

References and Notes

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Arsenic and Water Pollution Hazard

Undue speculation in the press, scare headlines, and the drawing of wide-sweeping conclusions about alleged hazards to consumers have resulted from the report by Angino *et al.* (1). The full set of facts for evaluation should include the following.

1) Arsenic in trace amounts ranging up to a maximum of 70 to 80 parts per million (ppm) is introduced into detergent products as a constituent of the phosphate builder. Arsenic in trace quantities is also widely found in both animate and inanimate nature. For example, an average of 5 ppm in soils has been reported, with soils of volcanic origin having as much as 20 ppm. Arsenic occurs in most plants and animals, especially in fish and shellfish (amounts considerably higher than anything found in washing products have been reported in some seafoods; for example, 174 ppm in prawns, 42 ppm in shrimp, and 40 ppm in bass have been noted in local areas). A survey of arsenic in food that had been purchased in New England chain stores showed detectable amounts in most of the items (2).

2) Schroeder and Balassa (2) state, "pentavalent arsenic as arsenate is non-toxic in normal concentrations, is excreted rapidly largely through the kidneys, probably does not accumulate in human tissues, is a normal constituent of food, and may perform some unknown physiological function."

3) Arsenic exists in detergents in the pentavalent state (arsenate) and not in the highly toxic trivalent form (arsenite) that is prepared commercially for pesticide use. Moreover, any

human exposure to arsenic due to the trace amount in washing products is negligible in terms of the normal dietary intake. Even at the highest level reported by Angino *et al.*, the quantity is far too small to have a measurable effect on the acute toxicity of the product itself or to have any effect as a result of chronic ingestion. Typical wash water, even with the product that contained 80 ppm (the highest level detected) would contain only about 0.15 ppm arsenic, about one-fourth the amount estimated in the average diet. The skin effects of arsenic in detergents are equally negligible, both because of the valence form of the material and the exceedingly low concentration in which it would contact the skin.

4) Angino *et al.* have attempted to show that arsenic found in trace quantities in some detergents constitutes a potential threat to water quality. However, in only 4 out of 27 combinations (dilutions made in containers of different sizes) did the concentration of arsenic in wash water exceed drinking water standards. Angino *et al.* also measured the arsenic concentration of water and sewage in Lawrence, Kansas. The concentration in the raw drinking water ranged between 2.6 and 3.6 parts per billion (ppb), and in the finished water (after cold lime softening) arsenic ranged between 0.4 and 0.5 ppb. It was noted that 0.4 ppb is at the lower limit of detection for the analytical procedure used. The highest amount found, 0.5 ppb, is 100 times below the Public Health Service's mandatory limit. Arsenic in raw sewage in Lawrence ranged between 2.0 and 3.4 ppb and

averaged 2.7 ppb. After treatment, the range was from 1.5 to 2.1 ppb and averaged 1.8 ppb. Treated sewage thus contained a maximum of 1/25th of the arsenic permitted in drinking water. A single determination made in the Kansas River at Lawrence indicated an arsenic concentration of 3.3 ppb and 8.0 ppb in the same river at Topeka. If these data are accurate (particularly that for Lawrence), it is obvious that a major input of arsenic to the Kansas River comes from a source other than Lawrence sewage.

5) The USPHS Drinking Water Standards (1962) set a recommended arsenic limit of 10 ppb (10 µg/liter) and considers amounts in excess of 50 ppb grounds for rejection of a water supply. These standards are legally applicable only to water supplies subject to federal quarantine regulations (such as interstate common carriers) but have been generally adopted by most state departments of health or sanitation commissions as their individual standards. Generally speaking, the USPHS standards are predicated on the assumption that the daily intake of water over a lifetime is 2 liters per day.

Under no circumstances (whether examining raw and treated sewage, raw and treated drinking water, and river water) were Angino *et al.* able to present data that indicated that arsenic concentrations remotely approached those that would disqualify these waters as water supplies. Moreover, finding traces of arsenic in wash water (which is certainly not recommended for drinking, in any case) likewise does not constitute a threat. Angino *et al.* made no apparent effort to evaluate the impact of the use of farm insecticides (arsenicals) or industrial and municipal discharges on the one river they studied (the Kansas River). In fact, all they demonstrated was that arsenic, in trace quantities, is a ubiquitous material found widely in nature and that it does not constitute a hazard to water quality at the concentrations they reported.

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Angino *et al.* (1) imply that the arsenic they found in laundry detergent products was introduced by the enzyme added to improve the ability of the detergent to remove proteinaceous stains. These enzymes contain no arsenic. In the final stages of enzyme manufacture in our plant (Novo Industri A/S, Copenhagen) sodium chloride and other common salts may be used to precipitate the enzymes from the fermentation liquid. Such salts, which may also be used as "carriers" or "bulkings," do contain traces of arsenic, and these traces may then be carried into the final enzyme products that we supply to makers of laundry detergents. That such carry-over of arsenic is completely insignificant is demonstrated in tests conducted in the independent laboratories of Foster D. Snell, Inc. (2), where it was found that the arsenic content of Novo's Standard Alcalase was only 50 parts per billion (ppb).

Because consumer-type laundry detergents contain no more than 1 percent of Alcalase, the arsenic contributed by the carrier salts could thus not exceed 1 part in 2 billion. In fact, when diluted in the water load of a typical home laundry machine the arsenic falls to 1 part in 2×10^{12} . Continued references to the hazards of arsenic in "enzyme" household detergents would thus seem unwarranted.

Today's laundry detergents do contain phosphates, and phosphates contain traces of arsenic. Thus, detergents now on the market would contain traces of arsenic whether or not they were "enzyme" detergents. What Angino *et al.* were measuring in their detergent samples was primarily the arsenic in the phosphates.

I think that the report of Angino *et al.* is misleading for the following reasons. The Drinking Water Standards set up by the U.S. Public Health Service allow traces of arsenic in acceptable sources of drinking water. The term "recommended" therefore signifies that the PHS regards 10 ppb of arsenic as low enough to permit continued use of such water for drinking purposes. The so-called *mandatory* limit of 50 ppb indicates that only *at or beyond* this concentration of arsenic in a water supply does the PHS regard such supply as potentially dangerous for continuous use over a long period of time. Testimony before the U.S. Senate Committee on Public Works (3) indicates that experts are now pre-

pared to recommend raising the mandatory limit to 200 ppb. Nevertheless, Angino *et al.* say that 2 to 8 ppb (amounts *well below* those that the PHS recommends as "not cause for concern") are "high enough to cause a pollution problem and a potential health hazard."

Although housewives might immerse their hands in laundry water containing dilute detergents with trace quantities of arsenic, and residents of Lawrence, Kansas, may be exposed to arsenic in their drinking water, nevertheless Angino *et al.* have provided no basis for comparing these risks with those of other exposures to arsenic to which we are all regularly subject (4).

It is doubtful that the remark of Angino *et al.*, "the danger clearly exists that arsenic can be absorbed through the unbroken skin," can be supported by specific pertinent data.

Finally, arsenic in detergents is of the pentavalent and not of the trivalent form. The highly toxic trivalent arsenic used in pesticides, for example, is not present in detergents.

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3. Transcript of official record of Subcommittee on Air and Water Pollution of Committee on Public Works (U.S. Senate) 8 June 1970.
4. E. S. Pattison, *Science*, this issue.

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While arsenic is present in trace amounts in many natural food sources, soils, or fluids, such an argument is not reason for adding to the natural levels. That some of the arsenic in food sources is the result of arsenic pollution, although not necessarily from detergents, is documented (1). With much water being employed for irrigation and other uses, a most interesting study would be one of testing whether the amounts of arsenic in many foods, plants, fish, and the like have increased over the last 10 years, and whether detergents are contributing a significant portion of that arsenic. Why add a component which can be so easily kept out of the system if raw materials of higher purity are used?

Schroeder and Balassa also state (1, p. 95), "Differences in experimental results on feeding tri- and pentavalent arsenic largely points to the presence

of a homeostatic mechanism for arsenates (excretion) and a relatively less efficient one for arsenite. This mechanism has not been demonstrated in man because of the difficulty of distinguishing arsenate from arsenite by chemical analyses of fluids and tissues." They state again (1, p. 104), "Pentavalent arsenic as arsenate . . . *probably* does not accumulate in human tissue." The uncertainty in the above statement is clear—as these authors imply several times—that there is still much that we do not know concerning the chemistry of arsenate in humans.

Whereas the human intake of arsenic in streams may be low or negligible, such may not be the case for different species of plants or animals living in or using the water. What is the potential of these forms for reducing "non-toxic" arsenate (pentavalent state) to the toxic arsenite (trivalent)? Schroeder and Balassa's article lists many plants and animals that concentrate arsenic. How much of this arsenic taken from the water by organisms is consumed by humans?

Pattison stated that ". . . arsenic exists in detergents in the pentavalent state. . . ." We also question what evidence exists that the arsenic remains in that same oxidation state after entering the sewage system or the environment. If one considers the magnitude of the pollution in many of our streams as reported in several congressional hearings, then we do not believe that we can be so complacent about arsenic in waters. When the amount of dissolved oxygen in many streams or in portions of certain streams is zero or nearly so, then we must consider the real possibility that arsenic present in water as the arsenate stands a good chance of being reduced to the arsenite form. The dissolved oxygen in Houston Ship Channel, for instance, is zero or essentially so. This water course empties into the Gulf of Mexico where a large shrimp and fish industry exists—and one is led to wonder about possible buildup of arsenic over the last 10 years or more.

The tests of the water and sewage treatment plants showed that the arsenic was being added in a use cycle. If, in a nonindustrialized college town like Lawrence, the amount of arsenic indicated was being added, "What amounts are being added in heavily industrialized areas where other sources of arsenic exist?"

Detergents are not the only source of

arsenic pollution, so that, even if the amount of arsenic from detergents were not a hazard by itself, this amount coupled with the amount from other sources may be sufficient to lead to "40 ppm in bass." Finally, it seems to us a poor choice to allow the arsenic content in detergents to reach the levels we noted; especially when the technology exists to prevent the addition of arsenic in these widely used products. We suggest that in a time when pollution of all kinds in our environment is of obvious concern, why persist in adding to the system a potentially serious pollutant when it can be eliminated at the source?

In reply to Sollins, it was not our intent to imply that the arsenic found in detergents was introduced by the enzyme material. In response to the statement that the mandatory limit for arsenic may be raised to 200 ppb, it should be clear that this is not offered. On the contrary, the federal government and indeed several states are now closely looking at the environmental impact of many metals. Included at the head of the list of possible pollutants are lead, arsenic, and cadmium.

We suggest that failure to remove such potential pollution (and our report used the word potential) where possible, will lead to greater federal regulation which would require that it be shown that products (like detergents) will not contribute to pollution. We stand by our closing contention that a potential danger does exist and warrants further study.

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RNA Hybridization:

Competition between Species

Hansen, Spiegelman, and Halvorson (1) state that the equation

$$\frac{1}{1-F} = \frac{C'}{C_T} \cdot \frac{C_T}{C} \cdot \frac{A^*}{A} + 1 \quad (1)$$

"may be summed from species 1 to species i , where i is the number of competing species.

$$\sum_1^i \frac{1}{1-F} = \frac{C'}{C_T} \left[\sum_1^i \frac{C_T}{C} \cdot \frac{A^*}{A} \right] + 1 \quad (2)$$

Division of both sides by i gives

$$\frac{1}{1-F} = \frac{C'}{C_T} (\text{average slope}) + 1 \quad (3)$$

This is also a straight line with an intercept of 1." The summation from 1 to i of Eq. 1 does not yield Eq. 2. Division of both sides of Eq. 2 by i does not give Eq. 3; and Eq. 3 is not a straight line.

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Reference

1. J. N. Hansen, G. Spiegelman, H. O. Halvorson, *Science* **168**, 1291 (1970).

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The numeral 1 in Bolch's equation 2 should clearly be an i . To demonstrate, Bolch's equation 1 becomes, upon summation for each species of RNA from 1 to i ,

$$\sum_1^i \left(\frac{1}{1-F} \right)_i = \sum_1^i \left[\frac{C'}{C_T} \cdot \frac{C_T}{C_i} \cdot \frac{A^*_{i}}{A_i} + 1 \right] \quad (1)$$

After separation of the right side into a summation and the sum i , C'/C_T can be factored out to give

$$\sum_1^i \left(\frac{1}{1-F} \right)_i = \frac{C'}{C_T} \left[\sum_1^i \frac{C_T}{C_i} \cdot \frac{A^*_{i}}{A_i} \right] + i \quad (2)$$

Dividing both sides by i gives a normalized expression with an intercept of 1.

$$\left(\frac{1}{1-F} \right)_{\text{obs}} = \frac{C'}{C_T} \cdot (\text{average slope}) + 1 \quad (3)$$

The left side of the equation has been denoted $[1/(1-F)]_{\text{obs}}$ because it is an experimentally obtainable quantity, since by definition, F is the fraction of uncompleted RNA counts at any C'/C_T . The slope is denoted as an average slope because it is the sum of the slopes for all i , divided by i . The slope is a constant, because it is the product of two constants C_T/C_i and A^*_{i}/A_i ; C_T/C_i is the reciprocal of the fraction of total competing RNA consisting of species i and is fixed for any RNA mixture, and A^*_{i}/A_i is the reciprocal of the fraction of saturation for each respective species of labeled RNA in the absence of competing RNA. Since the concentration of labeled RNA is held constant throughout a competition experiment, A^*_{i}/A_i is also constant. Equation 3 is therefore correct, and is written in the standard form for a straight line with $1/(1-F)$ and C'/C_T as the variables.

In the legend to Fig. 9b (1), the equation should read " $1/A = (K/A^*C') + (1/A^*)$." The first sentence in the legend to Fig. 9 (1) should read: "Theoretical competition curves of homogeneous labeled RNA and an identical homogeneous competing RNA." In (1) on page 1296, column 3, second paragraph; the sixth sentence should read "When we solve for F , it seems that about 29 percent of the predominant radioactive species present in 8-minute RNA are absent from 80-minute RNA."

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Reference

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