TABLE I.
Survival Periods After Hemorrhage.

	No. surviving							Avg survival
Hours	0	1	2	3	4	5	6	hr
Depleted	14	14	14	9	2	0	0	3.3
Normal	10	10	10	7	6	0	0	3.7
3 wks horse meat	10	10	10	10	8	0	0	4.5
5 " " "	10	10	10	10	9	5	0	4.9

and the next group averaged 10% of the body weight. No other food was ingested. Analysis of the meat showed a content of about 20% protein (N × 6.25) and 4% fat. The average caloric intake was 120 per kilo per day.

Group 4 consisted of 10 animals given the same diet as in Group 3 for a period of 5 weeks before the experiment.

Experimental Findings. The data observed are described in Table I. It will be observed that the average survival time was but 3.3 hours in animals who had been previously on a non-protein regime, that the figure was 3.7 hours in the "normal" group, but that the animals on horse meat for 3 weeks survived 4.5 hours, whereas those on the same diet for 5 weeks survived 4.8 hours. That these averages are significant can be appreciated by consulting the number of animals actually surviving at each hour.

Comment. It seems clear from the evidence herein presented that dogs on a high protein diet consisting entirely of horse meat for 3 and 5 weeks are able to withstand significantly greater loss of blood than those not so prepared. While it is possible that the beneficial effect of the horse meat diet may be due to high protein content, some other factor or factors may have been responsible for the results. For example, experiments have been described[3] indicating the beneficial effect of thiamine in hemorrhage; however, horse meat has been reported as containing very little thiamine.[4] Regardless of the explanation, there would seem to be a practical inference from these experiments, *i.e.*, that the ingestion of a high protein diet of meat might be advisable as a prophylactic measure in patients in whom severe repeated blood loss may be expected.

Summary. Dogs given an *ad libitum* diet of horse meat for 3 and 5 weeks before repeated fatal hemorrhage are able to withstand a larger amount of blood loss than those on a "normal" or on a non-protein diet.

3 Govier, W. M., *J. Pharm. and Exp. Therap.*, 1943, **77**, 40.

4 Pyke, Magnus, *Biochem. J.*, 1940, **34**, 1341.

14656

Muscle Pain, Tendon Reflexes, and Muscular Coordination in Man.*

E. GELLHORN AND L. THOMPSON.

From the Laboratory of Neurophysiology, Department of Physiology, University of Minnesota.

The muscular action resulting from cortical innervation is greatly modified by afferent impulses. This is demonstrated by the inability of a monkey to use the hand after denervation of its posterior roots.[1] Moreover, distinct effects may be exerted on the cerebral cortex not only when the normal afferent impulses are absent, but also when they are increased as shown by observations of Uchtomsky.[2] It appeared, therefore, not un-

* Aided by a grant from the National Foundation for Infantile Paralysis, Inc.

1 Mott, F. W., and Sherrington, C. S., *Proc. Roy. Soc.*, 1895, **57**, 481.

2 *Cf.* Ufland, J. M., *Pflüger's Arch.*, 1925, **208**, 87.

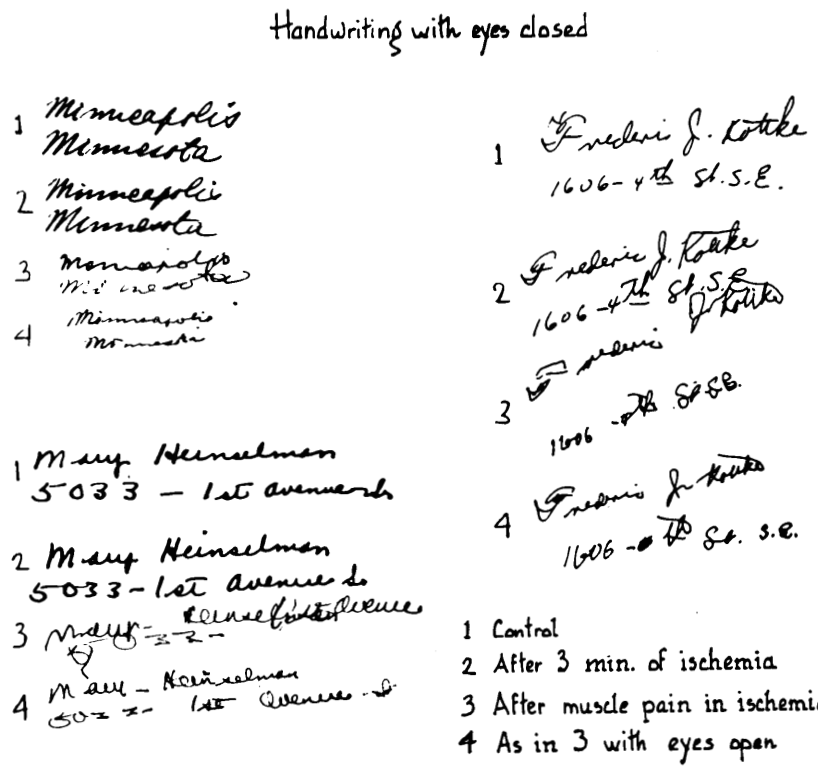

FIG. 1.

likely that pain originating in various peripheral structures may have a marked influence on the effects of central innervation. This problem was studied with respect to muscle pain which was produced by Lewis'[3] method of ischemic pain. Handwriting with closed eyes was used as an indicator of muscular coordination; in addition, the influence of muscle pain on tendon reflexes was studied.

Ischemia of the arm was produced by raising the pressure in a blood pressure cuff above the systolic blood pressure. Three minutes afterwards a writing test was performed in order to determine the effect of ischemia on muscular coordination. Then the fingers were alternately flexed and extended at a rate of 40 to 60 per minute for 40 to 60 seconds until the "end point" was reached. The muscle pain showed the diffuseness mentioned by Lewis. Its occurrence was indicated objectively by the appearance of a flushed face. While the pain persisted after the cessation of the movements the writing test was repeated with closed and also with open eyes. Then the pressure was released and the pain disappeared almost instantaneously.

Fig. 1 illustrates the effects on 3 subjects. It shows clearly that ischemia, at least within the time limits of our experiments, had no effect on handwriting. However, muscle pain initiated in the ischemic muscle by contractions lasting only 40 to 60 seconds causes a profound alteration in writing (*cf.* No. 3 in Fig. 1). These changes persist to a varying degree when the test is repeated with open eyes. The fact that there is an improvement in handwriting when the eyes are open distinguishes this form of disturbance of muscular coordination from that seen in anoxia[4] where the evaluation of optical sensations as well as of those originating in other sense organs is impaired.

As a further indication of disturbance in muscular coordination the finger-finger, and finger-nose tests were employed. It was found that the former was distinctly impaired during

[3] Lewis, T., *Pain*, N.Y., 1942.

[4] Gellhorn, E., *Am. J. Psychiat.*, 1939, **93**, 1, 413.

ischemic pain in 3 out of 6 tests, and the latter in 6 out of 8 tests. It is worthy of mention that in some instances repetition of the same test led to some improvement in spite of the persistence of pain.

Experiments to be reported elsewhere on action potentials in the human and on the effects of stimulation of the motor cortex under the influence of muscle pain give evidence that muscle pain produces changes in the reactivity of the motor cortex. It seems to be probable that these central changes are linked up with the disturbances in muscular coordination reported in this paper. Moreover, there is further proof of changes in the human central nervous system following muscular pain as shown by the following experiments.

The effect of muscle pain on the triceps reflex was studied in 10 persons in whom this reflex was very vivid. It was found that ischemia maintained for several minutes did not alter the reflex, nor had muscle pain, originating in the biceps through repeated contractions of this muscle during ischemia, any effect on the triceps reflex. However, when pain was elicited in the triceps by repeated contraction of the ischemic triceps, the triceps reflex disappeared completely even when stronger stimuli were applied to the tendon than under control conditions. These results were uniform in all 10 subjects. After restoration of the circulation the pain disappeared instantaneously and the triceps reflex returned within a few minutes. In some instances the reflex was fully restored after 30 seconds. In other cases it took one to 3 minutes. During this time a gradual increase in the responsiveness of the reflex was observed. This effect of muscle pain was absent in the contralateral triceps.

That the inhibition of tendon reflexes is due to pain and not to circulatory changes is indicated by the fact that ischemia lasting several minutes does not alter the triceps reflex. Moreover, the reflex remains inhibited immediately after restoration of circulation although the blood flow through the previously ischemic limb is greatly accelerated at this time (Lewis, Pickering, and Rothschild).[5]

[5] Lewis, T., Pickering, G. W., and Rothschild, P., *Heart*, 1931, **15**, 359.

The existence of nerve fibers originating in muscle which on stimulation lead to a reflex relaxation of this particular muscle was demonstrated by Sherrington.[6] These fibers may well be excited by pain and cause the tendon reflex to be absent since it is known that when the extensors are relaxed as under conditions involving reciprocal innervation tendon reflexes cannot be elicited. The fact that the painful triceps does not call forth reflex changes in other muscles such as the biceps is not surprising since stimulation of proprioceptive fibers produces a reflex only in the stimulated muscle and not in large groups of muscles which may be excited via cutaneous receptors. However, with increasing severity muscle pain may affect large areas of the motor cortex as will be shown elsewhere.

That muscle pain abolishes voluntary innervation is common experience; that it may lead to reflex inhibition was suggested by Sherrington.[6] However, no systematic studies seem to have been made previously on this subject. In view of the similarity of the symptoms produced by pain in various deep structures such as muscle, tendon, periosteum, etc. (Lewis)[3], a chance observation of Hess[7] that the muscle of a painful fractured leg failed to shiver on exposure to cold is of interest in that it likewise seems to demonstrate that pain originating in deep structures may inhibit the anterior horn cells. Inhibitory effects of pain on autonomic centers are well established. Ury and Gellhorn[8] showed that pupillary dilation following painful stimulation is due solely to an inhibition of the parasympathetic tone.

Summary. Muscle pain induced by 40 to 60 contractions of the ischemic muscle of the hand causes a severe disturbance in the coordination of voluntary movements as shown by the handwriting and finger-nose tests. That reflex movements are likewise altered is shown by the fact that the triceps reflex disappears regularly when ischemic pain is produced in this muscle. However, neither

[6] Sherrington, C. S., *Nature*, 1924, **113**, 929.

[7] Hess, W. R., *Pflüger's Arch. Ges. Physiol.*, 1924, **203**, 539.

[8] Ury, B., and Gellhorn, E., *J. Neurophysiol.*, 1939, **2**, 268.

ischemia nor ischemic pain involving the biceps has any influence on the triceps reflex. It is concluded that muscle pain interferes with muscular coordination through influences on spinal and possibly cortical centers.

14657

Pharmacological Observations on Crystalline Sodium Penicillin.

H. B. VAN DYKE.

From the Division of Pharmacology, the Squibb Institute for Medical Research, New Brunswick, N.J.

Through the kindness of Dr. O. Wintersteiner, of the Division of Organic Chemistry of this Institute, a small quantity of twice-crystallized Na Penicillin-G was made available for pharmacological experiments which had to be restricted owing to the great scarcity of this pure antibiotic. Its potency was 1650 Oxford units per mg.

Results. Hemolysis of washed rabbit erythrocytes suspended in 0.15 M NaCl or in 0.15 M Na penicillin was compared at 37° after the addition of one part of erythrocyte-suspension in isotonic saline equivalent to original blood to 10 parts of a solution of NaCl or of Na penicillin. No hemolysis occurred during 3 hours. After 17 hours, the erythrocytes suspended in Na penicillin solution had become converted into a chocolate-colored granular mass not easily homogenized; the suspending fluid was water-clear. Microscopically the brown material appeared to consist of granules about 0.5 micron in diameter; a few irregular "ghosts" could be found. No further change occurred during a total period of 48 hours after which only 4% of the original antibiotic activity remained in the supernatant fluid.

Dr. R. J. Dubos suggested that possibly adsorption of penicillin on the rabbit erythrocytes had occurred. In another experiment, the suspending fluid contained 1 mg (1650 units) of Na penicillin per ml together with sufficient NaCl to make the solution equivalent to 0.15 M NaCl. Erythrocyte suspensions with and without penicillin as well as Na penicillin-NaCl solutions without erythrocytes were incubated at 37° for 2 hours in duplicate. There were suitable control tubes from which erythrocyte suspension in saline or the supernatant fluid of such a suspension was added to incubated Na penicillin solution at the time of the antibiotic tests. No hemolysis occurred. Within the limits of error (± 20%) of the determination of antibiotic activity, which was kindly estimated by Miss C. M. McKee, there was no loss of activity in any tube including those from which the erythrocytes had been removed by centrifugation at the end of the period of incubation. It appears unlikely that important amounts of penicillin are adsorbed by rabbit erythrocytes suspended in isotonic saline solution.

Dr. Eric G. Ball, of Harvard Medical School, performed a careful experiment to determine whether yeast cells, duck erythrocytes, or rat liver slices consume oxygen at an altered rate if 3400 units (2.05 mg) of Na penicillin replace an osmotically equivalent amount of NaCl in each ml of Locke's solution containing phosphate buffer at pH 7.33 and glucose as the substrate. All observations were made at 37° C. The solution of Na penicillin was added to the suspensions after a control period of 20 minutes. The oxygen-consumption of other control suspensions was followed throughout the experiment. The total period of observation was 214 minutes. The Na penicillin caused no change in any of the 3 tissues with regard to the rate of oxygen-consumption.

The results of representative tests with a few isolated organs are illustrated in Fig. 1-3. A detailed description of the individual trac-