HISTORICAL PERSPECTIVES

From metabolism and behavior to respiratory physiology: an educational and research perspective

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Schlenker EH. From metabolism and behavior to respiratory physiology: an educational and research perspective. Adv Physiol Educ 44: 540–544, 2020; doi:10.1152/advan.00047.2020.—Throughout my academic career, I have been extremely fortunate to have as mentors, teachers, and advisors, remarkable individuals who helped me develop into a competent researcher and passionate teacher. This essay describes a period in my career as a graduate student that was challenging, but also rewarding. I was fortunate to have contact with world-class researchers and teachers, including Drs. Clyde Herried, Leon Fahri, Hermann Rahn, Donald Riggs, Verner Noell, and Barbara Howell. In addition, I attended excellent University of Buffalo Department of Physiology seminars presented by world-renowned scientists. Looking back on the experience, allows me to appreciate the large impact they made on my subsequent career in education and research in physiology and neurobiology.

behavior; CO₂; mentors; metabolism; mice

When I was a graduate student at the State University of New York at Buffalo (UB) in the 1970s, I worked on a dissertation project inspired by a small section of a larger article published in 1947 investigating the role of metabolism of small mammals (15). In the paper, a series of studies indicated that metabolic requirements of a mice dropped dramatically (30–50%) when housed in groups of two or three rather than alone, even when housed in their thermoneutral temperature. From an ecological physiological perspective, this was beneficial for the survival of mouse, since they expended less energy obtaining food, potentially putting them in harm’s way from predation. The author did not discuss the mechanism responsible for this effect, but it was thought to be the result of huddling. Other possibilities included pheromones or other sensory cues. Thus my project was to determine the factor or factors responsible for this physiologically advantageous phenomenon.

I was fortunate to have as my mentor for this project, Dr. Clyde Herried who had suggested at one time that, because of my interest in olfaction, I work on olfaction in vultures (I felt that I did not have the expertise to conduct the vulture study), but then agreed to have me pursue the metabolic project, which was in line with previous research he had conducted. Dr. Herried received his master’s degree at Johns Hopkins and his doctorate in physiology at Pennsylvania State University. He went on to do postdoctoral work at Duke with the renowned comparative physiologist, Dr. Knut Schmidt-Nielsen. As a visiting assistant professor at Duke University, Herried worked on temperature and oxygen consumption in bats. He published papers in the American Journal of Physiology, as well as in Science (7–9).

Aside from a researcher who had several doctorate students working in his laboratory during my tenure, Dr. Herried directed and taught in a very well-received upper level undergraduate comparative physiology course. Many of his students, including myself, were graduate assistants in this course, teaching in laboratories and giving a lecture or two in the course. The lectures were chosen to be outside our research direction, thus challenging us to learn new areas. I am indebted to Dr. Herried’s role model as an excellent and engaging teacher. Dr. Herried’s teaching skills and innovations were then and still are extremely well regarded, as witnessed by many prestigious awards he received (6), numerous publications about teaching modalities, and establishment of the National Center for Case Study Teaching in Science in the early 1990s (https://sciencecases.lib.buffalo.edu/about/staff/). The mission of the center is “... to promote the development and dissemination of materials and practices for case teaching in the sciences.” To accomplish the mission, the group has a website that provides instructors teaching at various levels a broad collection of peer-reviewed cases in particular topics. In addition, the group offers a summer workshop and fall conference to teach instructors how to use case-based science teaching. Over the years, the group has also investigated the outcomes of this form of education, resulting in over 100 publications, as noted in Dr. Herried’s resume. Having used this method in teaching myself, I concur that case-based learning is a powerful way to engage students to think critically. Only after I left UB did I come to appreciate that, aside from laboratory research, Dr. Herried’s passion was in the area of educational research and use of case-based teaching modalities.

Back to my dissertation work that examined two issues. The first was to determine how different types of social groupings in Mus musculus (the albino mouse) affected metabolism, and the second was to determine what factor or factors were responsible for the decreased metabolism noted in grouped animals. For both studies, I needed to construct a chamber that allowed measurement of flow rate through the chamber as well as concentrations of oxygen and carbon dioxide entering and leaving the chamber to calculate oxygen consumption, CO₂ production, and respiratory quotient. I was exceedingly fortunate that we had an excellent Department of Physiology in close proximity to Dr. Herried’s laboratory, including several world-renowned respiratory physiologists and very capable technical help.
The equipment I had available to measure the concentrations for oxygen, nitrogen, and CO₂ was a Scholander micrometer gas analyzer (21). This chemical- and volume-based gas analyzer was very accurate, depending on chemical reactions and the skill of the operator, which was to be me. I was fortunate to have Dr. Leon Fahri and his excellent technician, Sandra England, as my teachers. Both had the patience of Job. However, when I finally mastered the technique to their stringent dictates, I was rightfully told by Dr. Fahri to set the Scholander apparatus up in my own space instead of having to go down several flights of stairs to Dr. Fahri’s laboratory to analyze samples.

During the time I spent in his laboratory, little did I appreciate the remarkable and fundamental accomplishments of Dr. Fahri and his colleagues in the areas of gas exchange, alveolar-arterial oxygen gradients, noninvasive measurements of cardiac output using a single-step CO₂ rebreathing method, effects of gravity on gas exchange, analysis of gases in small volume samples, and mathematical models. The latter were also applied to teaching concepts in gas exchange and acid-base balance (4, 13). These studies were seminal contributions to the area of respiratory physiology. Dr. Leon Fahri was also an excellent teacher of respiratory physiology and an historian writing an account of the Fenn, Rahn, and Otis laboratory in Rochester during WWII that I discuss later (3). Sandra England went on to receive a doctorate and did a postdoctoral fellowship in the Department of Physiology at Dartmouth Medical School under Donald Bartlett and became a well-regarded pediatric respiratory scientist at the Research Institute, Hospital for Sick Children, University of Toronto, and finally at the Rutgers Robert Wood Johnson Medical School. Moreover, Dr. England became a colleague of mine, promoting the role of women scientists and clinicians in leadership roles at the American Thoracic Society.

During this time, I also took courses, including Medical Physiology directed by Dr. Herman Rahn, Medical Neuroanatomy, neurophysiology courses in the Physiology Department with the well-known neurophysiologist, Dr. Werner Noell, where I learned to do electro-olfactograms in frogs, and animal behavior courses under Dr. Peter Gold. Moreover, a course that would have a major impact on my future involved understanding mathematical approaches to physiological problems and was taught by Dr. Douglas Shepard Riggs, a faculty member and former Chair of the Pharmacology Department. In addition, I took two courses in statistics in preparation of data analysis. All courses were challenging and prepared me for critical thinking skills to this day.

Returning to my dissertation work, I determined that the first question of investigating the behavior of different social groupings was relatively easy to investigate. I constructed three groups of mice. One group consisted of animals that were housed singly, another group consisted of animals that were consistently housed together (stable group), and the third group of mice were housed with a new member appearing daily (unstable group). During this study, I evaluated the effect of social groupings on a variety of behavioral interactions (sniffing, digging, grooming, tail rattling, fighting, and latency to fight). The second part of this project was to determine CO₂ production over a 5-day period. For that I needed to construct chambers using Plexiglas, since mice chewed their way through less substantial (and less expensive) constructed chambers, much to my chagrin. I used the Scholander device to determine the CO₂ levels entering and leaving the chambers, as well as the flow rate of air traversing each chamber. The lowest CO₂ production was determined in the stable group, followed by that in the unstable group. The highest CO₂ production was found in the singly housed mice. It was 167% greater than that of the stable group. Thus the type of grouping had profound effects on behavior and CO₂ production, possibly associated with increasing levels of stress of isolation.

In contrast to the project described above, trying to delineate factors that caused grouped mice to decrease their metabolic requirements proved to be an engineering project. The approach I developed consisted of having a donor chamber attached in series to a recipient chamber. The donor chamber could contain a male or female mouse, dirty litter from mice, clean litter, or gas from a tank. Variables evaluated included flow rate through the chambers and concentration of O₂ and CO₂ entering and leaving each chamber. Moreover, both the behavioral and metabolic studies needed to be done in a relatively quiet environment. A laboratory with three other student investigators engaged in their own projects would not work. Thus I was relegated to do the projects in a turned-off walk-in cold room. Another factor to overcome was getting a clean supply of air to flow into the chambers. I found to my dismay that the compressed air in the building contained a respectable level of CO₂ (>0.5%), surely a major confounder. To deal with the issue, I received funding to purchase an air compressor that was placed in another room to mitigate the loud sound it produced. I have to thank the Physiology Department and extremely capable technicians in their shop for helping me devise this system. I was able to study up to 10 mice at a time. In the process, I learned a great deal about ventilation of chambers, knowledge I used in my postdoctoral work at the University of Florida and then my faculty position at the University of South Dakota to understand ventilation of confinement barns housing hogs on respiratory health in farmers (5).

After several studies using dirty litter and just a clean donor cage, it became apparent that a living donor mouse was required to explain the 30–50% decrease in metabolism of the recipient mouse. Results also indicated that the sex of the donor or recipient mouse equally elicited a decrease in metabolism over several hours (10). Moreover, this study also indicated that the social status of a mouse also affected its metabolic rate. Thus mice maintained in stable social conditions had considerably lower metabolic rates than those maintained in unstable social situations. The highest metabolic rates were found in mice maintained in isolation, as previously mentioned.

I still held onto the hypothesis that a pheromone acting through the olfactory neurons was required to produce the decrease in metabolism noted in the recipient mouse. However, this hypothesis was quickly debunked when I did an experiment making mice anosmic by irrigating their olfactory cells with a zinc sulfate solution. Although the recipient mouse was unable to smell food pellets, it still responded to air coming from the donor mouse with a decreased metabolism (20). Additional studies included having donor animal air enter chambers of recipient mice with two pathways. One was containing air going through a soda lime and CaCl₂ filter to remove the CO₂ or a dummy filter consisting of sand and a CaCl₂ filter (Fig. 1). The results suggested that low levels of
CO₂ could elicit a 30–50% reduction of metabolism in the recipient mouse, results that were unexpected. At that point, Dr. Herried enlisted the help of several faculty members in the Physiology Department, including Drs. Herman Rahn and Barbara Howell, to guide my research. After my technique using the Scholander was thoroughly vetted against an HPLC and a paramagnetic CO₂ analyzer, Drs. Rahn and Howell agreed to direct my continuing research project. What I appreciated the most about the two investigators was their intelligence, research rigor, professionalism, and kindness. To investigate the level of CO₂ that was required to decrease metabolism in recipient mice, a dose-response approach was taken. This final dose-response CO₂ study clearly indicated that different levels of CO₂ ranging from 0 to 1.7% affected O₂ consumption and CO₂ production differently, with the lower levels of CO₂ of 0.14, 0.23, and 0.5 depressing metabolism up to 50% (19). I also went on to test my hypothesis on another species of rodent, the very jumpy deer mouse (*Peromyscus maniculatus bardi*). The results of the unpublished study with the deer mice were similar to those I found in *Mus musculus*. Subsequent studies in a variety of species confirmed these results (1, 12, 22).
Most importantly, over time I came to appreciate the remarkable research into fundamental and applied respiratory physiology associated with gas exchange and acid-base balance conducted by these mentors. Dr. Hermann Rahn (1912–1990) began his research career studying endocrinology of the avian pituitary gland and reproduction in snakes and sexual dimorphism in the sage grouse as a faculty member at the University of Wyoming. During a stop in Rochester, New York, on the way to visit his parents in Ithaca, he met Dr. Wallace Fenn, who offered Dr. Rahn a position in the Physiology Department. Quickly, Dr. Rahn became involved in seminal work associated with high-altitude studies required by the military during World War II. Dr. Rahn worked with Drs. Wallace Fenn and Arthur Otis. Their work was described eloquently by Dr. Leon Fahri in 1990 in an article entitled, “World War II and respiratory physiology: the view from Rochester, New York” (3) and by John West in a 2012 paper entitled “The physiological legacy of the Fenn, Rahn, and Otis school” (23). One of Dr. Rahn’s many classic papers was entitled, “A concept of mean alveolar air and the ventilation-blood flow relationships during pulmonary gas exchange” (17). In this paper, Dr. Rahn showed how regional differences in the ratio of alveolar ventilation-to-perfusion ratios influence the alveolar-arterial oxygen gradient. The clinical relevance of these findings is especially important today in managing patients with lung diseases that lead to hypoxemia. Dr. Rahn’s draw to high altitude resulted in several studies of acclimatization to high altitude. At the University of Buffalo, Dr. Rahn also investigated diverse topics, including the effects of free diving on the respiratory system, gas exchange in eggs from a variety of environments, gas exchange in fish, acid-base regulation, temperature, and evolution. His contributions to respiratory physiology culminated in the 1960s with his election to the American Academy of Arts and Sciences, the presidency of the American Physiological Society from 1963 to 1964, and in 1968 as a member of the National Academy of Sciences (2).

The last series of studies that Dr. Rahn pursued with several investigators around the world involved factors influencing gas exchange in eggs during embryonic development. Hermann Rahn was a consummate and engaging teacher whom I first met when I took Medical Physiology, a course that he developed and continued to teach. The first lecture he presented in that course involved understanding basic concepts in physiology that were applicable to any organ system. That approach resonated with me and allowed me to apply these concepts to different aspects of research I engaged in throughout my career.

A few years after his death, an excellent biography of Dr. Hermann Rahn was written by Dr. John Pappenheimer (14).

Dr. Barbara Howell was an early recruit for the Physiology Department when Hermann Rahn became Chair of Physiology at UB in 1954. A photograph of the Department in 1962 includes Dr. Howell and another distinguished Professor, Dr. Barbara Bishop, who became my role model (Fig. 2) (16). Dr. Howell was a faculty member in the Physiology Department for 35 yr, coming from the University of Montana in the late 1950s and conducting research in the areas of the ontogeny of acid/base balance, effects of temperature on acid/base balance, and acid/base regulation in air-breathing fish. Dr. Howell was a mentee of, and close research collaborator with, Dr. Rahn, demonstrated by their co-authorships on several papers (11, 18).

Overall, I was extremely fortunate as a graduate student and later as a Parker B. Francis postdoctoral fellow (in pulmonary mechanics with Drs. Marc Jaeger and Arthur Otis at the University of Florida) to have as mentors individuals who were excellent researchers and also insightful, innovative teachers. My career is a legacy to their support of me at a critical period of my academic development. From them I learned not only how to conduct great research and ask critical questions, but also how to teach, mentor, and inspire students at all levels.

DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the author.

AUTHOR CONTRIBUTIONS

E.H.S. conceived and designed research; prepared figures; drafted manuscript; edited and revised manuscript; approved final version of manuscript.

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