bumin effect is that it combined with an uncoupler of oxidative phosphorylation which becomes manifest on aging of the mitochondria.

Release of an Uncoupler

When mitochondria prepared in the usual manner were diluted in distilled water, a substance was released that, upon addition to fresh mitochondria, produced an inhibition of aerobic phosphorylation. As in the case of aged preparations, the inhibition could be reversed by the addition of bovine serum albumin. The inhibitor was prepared as follows: freshly prepared mitochondria from 15 grams of liver were diluted with 450 milliliters of water (to give a final protein concentration of 0.1 to 0.15 percent) and kept at room temperature for 1 hour. The extract was centrifuged at 18,000*g* for 20 minutes, and the residue was discarded. The inhibitory factor was concentrated either by lyophilization or by sedimentation at 144,000*g* for 1 hour in a Spinco preparative ultracentrifuge and then suspended in water. As shown in Table 3, the factor specifically inhibits phosphorylation without affecting the oxidation of β-hydroxybutyrate and thus simulates the action of 2,4-dinitrophenol and other known uncouplers. The addition of serum albumin to this system counteracts the effect of the inhibitor.

During the course of this work, our attention was drawn to the studies of Polis and Shmukler, who reported their findings at the meetings of the American Chemical Society (*16*). These authors have isolated from liver mitochondria an electrophoretically homogeneous heme protein which inhibits aerobic phosphorylation and which is counteracted by serum albumin. They suggest that this factor participates in the process of phosphorylation as an acceptor of energy-rich phosphate.

In view of these results, it appears likely that the generally recognized lability of aerobic phosphorylation in mitochondria may be partly explained on the basis of the release of this inhibitor.

References and Notes

1. The following abbreviations are used in this article: AcAc, acetoacetate; ADP and ATP, adenosine di- and triphosphate, respectively; DPN and TPN, di- and triphosphopyridine nucleotide, respectively; DPNH and TPNH, reduced DPN and TPN, respectively; EDTA, ethylenediamine tetraacetic acid (Versene); G-6-P, glucose-6-phosphate; P_i, orthophosphate; 6-PGA, 6-phosphogluconic acid.
2. E. C. Slater and K. W. Cleland, *Biochem. J. London* 55, 566 (1953).
3. S. E. Lewis and E. C. Slater, *ibid.* 58, 207 (1954); B. Sacktor, *J. Gen. Physiol.* 37, 343 (1953–54).
4. M. I. Watanabe and C. M. Williams, *J. Gen. Physiol.* 37, 71 (1953–54).
5. A. B. Brodie and C. T. Gray, *Biochim. et Biophys. Acta* 17, 146 (1955); G. B. Pinchot, *J. Biol. Chem.* 205, 65 (1953).
6. C. Cooper, T. M. Develin, A. L. Lehninger, *Biochim. et Biophys. Acta* 18, 159 (1955); I. Raw, *J. Am. Chem. Soc.* 77, 503 (1955).
7. It is a pleasure to acknowledge the collaboration of Harvey Pinefsky in some of the work reported here, as well as the valuable technical assistance of Michael Kandrach.
8. W. C. Schneider, *J. Biol. Chem.* 176, 259 (1948).
9. W. W. Kielley and R. K. Kielley, *ibid.* 191, 485 (1951).
10. A. L. Lehninger, *Harvey Lectures Ser.* 49, 176 (1954).
11. J. H. Copenhaver and H. A. Lardy, *J. Biol. Chem.* 195, 225 (1952); A. L. Lehninger and S. W. Smith, *ibid.* 181, 415 (1949).
12. B. Chance and G. R. Williams, *ibid.* 217, 383 (1955).
13. A. L. Lehninger, *ibid.* 178, 625 (1949).

13*a*. Recently it has been possible to incorporate into this assay system the oxygen electrode described by Chance and Williams (*12*). The combined system—that is, the spectrophotometric assay of phosphorylation and the polarographic assay of oxygen concentration—makes it possible to determine P/O ratios directly.

14. R. E. Beyer *et al.*, *Exptl. Cell Research* 8, 586 (1955); F. E. Hunter and L. Ford, *J. Biol. Chem.* 216, 357 (1955).
15. H. Stern and S. Timonen, *Exptl. Cell Research* 9, 101 (1955).
16. B. D. Polis and H. W. Shmukler, *Abstr. Am. Chem. Soc. 126th Meeting,* 72c; *Abstr. Am. Chem. Soc. 128th Meeting,* 19c; H. W. Shmukler and B. D. Polis, *Abstr. Am. Chem. Soc. 124th Meeting,* 13c.

16*a*. Subsequent to the submission of this paper for publication, it was found that the addition of adenylic acid ($5 \times 10^{-4}M$), EDTA ($1 \times 10^{-2}M$) and Mg^{++} ($1 \times 10^{-2}M$) to the assay system maintained the rate of phosphorylation for longer periods and improved the reproducibility.

17. L. Berger *et al.*, *J. Gen. Physiol.* 29, 379 (1946).
18. A. Kornberg, *J. Biol. Chem.* 182, 805 (1950).
19. H. W. Robinson and C. G. Hogdin, *ibid.* 135, 727 (1940).
20. K. Lohmann and L. Jendrassik, *Biochem. Z.* 178, 419 (1926).
21. S. S. Barkulis and A. L. Lehninger, *J. Biol. Chem.* 190, 340 (1950).

Evis Water Conditioner

Graham DuShane

We feel obliged to let our readers share our interest in some of the highlights of a recent case before the Federal Trade Commission. It is in many ways parallel to that of the battery additive case [*Science* **123,** 1059 (15 June 1956) and page 1099 in this issue].

On 5 Feb. 1954, the Federal Trade Commission issued a complaint against the Evis Manufacturing Company of San Francisco, Calif. The company manufactures a product, the Evis Water Conditioner, which looks like an expanded pipe coupling with a vertical post integrally cast in the center of the internal chamber. The "conditioners" range in size from those that may be fitted into a 0.5-inch pipe to models that are intended to be fitted into large industrial or marine pipes and in price from $25 for the smallest model to $3700 for the largest bronze model. All models are made of either zinc-coated cast iron or bronze, and they are "intended to be fitted into water systems for the purpose of beneficially treating and conditioning water."

The task for the Government in pressing its charges of false advertising was made difficult by the fact that the respondents averred that treatment with the "conditioner" did not affect the chemical or physical properties of the water in any detectable way but only the *behavior of the water in use.*

Burden of Proof

The company further claimed that both castings were processed by a secret method in such a way that they differ from ordinary cast iron and bronze. Metallurgical and spectroscopic examination of the iron castings (the bronze castings were not examined) failed to show that they differed from ordinary cast iron, but the hearing examiner ruled the evidence inconclusive when a metallurgist admitted that certain minute areas in the etched surface could not be identified and the spectroscopist admitted that the presence or absence of some 26 elements

could not be detected by spectroscopic examination. The elements that could be detected appeared to be those of ordinary cast iron.

In a similar way, when a physical chemist, James I. Hoffman of the National Bureau of Standards, testified that there was no known way in which the castings could exert their effect and that such action would be contrary to the second law of thermodynamics, he was forced to admit that our scientific knowledge is still incomplete. Quotation from the hearings will serve to amplify this point:

"Q. Doctor, those are various types of energy that might be existing in any given water system and if you imposed upon that system some influence, either by the water coming into contact with the interface with metal or by some other means, would not that inherent energy in the system, at least would there not be a scientific possibility that in converting that inherent energy in one of those forms into energy in some other form that you could bring about a change in the physical characteristics of the body of water without violating this first fundamental law of thermodynamics?

"A. It would be beyond my comprehension, if it could be done.

"Q. Well, that is based upon the knowledge that you have today, is it not, Doctor.

"A. Yes, sir.

"Q. And again I assume I would be correct in saying that as a scientist you would not foreclose the possibility that at some future date that could be established?

"A. All right."

The hearing examiner commented, in part: ". . . we must conclude that Dr. Hoffman's testimony as a whole, like that of other witnesses previously considered in relation to this issue, fails to establish that the Evis Water Conditioner does not have a catalytic or other effect upon water passing through it so as to change the physical behavior of such water in a beneficial way." It is logically impossible to prove a negative.

Perhaps something further should be said about the claimed catalytic effect. The respondent claimed that "his device resembles a catalyst because the results accomplished, like those of a catalyst, are accomplished by mere physical contact, and because the device, also like a catalyst, remains unchanged after the reaction has taken place. He recognizes that it differs from a true catalyst in that the change accomplished is not chemical in nature." Hoffman's testimony on this point is of interest:

"Q. We have been told by the inventor of this unit that the action of it is closely related to catalysis, though it is not a true catalyst, because there is no attendant chemical change, but only the physical effect of the catalyst is involved. Is that theory scientifically sound, so far as you are concerned, Doctor?

"A. It is not.

"Q. Can you explain it?

"A. A catalyst can only change the rate of a reaction. It cannot make a reaction go that would not go otherwise.

"Q. You say that it would change the rate of the reaction. Is that a physical or chemical reaction?

"A. That is a chemical reaction."

And, later on, we have an exchange between Hoffman and his interrogator in which Hoffman replied to a question about whether water that had passed through the "conditioner" would be changed physically and, if so, ". . . would that effect persist after the water had gone beyond the Evis unit?" Hoffman replied:

"It could not . . . if it did it would violate one of the fundamental laws of thermodynamics."

The Government contended "That the Evis Water Conditioner will not cause dishes or glassware to dry without leaving water stains." One of the tests bearing on this point shows the technique in operation. Eight identical glasses were washed in a solution of warm water and soap. Four were rinsed in "treated" water and four in untreated water. After they were dried, two people examined the glasses. For the dirtiest ones, they picked two of the controls and two rinsed in "treated" water; for the cleanest glasses, they again picked two of the controls and two rinsed in "treated" water.

Of this experiment the examiner had this to say: "It appears that the tests . . . as to water stains prove nothing, because the negative and positive thereof were exactly equal. Accordingly we must conclude that the testimony offered in support of the allegation that the Evis Water Conditioner will not cause dishes or glassware to dry without leaving water stains has too little probative value to be regarded as substantial proof."

It was, of course, up to the Government to prove that the claims were false. But a scientist would turn the proposition around and say, with the support of logic and several hundred years of experience of the experimental method to back him up, that this test failed to show that there was any significant difference in "treated" and untreated water, but this was not the question at issue.

Making Proof Difficult

The company further claimed that the mode of installation is important. The "conditioner" should be grounded; pipes carrying "treated" water should not be located near pipes carrying untreated water; "treated" water would lose its special virtues if mixed with untreated water. If any of these conditions were ignored, the respondents claimed that the device could not be expected to work. Furthermore, they did not claim the conditioner would be effective with all water but only with "some water." This last claim, if accepted, as it was by the hearing examiner, clearly precludes any significant testing of the device. One might be using the wrong kind of water.

One of the reasons that the examiner gave for ruling evidence of rinsing tests inconclusive appears in his statement that the tests were ". . . laboratory tests rather than practical tests." In short, we suppose what this means is that laboratory tests on scale formation, rinsing, scum formation, soap use, and so on, are not held to be relevant to the question of the merit of a product.

We are not concerned with the decision of the hearing examiner on 26 Apr. 1956 to drop the complaint. The burden of proof rests on the Government. But we are astonished at the following statements by the examiner: "From the record as a whole, it appears that we may here be confronted with a device operating upon a principle unknown to or unrecognized by present-day science. The strongest indications of this possibility lie in the scientific testimony in support of the complaint, wherein the scientists admitted that they did not understand the theory upon which the Evis Water Conditioner purports to operate. . . ." *Mirabile dictu*! And ". . . we must not take the risk of interfering with the development of a device which may prove to be the first practical application of a scientific principle heretofore undiscovered." And "We cannot . . . justify the issuance of an order which might act as a brake on the wheels of progress."

Nature is inexorable; it punishes the child who unknowingly steps off a precipice quite as severely as the grown scientist who steps over, with full knowledge of all the laws of falling bodies and the chances of their being correct.—HENRY A. ROWLAND.