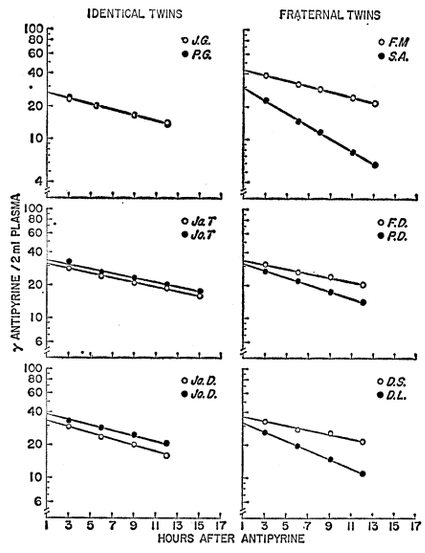

Fig. 1. Decline of antipyrine in the plasma of three sets of identical twins (left) and three sets of fraternal twins (right). The log of the antipyrine concentration in 2 ml of plasma is shown at intervals after a single oral dose (18 mg/kg).

Table 1. Half-life of antipyrine in plasma. The difference between monozygotic and dizygotic twins in intrapair variance is significant: $P < .001$ ($F = 64.5$, $N_1 = N_2 = 9$).

Twin	Age, sex	Half-life (hr)
	Identical twins	
JG	22,M	11.5
PG	22,M	11.5
DH	26,F	11.0
DW	26,F	11.0
JaD	29,M	11.0
JoD	29,M	12.0
SD	34,F	11.1
ML	34,F	11.5
DT	43,F	10.3
UW	43,F	9.6
JaT	44,M	14.9
JoT	44,M	14.9
GeL	45,M	12.3
GuL	45,M	12.8
HeM	48,M	11.3
HoM	48,M	11.3
CJ	56,F	6.9
FJ	56,F	7.1
	Fraternal twins	
AM	21,F	15.1
SM	21,M	6.3
LD	21,F	8.2
LW	21,F	6.9
JaH	24,F	12.0
JeH	24,F	6.0
EK	31,F	7.7
RK	31,M	7.3
SA	33,F	5.1
FM	33,F	12.5
DL	36,F	7.2
DS	36,F	15.0
NM	39,F	16.7
JR	39,F	13.4
LL	44,F	12.0
LR	44,F	16.7
FD	48,M	14.7
PD	48,M	9.3

Each twin received at 0830 hours a single oral dose of antipyrine (18 mg/kg); at intervals of 3, 6, 9, and 12 hours thereafter blood samples were drawn in tubes containing oxalate, and the plasma was assayed for antipyrine by the method of Brodie *et al.* (*7*). After 12 hours the levels in blood in certain twins were so low that measurement was inaccurate.

Figure 1 shows the decay of antipyrine in plasma from three sets of identical and three sets of fraternal twins. Table 1 gives the half-lives determined from such curves for each subject; the lines were fitted by eye. Threefold variations in antipyrine half-life occurred in these 36 subjects; the range was from 16.7 to 5.1 hours (Table 1). Antipyrine half-life appears to be a stable trait; in two individuals given a second dose 6 weeks after the first, the half-lives were unchanged.

The contribution of heredity was calculated from a formula in which the difference between the variance within pairs of fraternal twins and the variance within pairs of identical twins is divided by the variance within pairs of fraternal twins (*8*). The value of 0.98 indicates that control of antipyrine variability in normal humans not receiving other drugs depends on hereditary rather than environmental factors; the formula permits a range of values from 0 (suggesting negligible contribution by heredity) to 1 (indicating strong hereditary influence). Variance within pairs was calculated from a formula in which the sum of the squares of the difference between twins is divided by twice the number of pairs of twins (*8*).

The intrapair correlation coefficient was 1.0 for identical and 0 for fraternal twins. Differences between fraternal twins in the half-life of antipyrine varied from 0.4 hours in E.K. and R.K. (values in the range of those observed in identical twins) to 8.8 hours in A.M. and S.M. This result probably depends on the degree of genetic similarity between the parents of each pair of twins.

The antipyrine half-life in the 36 subjects was 10.9 ± 0.5 hours (mean $\pm$ S.E.); a value of 10 hours appears in the literature (*3, 4*). The half-lives in the 22 female and 14 male subjects were 10.6 ± 0.7 and 11.5 ± 0.7 hours (means $\pm$ S.E.), respectively. Similarity of half-lives for males and females makes unlikely any strong contribution by Y-linked genes to the control of antipyrine half-life.

For the 28 individuals receiving both antipyrine and phenylbutazone, the possibility of correlation in their rates of metabolism was studied, particularly in view of the drugs' close resemblance in structure. No correlation between half-lives of antipyrine and phenylbutazone in an individual was found by Bartlett's method (*9*). This lack of correlation suggests that antipyrine and phenylbutazone are metabolized by two functionally distinct enzymes, a conclusion supported by differences between the drugs in the chemical sites at which hydroxylation occurs (*1, 4*). Knowledge of an individual's genetically dependent half-life might be utilized to improve the therapeutic effectiveness and to avoid the toxic side effects of many drugs.

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Retrograde Amnesia Produced by Hippocampal Spreading Depression

Abstract. *Injection of potassium chloride into the hippocampus produces a disruption of electrical activity; a concomitant of this disruption is a deficit in retention of conditioned suppression learned 24 hours before injection.*

A marked decrease in the amplitude of the electrical activity of the hippocampus is seen after injection of puromycin (*1*). Puromycin is a potent inhibitor of protein synthesis and produces retrograde amnesia (*2*). The

demonstrated effect on messenger RNA and thereby on protein synthesis has been proposed as an explanation of this amnesia (*3*).

An alternate account of the retention deficit could be that the electrical abnormality caused by puromycin is sufficient to produce amnesia. Thus, altering electrical activity by other means might also produce a retention deficit. The experiment we report here bears on this possibility.

Twenty male albino rats of the Sprague-Dawley strain were used as subjects. The measure of retention was the conditioned emotional response (*4*). In this procedure a tone was presented while the animal was drinking from a water-filled tube. The tone had previously been paired with shock. The degree of suppression of drinking (measured in seconds) during the tone was taken as a measure of conditioned suppression.

Disruption of hippocampal electrical activity was produced by injection of KCl. When KCl is injected into the hippocampus, it results in spreading depression. One consequence of spreading depression is a marked decrease in the amplitude of the electrical activity. The depression evidently spreads throughout the hippocampus without affecting the cortex (*5*).

On day 1 the animals, who had been deprived of water for 24 hours, were placed in a response chamber with a drinking tube at one end. When the animal drank from the tube, it closed a low ampere contact relay. All animals were allowed 110 licks. Four days later, the animals were given four tone-shock pairings in the same apparatus but with the drinking tube removed. The tone was of 15 seconds duration, and its termination was coincident with the delivery of a 1-second, 2-ma shock delivered to the animal's feet through the grid floor of the chamber. On day 6 the animals were anesthetized first with chlorprothixene (10.0 mg per kilogram of body weight) and then with pentobarbital (40.0 mg/kg). All injections were intraperitoneal.

Stainless steel 23-gauge hypodermic needle tubing, insulated except for 1 mm at the tip, was used for both recording and injection. The placements were bilateral: 5 mm posterior to bregma, 5 mm lateral to the midline suture, and 5 mm below the dorsal surface of the skull (*6*). The electrodes were held in place by cranioplastic cement anchored to stainless steel jewelers' screws drilled into the skull. A copper wire was wrapped around one of the screws and served as ground. Electroencephalographic recordings were made with a Beckman type RB dynograph by connecting the upper portion of the implanted tube to Microdot cable with gold Amphenol connectors. All recordings were made while the animal was surgically anesthetized. Monopolar leads were used at all times.

Table 1. Retention scores (suppression of drinking) and EEG depression.

	Retention score (sec)	EEG depression (sec)
	Saline control	
	270.9	0
	11.7	0
	300.0	0
	300.0	0
	163.4	0
	300.0	0
	300.0	0
	300.0	0
	300.0	0
	300.0	0
Median	= 300.0	= 0
	KCl injected, group A	
	42.3	600.0
	14.3	405.0
	2.4	1049.0
	70.2	575.0
	17.6	1746.0
	2.2	437.0
	62.9	718.0
Median	= 17.6	= 600.0
	KCl injected, group B	
	300.0	200.0
	300.0	120.0
	300.0	217.0
Median	= 300.0	= 200.0
A + B median	= 52.6	= 524.0

Direct injections into the hippocampus were made by inserting a 30-gauge tube through the electrode. This tube was inserted into one side of the hippocampus and an injection was made; the tube was then removed and a contralateral injection was given. For ten of the animals, each injection was 5 to 12 μl of KCl (25 percent solution); comparable volumes of physiological saline were given to the other ten animals.

The electrical activity of the hippocampus was monitored before and after the bilateral injections. In the group injected with KCl, monitoring was continued until the record approximated the preinjection activity. The activity of the group injected with saline was monitored for 15 minutes after injection. Injections of saline had no effect on the electrical activity of the hippocampus. In all cases monitoring began 1 minute after injection.

In all groups, recordings were made from one hemisphere at a time. By switching the leads several times during recording, we determined that similar changes in activity resulted from the bilateral injections. A sample set of EEG records is given in Fig. 1.

The animals were tested for retention 4 days after the operation. The subjects, who had been deprived of water for 24 hours, were placed in the apparatus and permitted to make 100 licks. The tone was then presented and the time it took the animal to make ten additional licks was recorded. If the animals did not respond after 300 seconds, the trial was arbitrarily terminated. No shocks were delivered in the test session.

Seven of the controls reached the 300-second criterion, and only one failed to show substantial retention. In contrast, only three of the ten animals injected with KCl showed retention. The intergroup differences in the time required to make ten licks were significant ($P<.02$ by two-tailed Mann Whitney U test). The data are presented in Table 1 (*7*).

The duration of maximum EEG de-

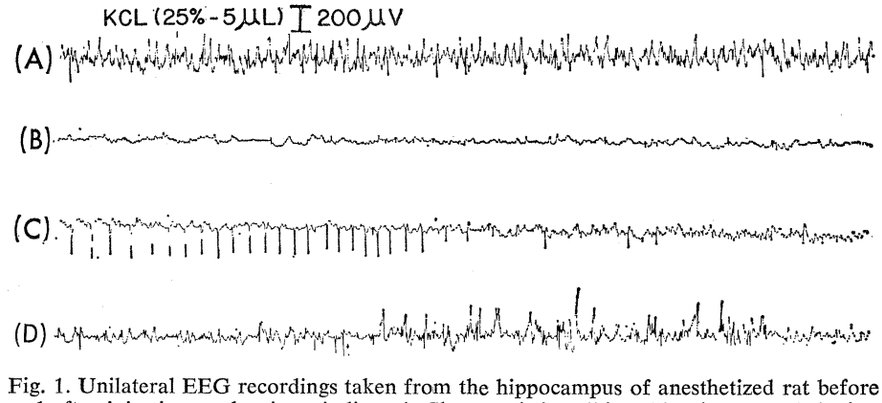

Fig. 1. Unilateral EEG recordings taken from the hippocampus of anesthetized rat before and after injection at the times indicated. Characteristic spiking, like that recorded after 30 minutes, was seen sporadically and infrequently during the period of depression. (*A*) Before injection; (*B*) 15 minutes after injection; (*C*) 30 minutes after injection; (*D*) 60 minutes after injection.

pression differed in the KCl group. When this was estimated from the records, it was found that the three subjects that did show retention (group B in Table 1) showed a shorter duration of maximum EEG depression (*8*) than those that did not show retention (group A in Table 1). This difference in depression was a reliable one ($P<.016$; two-tailed Mann Whitney U test); thus duration of EEG depression was inversely related to retention.

As a test for the possibility that the injections had produced hippocampal lesions, three of the group that had been injected with KCl and that showed a retention deficit were retrained in the apparatus. All three subsequently showed retention, indicating that neither performance nor the ability to retain was permanently impaired by the injection (*9*).

After being tested, the animals were perfused with formalin, and the brains were removed. After several days, the frozen brains were sectioned (50-μ sections) and stained to verify electrode placements. All electrodes were located in the ventral posterior hippocampus. Some of the animals that had been injected with KCl sustained small, uniformly unilateral lesions. In fact, one of the animals with the most extensive damage was a saline control. Furthermore, the animals injected with KCl could not be differentiated from those injected with saline on the basis of extent of lesion. Finally, the animal that had sustained the largest lesion was later retrainable. Thus, even the largest histologically estimated lesion was insufficient to interfere with retraining or retesting.

The results indicate that temporary interruption of hippocampal electrical activity as much as 24 hours after learning can produce retrograde amnesia. This finding is unusual in that it has been generally acknowledged that electrical activity need only occur for a few minutes after learning, after which permanent storage is assumed to have taken place. Our findings indicate that, at least in some areas of the brain, and with our particular experimental technique, electrical activity must continue for at least 24 hours. Somewhat similar results have also been found with bilateral hippocampal lesions and with deafferentation (*10*).

It has been reported that potassium chloride can inhibit protein synthesis (*11*), but neither the percentage nor duration of inhibition approaches that which puromycin must evidently produce in order for amnesia to occur (*12*). Although procedural differences may account for the differences in degree of inhibition, there is at least the suggestion that the deficit seen with KCl is not mediated by an inhibition of protein synthesis.

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7. There were no differences between the group injected with saline and that injected with KCl in the first 100 and the subsequent ten licks in the training session as evaluated by the U test. For the saline animals the medians were 53.8 for the first 100 and 4.6 seconds for the subsequent ten; the corresponding medians for the animals injected with KCl were 98.8 and 4.8. The times for the first 100 licks between training and test were, however, significantly different for the saline group but not the KCl group ($P < .05$; Wilcoxen matched-pairs test). Similarly, the KCl and saline groups differed in time to 100 licks during the test session ($P < .05$; U test). The medians were 64.6 seconds for KCl and 190.5 seconds for saline.
8. Maximum depression is defined as activity showing an amplitude of less than 10 μv.
9. As an added check on the possibility that KCl produces damage and thus a deficit in suppression, other animals were first given bilateral injections of KCl, were trained 3 days later, and then were tested 1 day after training. The mean duration of suppression in tone was 232.0 seconds (see the saline controls in Table 1). The water intake was also recorded in these rats; no effect on intake was noted.
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27 May 1968

Premature Citations of Zoological Nomina

While agreeing with Sohn (*1*) that premature citations of zoological nomina (technical names of animal taxa) may be undesirable, I believe that the argument should be modified in two respects.

Sohn gives as one example the publication by Egorov in 1953 of a previously unpublished specific nomen as *Mossolovella incognita* (Glebovskaja and Zaspelova in litt.) and states that the taxon (that is, its nomen) should be cited as *M. incognita* Egorov, 1953. In my opinion, that is probably incorrect under the present Code (*2*). Egorov's wording suggests that "some other person (or persons) is alone responsible both for the name and the conditions that make it available" (*2*, Article 50). If that is true, the name should be cited as *M. incognita* (Glebovskaja and Zaspelova in Egorov, 1953) [*2*, Article 51(c)]. The expression "the conditions that make it available" is not explained in the Code. In this particular example, it can be debated whether Glebovskaja and Zaspelova were in fact alone responsible for such conditions, although from Sohn's statement it would appear that they are. I am not particularly concerned with possible disagreement about the specific example but with the fact that the Code does provide for crediting nomina and their definitions to others than the authors of the paper in which they first appear.

Just this point is not covered by the inadequate and nonmandatory published Code of Ethics (*2*, Appendix A), but it is obviously simple good manners to seek permission from the authors, if possible, before publishing their nomen. I can see no cause for confusion in this practice, and nothing objectionable if done with permission.

A second point, not covered by Sohn except by implication, is that publication of *nomina nuda* may be justified and even desirable under special circumstances. If a nomen and its definition by author A are known by B to be in press or in a manuscript assured of publication and if the corresponding taxon is involved in publication by B, in my opinion it is desirable that B publish the nomen with or without quotation of A's definition and in either case with ascription to A, "in litt." or "in press." If published by B with definition quoted from A, under the Code the nomen dates from B's publication but is correctly ascribed to A, not to B. If published by B without definition, the nomen is a *nomen nudum* but will assuredly cease to be so when published by A and will take the authorship and date of the latter publication. In either case the convenience and accuracy of B's work are promoted, because the eventual nomen of a taxon discussed is given, without such ambiguity as, for example, writing about "a species later