Letters

Digestive Tract of the Oyster

According to an item in *Science* [**135**, 360 (2 Feb. 1962)], the Food and Drug Administration refused to certify whole fish flour because the viscera were included in its manufacture. If the product is chemically and bacteriologically clean, it is clean, but I do not wish to join the controversy.

However, one statement startled me: "[Commissioner George P.] Larrick agreed that a number of whole-fish products—sardines, shrimp, oysters, and clams—have FDA approval, but he noted that these had gained consumer acceptance before FDA was established." Apparently Larrick believes that the intestine of an oyster contains putrid fecal material. That concept is erroneous. A raw oyster from an unpolluted bed is as clean as a cabbage growing in a field. Up until a few years ago one could order "dressed" oysters at some hotels, which had the green digestive gland removed because of the mistaken idea that it was feces. This gland is sometimes called the hepatopancreas, although its functions are somewhat different, and it is one of the more nourishing and better parts of the oyster.

The digestive tract of an oyster is ciliated. Diatoms often go through the whole system alive and are deposited with the little pellets of sand, silt, diatom shells, and mucus, which are called feces. Decay does not take place in the oyster's short intestine, and the ejected material is not comparable to mammalian feces, which are largely products of bacterial decay. In fact, it is indistinguishable in gross characteristics from material rejected before it reaches the oyster's mouth and cast off into another little pile.

In small Virginia oysters growing on glass slides the lower valve is transparent until the oyster is several millimeters long, and its internal workings can be viewed under the compound microscope. I have watched balls of food formed by the labial palps go into the mouth and traverse the whole digestive tract of these small oysters in less than a minute. R. W. Menzel [*Univ. Texas Inst. Marine Sci. Publs.* **4**, 123 (1955)] has shown that oysters 12 millimeters long passed carmine particles 6 minutes, and stained plankton 10 minutes, after ingestion. Food particles go through the digestive tract of oysters much too fast for bacterial decay to take place.

There is an injustice concerning the use of American oysters which deserves comment. Oysters from foreign countries coming originally from polluted beds are imported and pass Food and Drug Administration standards, as they should, because they are sterilized in the canning process. On the other hand, oysters from polluted beds in this country cannot be used for canning, and hundreds of acres of oyster beds are unused every year because domestic canners are not permitted to produce clean products under the same conditions that foreign canners are.

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The Basic Variable in the Early Handling Phenomenon

I wish to direct attention to some logical and procedural difficulties contained in the recent report by Schaefer *et al.* (*1*). The authors hypothesize that "the effects of early handling are due to lowered skin or body temperature." The hypothesis was tested by handling one group of rats ("handled" group), placing the litters of another group in a refrigerator set at 7° to 10°C ("cold-exposed" group), placing the litters of a third group in a nonfunctioning refrigerator maintained at room temperature (23°C) ("cold-control" group), and not manipulating a fourth group ("nonhandled" group). These treatments were continued daily throughout the first week of life. At 12 days of age half the pups from each group were subjected to stress by being placed in a refrigerator at 55°C for 90 minutes. After this they were sacrificed, and the ascorbic acid content of the adrenals was determined. The ascorbic acid content of the adrenals was determined for the remaining animals without their undergoing the "cold stress."

Measurement of the levels of adrenal ascorbic acid in four differentially manipulated groups under two conditions ("cold stress" and "nonstress") involves a 4×2 factorial analysis. The only analysis made, however, was a test of the difference in findings for "stressed" and "nonstressed" animals *within* a given group. The adrenal ascorbic acid content of the cold-stressed handled animals was significantly lower than that of the nonstressed handled animals, and animals previously subjected to cold had a significantly lower level of adrenal ascorbic acid after cold stress than the similarly manipulated nonstressed animals. There were no differences in findings for the stressed and the nonstressed animals of the nonhandled or cold-control groups.

On the basis of this analysis the authors state, "These results indicate that the essential aspect of the handling procedure is a drop in environmental temperature accompanying removal from the nest." Further, it is stated, "Subjecting the pups to low temperature on days 2 through 7, although they were somewhat insulated in the nest by the mother, produced the same effect as handling (which exposed pups to room temperature for the same amount of time)."

That the effects of handling and exposure to cold are the same (or different) was, technically speaking, not tested in this experiment. Clearly, there is a significant *depletion* of adrenal ascorbic acid in response to cold stress in both handled and cold-exposed groups; however, there was no reported test of the possible difference between the depletion scores of handled and of cold-exposed groups.

Figure 1 of the report suggests that there was no significant difference between the *depletion* scores of handled and of cold-exposed animals. The significance of the difference in the scores for the cold-exposed and the cold-control groups cannot be determined from the figure. If the difference for these groups is significant, then it is likely that results for the cold-control group also differ significantly from those for the nonhandled animals, since the

absolute difference between the data for these latter groups is greater than that between the data for the cold-exposed and the cold-control animals, and there is considerably less variability and overlap between the cold-control and the nonhandled animals. A difference in results for these latter groups, neither of which had been previously exposed to cold, would be difficult to explain on the basis of a hypothesis which places emphasis on prior experience with cold.

The main difficulty, however, lies in the fact that only *depletion* scores are presented. In this the authors are apparently following an unfortunate precedent (*2*), but a depletion score is a poor substitute for the actual values observed in stressed and nonstressed animals, since the same depletion score may be a resultant of a variety of actual values. Also, the graphic presentation of depletion scores rather than of actual values for adrenal ascorbic acid in stressed and nonstressed animals provides the reader with insufficient information for making his own interpretation of the results. In any case, since one cannot prove the null hypothesis, even if the authors had compared the data from handled and cold-exposed animals and had found no significant difference, they would not be justified in stating that exposure to cold effects the *same* response to cold stress that handling does. That both handling and exposure to cold during the first week of life can result in a significant depletion of adrenal ascorbic acid in response to cold stress at 12 days of age has been established. That the effect of exposure to cold and handling effect the *same* change in the level of adrenal ascorbic acid remains to be determined. In this respect the authors, in presenting depletion scores rather than the stress and nonstress values for all animals, may have been doing an injustice to their own data.

One final point requires comment. There is no logical justification for the authors' contention that, because handled and cold-exposed animals show a significant depletion of adrenal ascorbic acid in response to cold stress, the essential variable in the handling procedure is a decrease in environmental temperature. Merely because event *B* produces the same effects as event *A* (this was not demonstrated by the data presented), it does not necessarily follow that the effects of *A* are therefore mediated by *B*, as the authors have implied.

In these comments I do not mean to

deny that reduction in environmental temperature may be a contributing factor in the behavioral and physiological changes effected by handling. However, the data presented by Schaefer *et al.* hardly warrant the conclusions that change in environmental temperature is the basic variable in the early handling phenomenon. As Schaefer *et al.* and others (*3*) have pointed out, what is really required in approaching this question is the direct measurement of a variety of physiological changes concomitant with handling and other types of early manipulation.

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The interesting results notwithstanding, the thesis and the conclusions drawn in the recent study by Schaefer, Weingarten, and Towne (*1*) are not convincing. Several points raise serious doubts about the validity of considering decrease in temperature *the* variable in studies of early handling phenomena, and about the applicability of this experiment in particular.

1) The effects of handling in infancy have been noted even in those instances in which a split-litter technique was employed, with removal of the mother at the time of experimental treatment (*2*).

2) The stipulation that vision and hearing are nonfunctional during the first postnatal week in the life of a rat is tenuous at best. A similar belief prevails for the cat, yet data that I have collected indicate that electroencephalographic and electromyographic responses to visual and auditory stimuli can be recorded from birth. Although visual and auditory stimulation may not be the most promising of the potential variables in the early experience phenomena, they have not been decisively excluded.

3) In view of the recognition given the view that "any of several modes of extra-stimulation" will induce the handling phenomena (*3*), the failure to expose a group of subjects to an increase in temperature is difficult to comprehend. The results reported would have

been much more impressive had such an experimental group been included.

4) Even when one grants the uniqueness of some of the early experience phenomena or the possibility of species specificity, the critical experimental condition of this study is not a reasonable one. A temperature of 7° to 10°C is hardly one that would be found in any of the laboratories in which handling phenomena have been investigated. I concede that exposure to this temperature is justifiable to show that temperature is *a* variable, but such a condition is preposterous as a means of showing that temperature is *the* variable in the handling phenomena.

5) Finally, what was the mother animal doing while she and her litter were in the cold environment? Could her reaction to the pups be such that tactile stimulation was increased, and a "normal" temperature for the pups thus maintained? Studies of other animals under even more extreme conditions indicate that, in this case, parental behavior may have constituted a homeostatic mechanism.

On the whole, the study and the investigators widely miss their mark.

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It should have been clear from our report that we considered our work only "an initial test of the hypothesis that the effects of handling are due to lowered skin or body temperature." The experiment was an attempt to demonstrate the importance of temperature change without making direct measurements. The investment in equipment and time needed to perfect techniques for such measurement did not seem warranted unless it could be shown that the temperature variable was worth investigating further. Both the title of our report and our conclusion that temperature change is a basic variable in the early handling phenomenon were presented tentatively. We stated that "direct measurements of skin or body temperature, and of neurologic, metabolic, or physiological changes concomitant with handling, would be needed to verify these speculations," and that "it re-

mains to be demonstrated that handling does, in fact, affect skin or body temperature in infant rats, and that such a change has the same effects on later behavior as handling."

More recently we have determined that lower body temperature does result when rat pups are removed from the nest. In addition, the magnitude of the temperature changes in the handled, refrigerated, and refrigerator-control animals is proportional to the magnitude of the depletion effects we found in using these treatments. During the first week of life the oral temperature of the rat pup drops approximately 2°C within the first 2 minutes after removal from the nest and about 5°C during 10 minutes of exposure to room temperature (23°C). By comparison, the temperature of pups left in the intact nest after the mother is removed declines less than 1°C in 20 minutes. However, placing the nest cage containing the mother and pups into a refrigerator at 10°C causes the pups' temperature to fall, on the average, 1.75°C, whereas the temperatures of control pups in cages similarly placed in the nonfunctioning refrigerator at 23°C drop less than 1°C within the 10-minute period. The slight temperature drop in the controls may be attributed to the failure of the mother to return her pups to the nest promptly after they have been scattered by her excited activity during the movement of the cage to the refrigerator. In contrast, the mothers placed in the refrigerator at 10°C gather their pups into the nest as soon as they enter the cold environment.

We have also extended the parallel that we found between the results of handling and of exposure to lower temperature by comparing the behavior of handled, nonhandled, and cold-treated animals in studies of lever-pressing for water and of conditioned avoidance. We found a curvilinear relationship between severity of treatment and behavioral effect which is similar to the relationship between severity of treatment and handling effects. In these studies we exposed the pups to cold in a different way, placing them in metal containers maintained at various temperatures.

In the light of these new data it seems unnecessary to discuss several of Meier's and Ader's criticisms. Ader's concern about our analysis of the data would be justified, perhaps, if adrenal function were the issue. It is not. We merely used a technique [adrenal ascorbic acid (AAA) depletion as a response to stress] and a simplified statistical analysis

which have evolved as a measure of the effectiveness of early treatment; these have been described in at least four published papers by Levine *et al.*, cited in our report. We chose this measure because of its demonstrated sensitivity and reliability, and because it permits early determination of the effects, making it unnecessary to keep the animals until they are adults. Our very close replication of Levine's values for AAA depletion in 12-day-old handled animals further supports the validity of this technique as a measure of the effectiveness of early treatment. However, we certainly agree with Ader's general criticism that the hypothesis was not *proved* by our study. Our continued work along the lines he recommended is evidence of this.

The results of our temperature determinations are more directly pertinent to Meier's criticism. As to Meier's point 1, his demonstration that cats did not show an effect when the mother was removed from the nest, possibly subjecting the kittens to lower temperature, does not vitiate our suggestion. Our temperature measures show that the control pups do not necessarily undergo a temperature loss when the mother is absent if the control pups are left in an intact nest. Perhaps this would be as true for the kittens Meier used in his experiment as it is for rats, though we feel less ready to generalize from our rats to his cats than Meier is to draw analogies between his split-litter cats and our rats. Yet, for hypothesis making, such analogizing may be valuable. In fact, Meier's finding (*1*) that early-handled Siamese kittens developed pigmented fur sooner than nonhandled littermates was one of the considerations which led to our hypothesis. In the Siamese cat, as in the Himalayan rabbit, pigmentation of the fur is known to be thermolabile (*2*), the differential development of pigmentation being very sensitive to slight differences in environmental or skin temperature. When Meier's findings first appeared we considered the possibility that this thermolabile pigmentation mechanism might explain the darker pigmentation in kittens which had been removed from the nest daily for 10 minutes of handling. Although Meier concluded that the pigmentation was evidence of hastened maturation resulting from early stimulation, Iljin's work (*2*) indicates that even so mild an exposure to lower temperature might be sufficient to produce the thermolabile effect of darker pigmentation in Siamese kittens.

Our temperature determinations suggest that the 7° to 10°C environment for cold-treatment was not so preposterous as Meier suggests in his point 4. We selected this extreme temperature because we had observed the mother's behavior as "a homeostatic mechanism" (Meier's point 5) and sought to maximize the likelihood of producing a temperature effect in spite of the mother's attempt to keep her litter warm. We did not include a group of animals subjected to higher temperature, as Meier suggested in his point 3, because it did not occur to us that handling, as typically carried out, could possibly raise temperature in the pup. Again, direct measurement of body temperature during various early treatments has supported this assumption.

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Pay and Promotion

Concerning the item in "News and comment" [*Science* **135**, 713 (1962)] on the urgently needed federal pay reform, it should be emphasized that the term *federal service* is limited to service performed by civil servants and does not apply to the equally well trained, experienced, and responsible scientists in the military. Although the staff of several government research and development establishments is as much as 80 percent civilian, it should be realized that the presence of this staff is, in many cases, the direct result of the prior and continuing existence of a military nucleus. If this bill is passed, and it *must* be for the very reasons you indicate, the scientific and engineering colleagues of the civil servants who are in uniform will be three pay reforms in arrears.

Scientists in the military must await separate pay and promotion legislation. The latter is keyed to the military population as a whole. Military pay and benefits for the average officer (55 percent or more, depending upon the