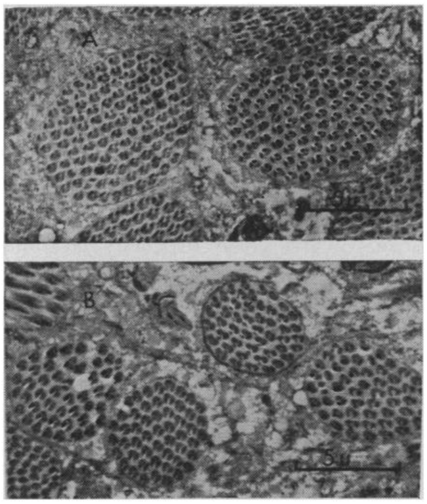

Fig. 1. (A) Electron micrograph of a section of a whole testis from a normal *D. pseudoobscura* male, showing cross sections of sperm bundles. (B) A section from a "sex ratio" male.

chromosome produced progeny consisting almost entirely of females because only X-bearing sperm function in fertilization; it was also reported that the full complement of 128 spermatids (*2*, *4*) and sperm (*4*) per bundle was present. In addition, it was reported that the X chromosome divides twice at the first meiotic division (*2*), thus giving rise to the extra X-bearing sperm, but Novitski, Peacock, and Engle (*4*) were unable to find evidence of any abnormality of the X chromosome at meiosis. They explained the phenomenon of "sex ratio" on the basis of the regular nonfunctioning of meiotic products, as proposed for *D. melanogaster* (*5*). In the case of the "sex ratio," according to this hypothesis, the X chromosome is favored and the unisexual progeny can be explained by "preferential movement of the X chromosome to the functional pole at anaphase I" (*4*). We have been able to confirm the earlier reports that over 100 spermatids per bundle are present in "sex ratio" males (*2*, *4*), but we find only about half that number of mature sperm per bundle. As a result of these findings, we suggest that the action of "sex ratio" cannot be explained on the basis of regular nonfunctioning of meiotic products, but rather should be described as spermiogenic failure.

For this study we used descendants of the flies collected by Novitski and Sturtevant (*4*), as well as male progeny of wild females recently collected near Tucson, Arizona. Whole testes of day-old males were fixed in 2 percent osmium tetroxide, embedded in Epon, and sectioned. Smear preparations of testes were also made for combined light and electron micrography (*6*) and fixed as above. Photographs of the smear preparations were taken with the light microscope perpendicular to the plane of sectioning both before and during the sectioning procedure. This permitted identification of the sections with respect to their position on the bundle as well as with respect to the size of the bundle. Some of the sections were photographed on a Philips EM-300 electron microscope and some on a Hitachi HS-7S.

The mean number of sperm per bundle as shown by electron micrographs of sectioned testes was 112 ± 7.4 (nine bundles) in normal males and 55 ± 8.2 (38 bundles) in "sex ratio" males (Fig. 1).

Three different sizes of sperm bundles have been reported as a regular feature of spermatogenesis in *D. pseudoobscura* (*7*). There was no difference in number of sperm per bundle in the three sizes. When using squashes stained with aceto-orcein and Feulgen stains we experienced great difficulty in counting mature sperm, and we feel that these light-microscopic methods are inadequate for the counting of sperm bundles in *D. pseudoobscura*.

Since "sex ratio" males have only half as many sperm per bundle as normal males, it seems reasonable to conclude that the action of "sex ratio" does not depend on a functional pole. The problem arises of how "sex ratio" maintains a frequency as high as 30 percent in some populations (*2*) if "sex ratio" males produce only half as many sperm as normal. There is also laboratory evidence that "sex ratio" is selected against (*3*), which compounds the problem.

The population dynamics of "sex ratio" in nature is not understood, but, if females are not limited in their fecundity by the amount of sperm they receive, then "sex ratio" is probably a good example of a chromosome meiotically driven to a high frequency against adverse selection (*8*).

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Prolonged Learning and Split-Brain Cats

Abstract. *A definite relation is shown between interhemispheric transfer and speed of learning. Split-brain cats were trained in a variety of discriminations, and the success or failure of interhemispheric transfer was evaluated with regard to rate of learning. When interhemispheric transfer succeeds, learning time is normal; when interhemispheric transfer fails, learning time is prolonged, becoming at least twice that of normal. Retention is also poor in those animals that exhibit prolonged learning. A normal learning curve appears to be the product of interaction between both cerebral hemispheres during learning.*

Effects of hemispheric division on learning and memory are relatively unknown. Past experiments with "split-brain" cats and monkeys (*1*) and with cerebral deconnection in man (*2*) have helped to increase our understanding of the function of the corpus callosum and other midline structures in their role of transmitting sensory information from one hemisphere to the other. Little attention, however, has been given to these same studies with regard to the contribution of each cerebral hemisphere to memory and the learning process.

I now report experiments in which interhemispheric transfer is compared with the speed of learning in normal animals and in split-brain animals prepared with a variety of commissurotomies as well as with cortical ablation. Retention is also compared between the two groups. Some of these investigations have already been reported, but they deal mainly with the success or failure of interhemispheric transfer of learning (*3*). The data demonstrate that, when only one hemisphere participates during learning, the rate of learning is prolonged and becomes two to three times that of normal. Retention is markedly decreased.

Of 84 cats studied, 35 were normal (controls), and 49 were split-brain cats prepared before any test procedures. Surgery included midsagittal section of the optic chiasm and corpus callosum as well as unilateral removal of the striate cortex (*4*). Interhemispheric transfer of learning was measured by the success or failure of interocular or somesthetic transfer of respective visual and tactual discriminations.

Visual tasks included brightness and pattern discriminations acquired monocularly with food-reward or shock-avoidance techniques, or both. These experiments were conducted with normal cats and cats with chiasm and callosum section with and without unilateral ablation of the striate (visual) cortex (hereafter referred to as split-brain striate cats). In the brightness test, each animal learned to choose the lighter of two stimuli in order to obtain food. In the pattern test, each animal learned to select the correct one of two patterned stimuli either to obtain food or to avoid shock. For these test procedures, each of the two patterns was identical and differed only in orientation (for example, upright and inverted triangles, horizontal and vertical stripes). Brightness and pattern differences were alternated from left to right in random sequence (*5*).

Every animal in these experiments was given daily monocular training sessions until its performance was 90 percent or better for two consecutive days and showed nine correct responses on the first ten trials of the third day. Interocular transfer of the learned visual discrimination was tested by masking the trained eye and exposing the untrained eye to the test procedure. Training was continued until the animal's performance with the second eye reached the same criterion that was required with the first eye.

Tactual tasks consisted of a unilateral limb-flexion response conditioned to unilateral tactile stimulation in an instrumental avoidance procedure. This study was conducted with normal cats and cats with section of the callosum. In order to prevent the cats from obtaining visual cues, both eyes of the animals were masked throughout all training and testing.

Daily training continued until discrete, limb-flexion avoidance responses reached the same 90 percent, 3-day criterion stipulated above. Tactile stimulation was applied to the opposite untrained forelimb of the same animal to evaluate somesthetic transfer of the conditioned limb response. Training with the opposite forelimb was continued until the criterion of learning was reached.

To obtain an exact record of the number of training trials required with each eye or limb, no extra trials were given to break "position habits" when they occurred. Trials were numbered, and behavior was recorded in detail such that performance could be examined for the *n*th trial of any particular test session.

No split-brain cats with total section of optic chiasm and corpus callosum showed interhemispheric transfer of the pattern discrimination with food-reward procedures. Complete training with the second eye was necessary. Similar split-brain cats showed transfer of brightness discrimination at or above criterion levels. In contrast to the "no transfer" performance of split-brain cats trained with food-reward procedures, all split-brain cats with chiasm and callosum sections showed good transfer of pattern discrimination with shock-avoidance training. Normal cats demonstrated interhemispheric transfer of all visual discriminations in both food-reward and shock-avoidance procedures.

Total interocular transfer of brightness was obtained with split-brain striate cats from the eye on the cortically *ablated* side to the eye on the cortically *intact* side. Interocular transfer failed from the eye on the cortically *intact* side to the eye on the cortically *ablated* side. Complete training was carried out with the eye on the cortically *ablated* side.

Figure 1 compares the length of learning with the success and failure of interhemispheric transfer. Both clearly indicate that failure of interhemispheric transfer is associated with prolonged learning.

In the studies with food-reward training, pattern discrimination learning by split-brain cats was severely impaired. When compared with monocular training trials required by unoperated cats, split-brain cats needed at least twice as many trials to reach criterion in the same pattern discrimination task. Normal cats needed an average of 325 trials, and split-brain cats that failed to show transfer required an average of 815 trials. Brightness discrimination, on the other hand, not only transfers completely from one eye to the other in split-brain cats, but learning in this group proceeds like that in normal cats. Both normal and operated cats required an average of 250 and 295 trials, respectively. Results obtained with split-brain cats in the transfer of pattern discrimination with shock-avoidance procedures demonstrates the same phenomenon of normal learning time with successful interhemispheric transfer (400 trials).

Results of interocular transfer of brightness discriminiation with split-brain striate cats (chiasm and callosum section and unilateral striate ablation) confirm the previous findings. When these cats are trained first with the eye on the cortically ablated side, they show complete transfer to the eye on the cortically intact side and learn within the same rate as normal cats (290 trials). Opposed to this, split-brain striate cats trained first with the eye on the cortically intact side show "no transfer" to the eye on the cortically ablated side. This group needed an average of 780 trials to reach criterion, the time required to learn being almost three times as long as that of the split-brain striate, split-brain, and normal groups that showed complete interhemispheric transfer of brightness discrimination.

Cats with section of the corpus callosum showed no somesthetic transfer and relearned the conditioned limb-flexion response at a slower rate than the normal cats, which demonstrated transfer of the discrimination at high levels. By comparing the mean of 745 trials required by the operated cats to the mean of 370 trials required by the normal cats, we can see that the cats with the callosum section and no interhemispheric transfer took almost twice as long to learn as the normal cats.

The results of these studies (Fig. 1) point to a definite relation between prolonged learning and failure of interhemispheric transfer. These findings are not attributable to surgical insult, for interhemispheric transfer can occur or not occur, depending upon either the condition of training or the locus of initial input. Support for this association between length of learning and interhemispheric interaction comes from studies carried out by others. Prolonged learning was observed (*6*) when the transfer of brightness discrimination was abolished in cats after section of optic chiasm and of all forebrain commissures. A mean of 200 trials was compared with a mean of 524 trials by those cats that failed to transfer brightness discrimination.

A somewhat similar finding for the rat has been reported (*7*). Interhemispheric transfer of memory traces was tested in rats with one cerebral hemisphere subjected to Leão's spreading depression (*8*). Acquisition of an avoidance response in normal rats required 19.3 trials, whereas unilaterally depressed animals required 28.1 trials. Normal rats showed immediate transfer; the unilaterally depressed group showed none.

Additional, although indirect, support comes from a study of the effect of pyramidal lesions on the acquisition of a conditioned avoidance response (*9*). Prolonged learning occurred in two split-brain cats with pyramidal lesions. These animals took 2000 and 1300 training trials compared with 875, 850, and 755 trials needed by the other animals (pyramidal lesions only) to reach the criterion of learning. In a study of delayed response in monkeys with section of corpus callosum and hippocampal and anterior commissures (*10*), prolonged learning was evident.

In these studies, split-brain cats showing "no transfer" and prolonged acquisition have also shown a severe decrement in retention compared with normal cats and split-brain cats with successful transfer. Retention for 62 days at or above a 90 percent criterion has been obtained in normal and split-brain cats with successful transfer. In contrast, split-brain cats with no transfer show short-term retention of only 5 days at or above criterion.

Examination of individual training trials revealed two different patterns of behavior. The daily performance of normal cats and split-brain cats with successful transfer increases in increasing amounts. The performance of the "no transfer" split-brain cats suggests a time-dependent memory deficit. Although performance improves by the end of each daily session, the initial performance on the following day decreases. Thus, prolonged training is necessary before 24-hour retention can facilitate criterion performance. Even then, long-term memory is fragile (Figs. 2 and 3).

Further inspection of the learning curves of these animals suggests two different functions. Figure 2 shows the learning curve typical of normal and split-brain cats that show interhemispheric transfer, whereas Fig. 3 shows a learning curve typical of the split-brain "no transfer" group. The normal learning curve of Fig. 2 is that of a monotonic function which increases in increasing amounts early in training. It shows little fluctuation and rapidly reaches the criterion of learning. On the other hand, Fig. 3 is a curve of long duration with a trend toward flatness. There are small daily fluctuations throughout the major portion of training and a sharp increase toward criterion, marking the end of the learning period. Early performance of this type is always indicative of an animal that will fail to show interhemispheric transfer.

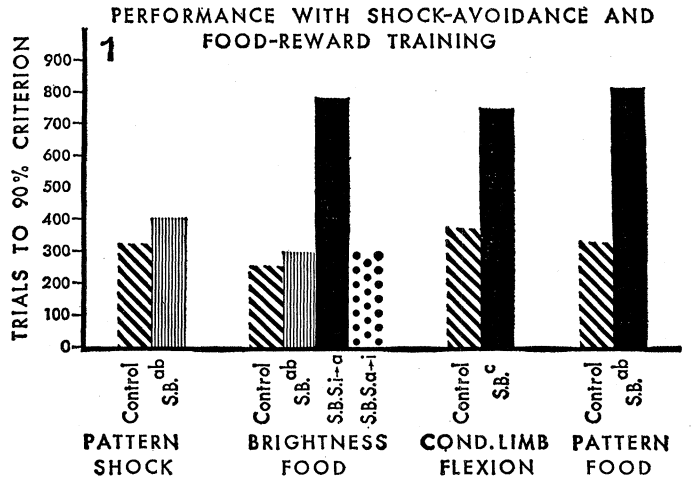

Fig. 1. Bar graph compares the mean number of training trials required by animals showing successful interhemispheric transfer and by animals showing no interhemispheric transfer. *S.B.*^{ab}, Split-brain cats with section of optic chiasm and corpus callosum. *S.B.*^{c}, Split-brain cats with section of the corpus callosum. *S.B.S.*$^{i-a}$, Split-brain striate cats with section of optic chiasm and corpus callosum and unilateral striate ablation. Transfer tested from the eye of the cortically intact side to the eye on the cortically ablated side. *S.B.S.*$^{a-i}$, Split-brain striate cats with section of optic chiasm and corpus callosum and unilateral striate ablation. Transfer tested from the eye on the cortically ablated side to the eye on the cortically intact side.

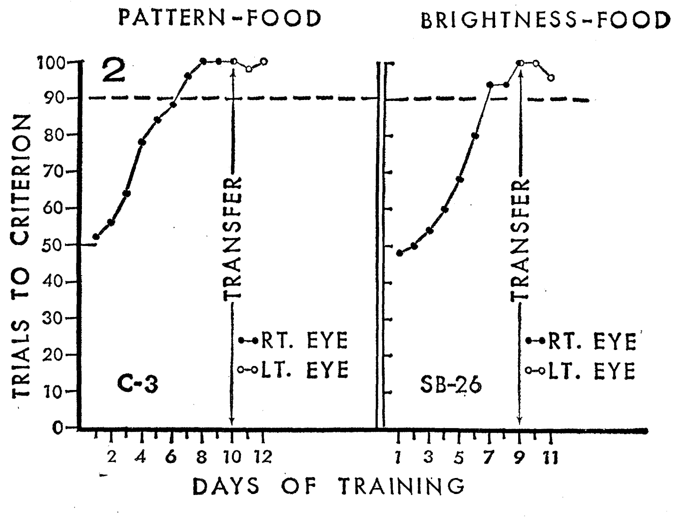

Fig. 2. "Bilateral hemispheric" learning curve of a normal cat and a split-brain cat, both of which show complete interhemispheric transfer (interocular) with normal acquisition.

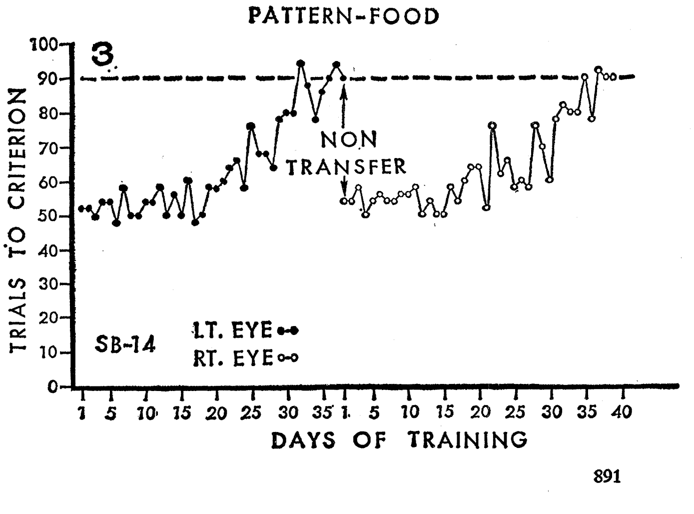

Fig. 3. "Unilateral hemispheric" learning curve of a split-brain cat that shows "no interhemispheric transfer" and requires prolonged training with the first as well as with the second eye.

Thus, the learning curve in Fig. 2, or a typically normal learning curve, represents the function of both cerebral hemispheres during learning. The learning curve in Fig. 3 must then represent the function of only one hemisphere during learning. They are respective "bilateral hemispheric" and "unilateral hemispheric" products.

Participation of only one hemisphere during learning would necessarily limit by one-half the available amount of brain tissue. If this is so, differences in learning rates between animals capable of interhemispheric transfer and animals incapable of interhemispheric transfer could be explained by Lashley's principle of cortical mass action (*11*). With this hypothesis it is not unreasonable to expect that the animal with both cerebral hemispheres interacting during learning should require less training than the animal with only one hemisphere participating in the same learning situation. We would also expect retention to be different since, in the unilateral hemispheric situation, there is less brain mass available for memory storage during and after acquisition.

Although the underlying mechanisms are unknown, the main conclusions suggested by these studies are that (i) both cerebral hemispheres usually participate during learning, (ii) the normal learning curve and thus the normal rate of learning appears to be a function of bihemispheric processing of information, and (iii) memory during and after acquisition is one of the functional relationships between the hemispheres.

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Photoreception in Sparrows: Response to Photoperiodic Stimuli

Underwood and Menaker (*1*) exposed blinded and normal house sparrows (in about equal numbers) to 18 different photoperiod treatments. Testicular weights were recorded at autopsy for each member of each group. The difference in testes size between the two groups (normal and blinded) in each condition was evaluated by Student's *t*-test. The *t* value obtained had a probability of less than .05 in only one condition. The authors conclude: "Our data offer no support for the hypothesis that the retina is involved in this [the testis] response." Their data not only do not support this conclusion, but very strongly support the opposite conclusion.

Their statistical logic is profoundly faulty, for they have confused failing to reject the null hypothesis with confirming it. Failure to reject the null hypothesis does not mean that it is true, but only that there is some chance that it is true. When the null hypothesis is not rejected at .05, we know only that there is better than one chance in 20 that it is true. Knowledge would progress very little if we accepted every hypothesis that has at least one chance in 20 of being true. Therefore, the null hypothesis is never confirmed by a statistical test, but only rejected or not rejected. Rozeboom (*2*) discusses this matter at greater length and with exceptional clarity.

Table 1. The number of blinded and sighted individuals in groups with larger or smaller average testes summed across all photoperiod conditions.

Average testes	Sparrows (No.)	
	Blind	Normal
Larger	48	114
Smaller	97	52

Thus this statistical analysis did not (and could not) support their conclusion. However, it seems quite likely that a different analysis of their data might have supported the opposite conclusion.

When the results of their experiment are considered as a whole, the differences between blinded and normal birds begin to look distinctly nonrandom. Their graphs show a consistent, though not invariable, tendency for normal birds to have larger testes. Their table shows that normal birds have heavier testes in 12 of the 18 different lighting conditions, while blinded birds have heavier testes in only six. The larger the group of animals in each condition (and therefore the better the sample) the more pronounced is this tendency.

One can fairly ask why this tendency, if it reflected a real difference, would not produce statistically reliable differences. There are two important reasons why it might not, both having to do with the lack of statistical power in Underwood and Menaker's experimental design and data analysis.

Some power was lost by their choice of a two-tailed rather than a one-tailed test. A one-tailed test (appropriate in view of the existence of a strong theory) would have led them to reject the null hypothesis in three of the 18 conditions. But this is not so much a problem in itself as a reflection of the second, more fundamental, problem—their experimental design had far too few animals in each condition. The number of subjects in several conditions is so small that it makes rejection of the null hypothesis very unlikely, no matter how much the groups differ.

Underwood and Menaker could still have salvaged some data despite their design by increasing the sizes of the groups during analysis. Probably the best technique would have been to calculate the mean testis weight for each condition and then to determine how many sighted and blinded subjects fell above this mean, and how many below it. The number of sighted and blinded birds in each category could then be compared by χ^2.

Unfortunately, this analysis cannot be performed on the data in Underwood and Menaker's report, but the χ^2 can be estimated. The fourfold table can be generated in the following way. For one cell, use the total number of sighted birds in all conditions where sighted birds had a larger mean testis weight than blinded birds as an esti-