				
0	13 099	8 815,7	825,2	65,5	0,0	2 082,7	1 309,9
15	13 054	8 289,3	809,3	117,5	52,2	2 584,7	1 201,0
30	13 123	8 634,9	787,4	91,9	52,5	2 414,6	1 141,7
45	13 350	8 717,6	814,3	133,5	26,8	2 416,3	1 241,5
60	13 350	8 904,5	760,9	106,8	93,4	2 630,0	854,4
90	13 421	8 508,9	926,1	107,3	40,3	2 711,0	1 127,4
120	13 004	8 192,6	741,2	78,0	26,0	2 912,9	1 053,3
180	12 572	7 920,4	842,3	276,6	25,1	2 539,6	968,0
240	12 095	7 293,3	701,5	145,1	24,2	2 673,0	1 257,9
300	12 670	7 982,1	874,2	88,7	25,3	2 318,6	1 381,1

V. G. Chizhikov [19] found that after bread and meat had been given there were two increases in leucocyte numbers, the first occurring after 50 minutes and the second after four hours. G. S. Belen'kii [2] showed that after dogs had eaten meat or mixed food there were two phases of the leucocytic response: there was at first a leucopenia, and then a leucocytosis in which the number of leucocytes returned to the original value five hours after the food had been taken. According to S. D. Orlova [11], 30 minutes after food has been taken a leucopenia occurs, and after one hour the number of leucocytes is increased and reaches a maximum after 3-4 hours, and then returns to the original value. M. F. Sirotina [14] found that after feeding dogs with 200-250 g of meat there was a transitory leucopenia followed after two hours by a neutrophil leucocytosis. Similar results were obtained in rats after they had been given 1.5 g of bread. A. Ya. Yaroshevskii [24] fed cats with 100 g of meat and 150 ml of milk, and found that after 15 minutes there was either an increase or a decrease in the number of leucocytes. After 2-3 hours a leucocytosis occurred, which ceased after four hours.

The change occurring after 15 minutes was attributed to stimulation of the gastric interoceptors, because it was found to disappear after treating the gastric mucosa with cocaine. The leucocytosis occurring 2-3 hours after feeding was thought to result from the influence of humoral factors, because the response was maintained even after applying cocaine to the gastric mucosa.

In view of the published reports, we chose times at which blood was taken after feeding which would enable possible changes in the leucocytes to be observed. We were, however, unable to confirm the results obtained by numerous authors [17,18,23,25,etc.] on the alimentary leucocytosis in dogs. Our results agree with those of E. I.

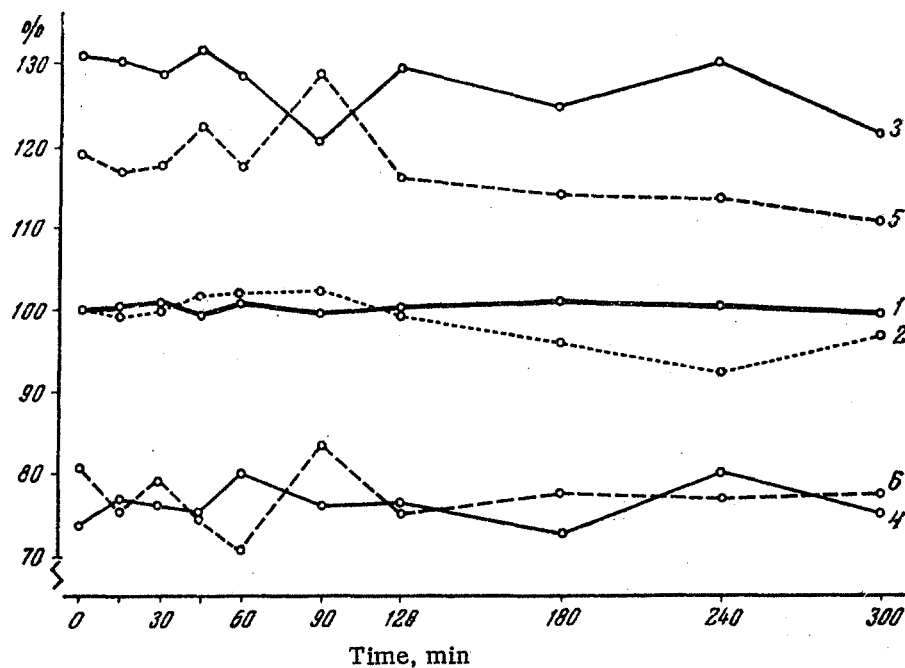


Fig. 2. Changes in the number of leucocytes in the circulating blood in dogs. 1) Number of leucocytes (mean results for 22 experiments) as a percentage of the original value, before and after feeding; 2) the same (mean results of 12 experiments), no food taken, and no sham feeding; 3,4) mean deviations in the experiments in which food was given; 5,6) corresponding deviations in the experiments made without giving food.

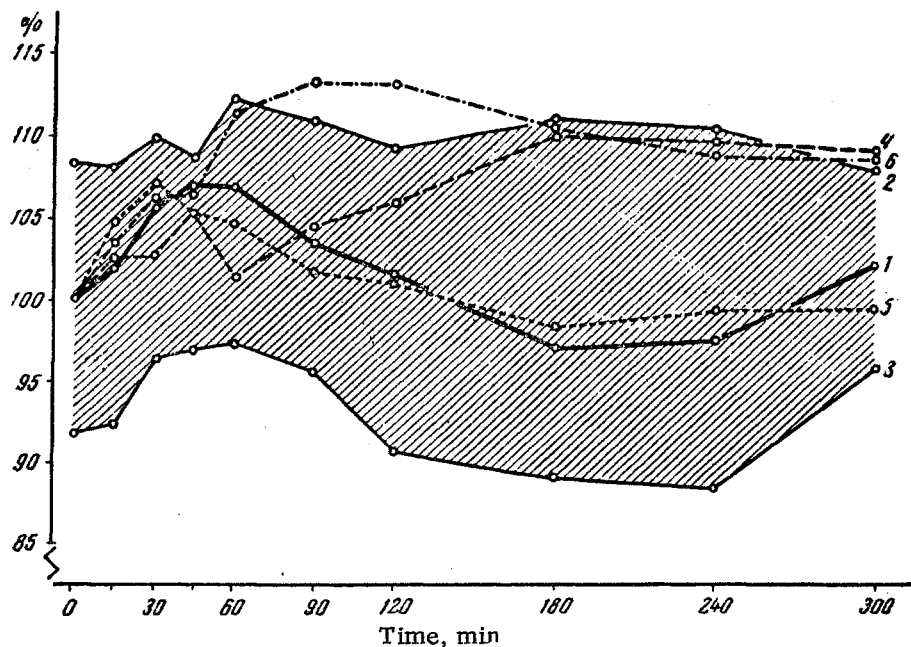


Fig. 3. Changes in the number of leucocytes in the circulating blood in dogs after receiving different kinds of food. 1) Number of leucocytes (as a percentage of the original value) in dogs not receiving food at the time of the experiment; 2,3) mean variations under the same conditions; 4) the same after feeding protein; 5) the same after feeding carbohydrate; 6) the same after feeding fat.

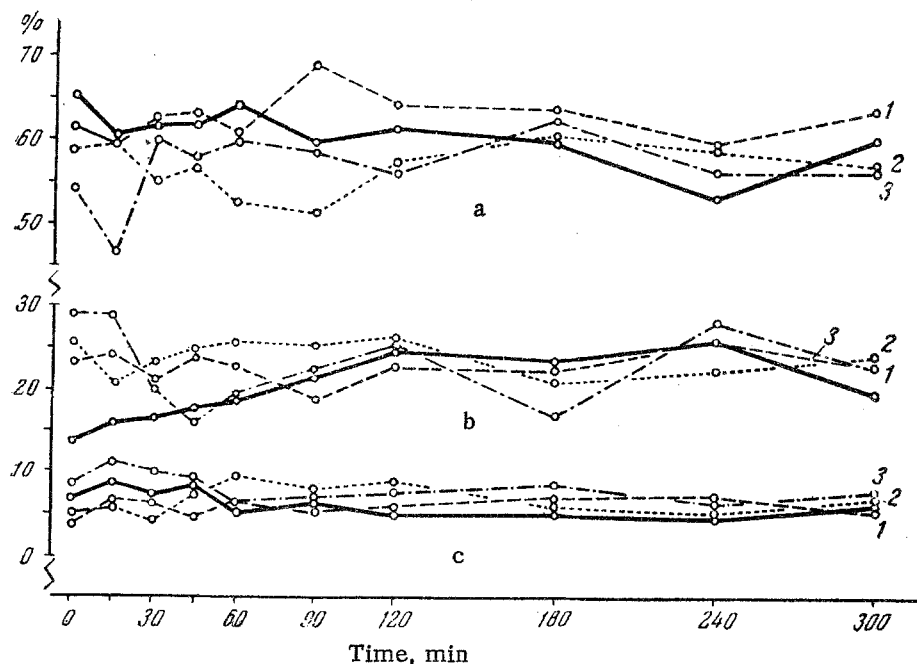


Fig. 4. Changes in the number of polymorphonuclear leukocytes (a), lymphocytes (b) and forms with rod-shaped nuclei (c) after consuming protein (1), fat (3) and carbohydrate (2). The solid line shows the number of the same kinds of cells present in the blood of dogs from whom food was withheld.

Freifel'd [16], who showed that no change in the white cells could be detected after the animals had been fed.

We consider that the difference between our results and those which had been published are due to experimental error: only small numbers of animals have been used, no statistical treatment of the results has been included and insufficient account has been taken of the variability in the number of leucocytes in the blood due to various internal and external factors.

SUMMARY

We were unable to confirm the assertions of numerous authors that an alimentary leucocytosis occurs in dogs. The reaction was studied in a sham feeding experiment.

In three dogs, after the sham feeding, there was no change in the white cell count. Repeated experiments on twelve dogs showed no significant difference between the leucocyte curves obtained before and after real feeding. The kind of food, whether protein, fat, or carbohydrate, had no influence on any change in the number of leucocytes or on the differential white cell count. The results were evaluated statistically.

LITERATURE CITED

1. Ts. I. Abakeliya and M. G. Gachechiladze, Ref. Zhurn. Biol., No. 4, 277 (1957).
2. G. S. Belen'kii, Klin. Med., No. 9, 52 (1950).
3. I. I. Il'in, Byull. Eksptl. Biol. i Med. 46, 7, 16 (1958).
4. S. K. Kiseleva, Klin. Med., No. 9, 43 (1951).
5. M. V. Krachkovskaya, Fiziol. Zhurn. SSSR 45, 5, 527 (1959).
6. M. V. Krachkovskaya, Zhurn. Vyssh. Nervn. Deyat. 9, 2, 205 (1959).
7. A. V. Kremer, Klin. Med., No. 9, 85 (1954).
8. I. T. Kurtsin, Arkh. Pat., No. 5, 3 (1952).
9. A. R. Makarova, Fiziol. Zhurn. SSSR 42, 2, 225 (1956).
10. I. I. Manukhin, Leucocytosis. Dissertation [in Russian] (St. Petersburg, 1911).
11. S. D. Orlova, Arkh. Biol. Nauk 44, 3, 123 (1936).
12. T. N. Ryantseva, Reports of the AN SSSR Institute of Higher Nervous Activity, Seriya Fiziol. [in Russian] (Moscow, 1956) No. 2, p. 254.
13. K. Ya. Sil'chenko, Zhurn. Vyssh. Nervn. Deyat, Issue 4, 482 (1954).
14. M. F. Sirotina, Fiziol. Zhurn. 3, 6, 77 (1957).

15. A. G. Urin, E. S. Zenkevich, Zhurn. Vyssh. Nervn. Deyat., Issue 5, 715 (1952).
16. E. I. Freifel'd, Hematology [in Russian] (Moscow, 1947).
17. V. N. Chernigovskii and A. Ya. Yaroshevskii, Problems of the Nervous Regulation of the Blood System [in Russian] (Moscow, 1953).
18. V. N. Chernigovskii and A. Ya. Yaroshevskii, Zhurn. Vyssh. Nervn. Deyat., Issue 1, 30 (1952).
19. V. G. Chizhikov, Russk. Fiziol. Zhurn., Nos. 5-6, 19 (1925).
20. V. G. Chizhikov, quoted by A. Ya. Yaroshevskii.
21. L. G. Shakleina, Byull. Éksptl. Biol. i Med. 39, 5, 27 (1955).
22. M. D. Shkol'nikova, Byull. Éksptl. Biol. i Med. 39, 1, 27 (1955).
23. S. V. Yaes, Vrach. Delo, No. 1, 87 (1956).
24. A. Ya. Yaroshevskii, Zhurn. Vyssh. Nervn. Deyat., Issue 5, 67 (1954).
25. A. Ya. Yaroshevskii, Fiziol. Zhurn. SSSR. 37, 175 (1951).
26. L. Dérer, Praktikum propedeutiky vnútorného lékařstva. (Bratislava, 1957).

All abbreviations of periodicals in the above bibliography are letter-by-letter transliterations of the abbreviations as given in the original Russian journal. *Some or all of this periodical literature may well be available in English translation.* A complete list of the cover-to-cover English translations appears at the back of this issue.
