NITRITE AND NITROSAMINE CONTENT OF FOODS:
UNSOLVED PROBLEMS AND CURRENT RESEARCH*

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The title of this presentation is sufficiently broad to cover a number of aspects of what we might call the "nitrosamine problem." In its own way, this problem is one of the toughest we have faced in the modern era of environmental concern and food safety, for it deals with some of the most difficult legal, analytical and predictive subproblems.

Since I have no training in the law, I will not deal in extenso with the legal aspect of nitrosamine occurrence in foods. However, we must accept a priori that most nitrosamines are carcinogens and, therefore, subject to regulation under the Delaney Amendment. We cannot change the fact of nitrosamine carcinogenicity, but a significant improvement in analytical methodology will necessitate either changing the present law or looking for a new food industry.

There are three topics on which I would like to comment more extensively:

1. The total environmental nitrite load on man.
2. The prospects for analytical methodology.
3. The prediction of nitrosamine occurrence.

Nitrite in the Environment

This is a particularly important topic, because processed meats have been singled out as a significant source. In considering the occurrence of nitrite one must also take into account sources of nitrates because of their ready conversion to nitrites by microbial systems. A listing of sources without regard to quantitative significance is given as follows:

1. Water supply (primarily nitrate)
   a. leaching from soil or geologic formation
   b. agricultural runoff
   c. contamination by sewage and/or other pollution
2. Vegetables—particularly leafy plants
3. Air pollution
4. Very high temperature food processes

5. Food additives

6. Saliva

The first category may become of increasing significance in the future. There are already numerous locations in the U.S.A. where nitrate concentrations in well water exceed 50 ppm (1) and it may prove necessary for the food plant of the future to purify its process water. As of today nitrate is a tertiary water treatment problem and there is no economic method for its removal. If nitrite load proves to have a positive correlation with nitrate occurrence, this category would by far be the most quantitatively significant source, because even low concentrations would be mathematically compensated by high consumption.

The second category is of only occasional significance, because consumption of leafy vegetables is low relative to our total food supply. However, under optimum conditions spinach can achieve nitrate concentrations as high as 1000 ppm and the accumulation of nitrate can be particularly aggravated by a soil molybdenum deficiency (1). Experiments showing lack of accumulation of nitrite in incubated foods high in nitrate (2) are unconvincing with respect to safety, because any nitrite formed might have further reacted with other food constituents.

The third category, air pollution, is probably not of great quantitative significance because of the relatively low concentrations of nitrogen oxides that are found even in highly polluted air. The fourth category, very high temperature food processes, may be of greater importance. Flame-heated spray driers have been shown to cause nitrite accumulation in non-fat dry milk (3). Any very high temperature system will oxidize the nitrogen in air to NO\textsubscript{x}, and contact of the heated air with food will leave a nitrite residue.

Foods containing added nitrites under the food additive regulations are of particular significance because of the relatively high residual nitrite concentrations compared to other incidental types of occurrence (4). Since the rate of nitrosamine formation is proportional to the square of the nitrite concentration (5), the rate of nitrosamine formation could be high in processed meats, if other conditions are favorable. A fuller discussion of this statement will appear later in this paper. The frequency and quantity of processed meat consumption in the general population is too low for this item to be considered a particularly significant part of the environmental nitrite load, but there may be individuals whose consumption pattern would generate a different conclusion.

The last category, saliva, may be of significance with respect to both nitrite load and nitrosamine formation in the oral cavity or stomach. The occurrence of nitrite in saliva has been known for many years but has heretofore been ascribed little importance. Our own work corroborates earlier findings (6) that pure ductal saliva contains no nitrite, but that whole saliva, containing large numbers of microorganisms has a relatively constant nitrite concentration. The salivary nitrite concentration for a number of individuals, measured before and after a defined meal is shown in
 TABLE 1. NITRITE CONTENT OF HUMAN SALIVA

<table>
<thead>
<tr>
<th>Individual</th>
<th>Date</th>
<th>Meal</th>
<th>Nitrite in saliva, ppm</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Before lunch</td>
</tr>
<tr>
<td>J. Tu.</td>
<td>8/6</td>
<td>canned ham, cake, soda</td>
<td>4.3</td>
</tr>
<tr>
<td></td>
<td>8/16</td>
<td>boiled ham, mortadella, soda</td>
<td>6.5</td>
</tr>
<tr>
<td></td>
<td>8/18</td>
<td>meat loaf, cake, soda</td>
<td>6.0</td>
</tr>
<tr>
<td>A.I.</td>
<td>8/6</td>
<td>meat, rice, tea</td>
<td>3.3</td>
</tr>
<tr>
<td></td>
<td>8/16</td>
<td>ham, bread, lettuce, tea</td>
<td>5.7</td>
</tr>
<tr>
<td></td>
<td>8/18</td>
<td>roast beef, lettuce, tea</td>
<td>4.6</td>
</tr>
<tr>
<td>J.F.</td>
<td>8/5</td>
<td>grilled cheese, ravioli, soda</td>
<td>1.0</td>
</tr>
<tr>
<td></td>
<td>8/16</td>
<td>peanut butter, cake, apple, soda</td>
<td>8.6</td>
</tr>
<tr>
<td></td>
<td>8/18</td>
<td>boiled eggs, celery, potato chips, coffee</td>
<td>8.7</td>
</tr>
<tr>
<td>J. Th.</td>
<td>8/4</td>
<td>luncheon meat, coffee</td>
<td>8.7</td>
</tr>
<tr>
<td></td>
<td>8/16</td>
<td>luncheon meat, coffee</td>
<td>9.4</td>
</tr>
<tr>
<td></td>
<td>8/18</td>
<td>luncheon meat, coffee</td>
<td>9.4</td>
</tr>
<tr>
<td>P.D.</td>
<td>8/6</td>
<td>corned beef, soda</td>
<td>5.5</td>
</tr>
<tr>
<td></td>
<td>8/16</td>
<td>Metrecal</td>
<td>4.3</td>
</tr>
<tr>
<td></td>
<td>8/18</td>
<td>canteloupe, salad, coffee</td>
<td>4.6</td>
</tr>
</tbody>
</table>
table 1. One can conclude from this data that neither time nor type of meal has great influence on salivary nitrite. Our experience with a large number of individuals over a period of about one year indicates a remarkable constancy for each individual with time and a concentration range of 1-10 ppm in the general population with no particular influence of sex, age, or race. The average level seems to be about 6 ppm, and considering a rate of salivary flow of 1-2 liters per day, this would imply consumption of 6-12 mg of nitrite per day. To equal the lower level of consumption from processed meat would require the daily consumption of 1/4 lb of material containing 50 ppm residual nitrite.

Analytical Methodology

A survey of analytical methodology for volatile N-nitrosamines has recently been presented by Wasserman (7), and surveys of meat products by Fazio and co-workers (8) and Telling and co-workers (9), and of a variety of food products by Sen (10). Unpublished, but well-publicized is the occurrence of nitrosopyrrolidine in cooked bacon. The conclusion of almost all investigators is that present clean-up procedures are sufficiently imperfect to require positive confirmation of all nitrosamine identifications by mass spectrometry. The complexity of this task is illustrated in the recent paper of Essigman and Issenberg (11).

All surveys to date indicate that the occurrence of nitrosamines in food appears to be an unpredictable event and that levels encountered are apt to be less than 25 ppb with a few exceptions. The arithmetic basis of the problem can be ascertained if one realizes that contamination of 1% of a weight fraction of a food item with 1 ppm of a nitrosamine will lead to an unacceptable level of 10 ppb in the well-mixed sample. The chemical nature of nitrosamine formation is such that rate is highly susceptible to specific reaction conditions (specifics to be covered in the last section). The control of reaction conditions in greater than 99% of any food item is essentially impossible given the variability of the real world. Therefore, although Fiddler and co-workers (12) have demonstrated the unlikelihood of nitrosamine formation in frankfurters under present idealized manufacturing conditions, we should not be surprised to occasionally encounter nitrosamine contaminated products in the supermarket.

It would appear therefore, that the prospects for analytical methodology lie not only in the improvement of present procedures of sample clean-up for mass spectrometry, but also in the development of methods which could be applied on a more routine basis. Such methods should be capable of demonstrating the absence of contamination and indicate those samples which require further investigation. A possible approach of this type has been suggested by Issenberg and Tannenbaum (13) and utilizes a gas chromatographic detector which is highly specific for nitrosamines. Interfering compounds such as amines can be eliminated by appropriate design of the sample preparation procedure.
It should be pointed out that even simplified procedures would be complex and costly relative to the types of analyses currently conducted by the meat industry. In addition, the nature of the contaminating process would make sampling procedures difficult to design in a statistical fashion. Finally, current methodology is suitable only for volatile compounds, while a monitoring program might ultimately have to deal also with non-volatile derivatives. The net conclusions on present analytical methodology are not encouraging and demonstrate the need for an intensified research effort. It is strongly recommended that the meat industry devote some attention to the problem of sample clean-up for various meat products.

**Prediction of Nitrosamine Occurrence**

I have already described some of the difficulties encountered in predicting nitrosamine occurrence, but this subject is worth exploring in greater detail. The essential conditions controlling the rate of nitrosation of simple amines have been described by Mirvish (5) for homogeneous solutions. The rate is proportional to the concentration of unprotonated amine and the square of the concentration of undissociated nitrous acid. These factors in turn lead to a pH-dependency profile where the rate is maximal at approximately pH 3.4 and falls off by about one order of magnitude for each one unit increase of pH.

Since a maximal rate is achieved at a fairly acidic pH, it follows from considerations of chemical equilibria that weak bases will give higher overall rates than strong bases at equal formal concentrations. This effect is demonstrated in table 2 where the reactivity of a number of groups on proteins, amino acids, and amines are given comparatively. The application of these principles to real systems should include some judicious consideration of expected concentrations in foods of the reacting amines and the individual rate constants, so that comparative rates can be calculated. One would predict from table 2 that morpholine would be readily nitrosated in protein containing foods, even at concentrations relatively low compared to the protein.

Up to this point we have considered the problems of predicting formation of nitrosamines in fairly simple systems. Foods however are fairly complex in composition both with respect to ionic environment and physical state. Unlike the simple case of reaction in a homogeneous system, one may have a system containing an aqueous phase, a solid matrix, and a lipid phase, and the distribution of reactants between these phases may influence the kinetics.

The influence of some ions on the rate of nitrosation of morpholine is shown in figure 1. Thiocyanate is a particularly potent rate enhancer (14) and is of some practical significance for its occurrence in saliva. One can see an additional complicating factor here in that the degree of enhancement is also pH-dependent. An opposing effect can be seen in table 3 where ascorbic acid, a common food constituent and a meat curing additive can be shown to suppress or prevent formation of nitrosomorpholine. Ascorbic acid is equally effective against a number of other nitrosation
TABLE 2. RELATIVE RATES OF NITROSATION

<table>
<thead>
<tr>
<th>Reactive group</th>
<th>Rate at pH 4.35, M⁻², hr⁻¹</th>
</tr>
</thead>
<tbody>
<tr>
<td>bovine thrombin</td>
<td></td>
</tr>
<tr>
<td>lysyl</td>
<td>0.6</td>
</tr>
<tr>
<td>tryptophyl</td>
<td>2.0</td>
</tr>
<tr>
<td>N-terminal leucyl</td>
<td>12.2</td>
</tr>
<tr>
<td>dimethylamine</td>
<td>1</td>
</tr>
<tr>
<td>proline</td>
<td>7</td>
</tr>
<tr>
<td>hydroxyproline</td>
<td>49</td>
</tr>
<tr>
<td>morpholine</td>
<td>200</td>
</tr>
</tbody>
</table>

Data for bovine thrombin calculated from Magnusson and Hofmann (5).

reactions (S. Mirvish, personal communication). Could there be other food constituents which are equally suppressive of which we are currently unaware? A different type of effect is seen in table 4 where the formation of nitrosomorpholine is shown in a 2-phase oil-water system. If no oil were present in the system, the amount of nitrosomorpholine formed at the end of 10 minutes would be a normalized value of 100. Due to the partitioning of nitrite into the oil phase this rate should be reduced as the volume of the oil phase is increased assuming no reaction occurs in the oil phase. The calculated rate using these assumptions is shown under "expected value" in table 4. The actual rate of synthesis of nitrosomorpholine is higher than expected as seen by the figures under "actual value." This difference is currently unexplained, but might be due either to a reaction in the oil phase or at the oil-water interface.

One might conclude from this discussion that the study of rate processes is not useful for prediction of occurrences of nitrosamines. This is not the intention. Rather one must be cautious in application of laboratory results to foods in the marketplace. Ultimately, we might know enough to make useful predictions and even now we can make useful recommendations concerning prevention.
### TABLE 3. EFFECT OF ASCORBIC ACID ON NITROSOMORPHOLINE FORMATION

<table>
<thead>
<tr>
<th>ascorbic acid concentration, ppm</th>
<th>NM formed (30 min.)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>14</td>
</tr>
<tr>
<td>120</td>
<td>11</td>
</tr>
<tr>
<td>720</td>
<td>approx. 0</td>
</tr>
</tbody>
</table>

pH 4.0, 25°C, nitrite and morpholine 600 ppm.

### TABLE 4. FORMATION OF NITROSOMORPHOLINE IN A TWO-PHASE SYSTEM

<table>
<thead>
<tr>
<th>oil:water, volume ratio</th>
<th>expected value</th>
<th>actual value</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>54</td>
<td>49</td>
</tr>
<tr>
<td>2</td>
<td>31</td>
<td>35</td>
</tr>
<tr>
<td>3</td>
<td>20</td>
<td>38</td>
</tr>
<tr>
<td>4</td>
<td>14</td>
<td>32</td>
</tr>
<tr>
<td>5</td>
<td>10</td>
<td>26</td>
</tr>
</tbody>
</table>

Nitrite and morpholine reacted 10 minutes at 25°C, pH 3.5; the expected value is calculated on the basis of no reaction in the oil phase and $K_{D} = 4$ for nitrite and 5 for nitrosomorpholine.
INITIAL RATE OF NITROSATION OF MORPHOLINE WITH NITRITE.

pH DEPENDENCY AND INFLUENCE OF CERTAIN IONS.

- O Nitrite (5 mM), Morpholine (10 mM), KSCN (2.5 mM)
- □ Nitrite (5 mM), Morpholine (10 mM), KBr (100 mM)
- △ Nitrite (5 mM), Morpholine (10 mM)

25 °C

The graph shows the initial rate of nitrosation of morpholine with nitrite as a function of pH.
Conclusions

I have tried in this paper to bring out some of the factors which influence nitrite and nitrosamine occurrence in foods and also some of the accompanying analytical problems. It would appear that there are a number of significant sources of nitrite in the environment other than that added to meat products. The most significant problem for meat products is the opportunity for nitrosamine formation presented by a relatively long storage time and our inability to maintain control of the reaction conditions of 100% of the product. Routine sampling and analysis of production lots is beyond present economic and technical feasibility. Although we cannot now predict the occurrence of nitrosamines with any certainty, research is the best present hope for the solution of the overall problem.

Acknowledgements

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REFERENCES


# # # # #

M. E. BAILEY: Thank you, Bob, for chairing this part of the program on nitrosamines. Do you have further questions concerning this problem?

R. P. DUDLEY: I would like to call on Ned Draudt to say a few words about the use of nitrite in meat processing.

H. N. DRAUDT: The major question is the efficacy of nitrites as an additive, what it does to the product. I am sure it does much more than preserve color. For instance, what does it do to the flavor? There are few clues to what happens when we use nitrite and nitrate.

M. E. BAILEY: Thank you very much for those comments, Ned. We are extremely fortunate to have a representative from three of the four laboratories that have the capacity of analyzing multicomponent mixtures of nitrosamines. I am sure these fellows do not always agree as to what method is most suitable for analyzing nitrosamines in a given situation. I would now ask if any member of the panel has a question for another member of the panel.

W. J. AUNAN: I have a question for Dr. Fiddler. What do you feel is the real importance to your discussion that certain amino acids contribute to nitrosamine formation?

W. FIDDLER: Woody is referring to data on one of my slides that indicate that certain amino acids could react with nitrite at high levels to form nitrosamines. I should point out that analytical procedures used in the studies cited are questionable. The methods used do not confirm the presence of nitrosamines unequivocally and this is the real question. The methods are not really valid for this type of conclusion.

M. E. BAILEY: Are there other questions by panel members?

W. J. AUNAN: Dr. Fazio referred to the level of sensitivity in nitrosamine analysis that was recognized. I would like for him to repeat that statement and explain why this level was selected.

T. FAZIO: We presently can confirm the presence of 10 ppb nitrosamine in samples. The real answer is that it is assumed that 10 ppb would be a relatively safe level of nitrosamine and this is why we are interested in this level.
S. R. TANNENBAUM: Below 10 ppb you are about as low as you can go and still identify the compounds with mass spectrometry. So, this is the level of positive confidence from an analytical point of view.

W. J. AUNAN: Concerning the legal status the use of nitrite is not governed by the Delaney Amendment, but has higher sanction.

S. R. TANNENBAUM: I wasn't referring to nitrite, but to nitrosamines.

W. J. AUNAN: But if nitrite is a precursor for nitrosamine formation and we add it where do we stand?

S. R. TANNENBAUM: I understand that the law reads in such a way that it covers any food additive should it be converted to a carcinogen.

M. E. BAILEY: The major question concerns whether or not the Delaney Amendment is applicable to the use of nitrite as a meat additive where it is a reaction product that apparently is carcinogenic and not the additive per se. I would like to ask Dr. Fazio a question. If we assume that all cured meat samples contained from 10 to 25 ppb dimethyl nitrosamine, can you suggest what this might mean in terms of human health.

T. FAZIO: Since I am not a toxicologist, I can't answer your question, but the levels we are working with are quite low.

M. E. BAILEY: The lowest levels of nitrosamine used in toxicological studies is 75 μg/kg/day for the rat. Have you seen data regarding use of nitrosamines at lower levels? It appears to me that these values are quite high compared to quantities of nitrosamines actually found in meat products.

S. R. TANNENBAUM: There have been but few studies carried out where low levels were used to measure toxicity. The study of the influence of additives on human health is a statistical problem. The toxicologist is interested in finding positive results; this is how he gets his research funds. Support for finding of negative results will come from the Meat Industry who are really interested in negative results on this problem.

It appears that from most reasonable observance from study of the biological data that it would be difficult to conceive that 10, 50 or even 100 ppb nitrosamine would be hazardous considering the frequency of intake of the product concerned.

M. E. BAILEY: That is exactly the point that I was trying to make.

FROM THE FLOOR: I would like to know the nitrite content of fresh meat. Does fresh meat contain nitrite?

W. FIDDLER: I am not sure, we do not have these data.

FROM THE FLOOR: Dr. Tannenbaum discussed the high level of nitrosamine formation at pH 3.5. What happens at pH 6.5, the pH of meat?
S. R. TANNENBAUM: It is difficult to measure the rate constant at pH 6.5. One has to run the reaction at this pH for a long time and its difficult to maintain nitrite concentration. The rate at any pH is approximately predictable, but the results have to be confirmed by experimentation.

FROM THE FLOOR: Have compounds besides ascorbate been found that would expedite the nitrite reaction? Can you predict the action of compounds that would have the same effect?

S. R. TANNENBAUM: Ammonium sulfamate and NADPH are two that might be useful. Anything that could compete for nitrite might be effective. There are many such compounds, but ascorbate is the most useful.

M. E. BAILEY: Is the major mechanism of the ascorbate reaction concerned with the reduction of nitrite to nitric oxide?

S. R. TANNENBAUM: Not completely.

T. BLUMER: In processing meats, the faster the reaction goes, the better the situation should be as far as nitrosamine formation is concerned. Faster processing would prevent amine formation and at the same time deplete the available nitrite.

S. R. TANNENBAUM: It would be desirable to prevent amine formation, but I thought it was also desirable to maintain a certain level of nitrite.

T. BLUMER: Assume you have the proper pH for nitrosamine formation (pH 4.0). Would it be desirable to reduce the pH to say pH 3.5 to help reduce nitrite content?

S. R. TANNENBAUM: The low pH would be more detrimental as far as nitrosamine formation is concerned.

T. BLUMER: Is there a desirable relationship concerning conditions for removing nitrite and those required for nitrosamine formation?

S. R. TANNENBAUM: In a system as complex as meat, nitrite is lost due to many different reactions, depending on pH. I think that a higher pH would be more desirable because reaction between nitrite and other compounds as pyrimidines and ascorbate would be favored. It would be desirable to maintain the nitrite at a certain low level.

C. E. ALLEN: What is the contribution of other materials such as packaging materials to nitrosamine formation?

S. R. TANNENBAUM: This is a possibility, but we haven't looked into it. This is a different ball game.

J. D. SINK: What does the future hold concerning regulation in the use of nitrite? Will the presence of nitrosamine be under surveillance in the future?
R. P. DUDLEY: There is residual nitrite in processed meat. I can assure you that all involved are trying to reduce nitrite residues in cured meat. We are attempting to find the lowest level that will maintain quality (flavor, color, etc.) and yet leave a minimum residue in meat per se.

T. FAZIO: The Food and Drug Administration is cooperating with the Meat Industry. We are trying to determine if there really is a problem.

W. J. AUNAN: I would like to emphasize the importance of maintaining sufficient nitrite to prevent growth of clostridium botulinum as a health hazard.

W. FIDDLER: Some of the work we are doing might be helpful in determining future direction. I agree with the others that much work needs to be done and that it would be desirable to reduce the levels of residual nitrite, but only to levels that would maintain quality.

FROM THE FLOOR: I understand that Dr. Brown of the University of California is studying the interaction of pyrimidines and myoglobin, do you visualize this as a possible substitute for nitrite?

W. J. AUNAN: We are looking for a substitute, but you need a very dynamic compound to compete with nitrite.

C. E. ALLEN: I would like a comment from someone in the Meat Industry concerning why we need to use nitrate in our rapid processing procedures for curing meat.

W. J. AUNAN: The use of nitrate is a hang-up in the sausage makers mind, but its use in individual products must be examined.

H. N. DRAUDT: Some nitrite is needed to help extend the shelf life of some products. We have found it useful for products that will be stored for long periods of time. However, the extended shelf life may not be worth it.

R. P. DUDLEY: This is because you have reducing bacteria present, is it not?

H. N. DRAUDT: Possibly, but nitrate does extend shelf life, but I am not praising its use.

E. WIERBICKI: Nitrate, particularly in combination with ascorbate helps prevent discoloration of cured meat color. We have measured the degree of fading of nitrite alone compared with a mixture of nitrite and nitrate. The combination treatment appears to prolong desirable color.

FROM THE FLOOR: Would KNO2 be as useful in the curing reaction as NaN02?
S. R. TANNENBAUM: It makes no difference which you use since both are salts of strong bases and a strong acid.

M. E. BAILEY: I would like to thank the speakers for their excellent presentations and for their contributions during this discussion. I would also like to thank Bob Dudley for his efforts in helping to organize this program; it has been a successful one.

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C. E. ALLEN: Good Morning. Glad to see those of you who are here this morning. We have two programs. The first one is Processed Meats and after that we have a concurrent session which I'll say more about a little later before our break. There are a couple of announcements. For those of you who are speakers today or tomorrow, I'd like to remind you that we would like to have the copies of your talks turned into Dr. Sherman and if any of you yesterday did not turn yours in we would like to have them today. Our first program committee which is reporting is Processed Meats Committee and this is chaired by George J. Cocoma of Peter Eckrich and Sons and we will again have what we consider to be a very appropriate topic this morning and this has to do with nutritional labeling which you've heard a lot about this last year. We feel that we have two very appropriate people to speak to you about this topic. So I'll turn the time over to George to carry on with his committee's program. George.

GEORGE COCOMA: Most of you have read that on March 30, 1972, the FDA published in the Federal Register proposed regulations dealing with nutritional labeling. We have with us today the person under whose direction this proposal was written, Dr. Ogden Johnson. He will discuss nutritional labeling from the Government's point of view, and will be followed by Dr. Stewart from Gerber & Company, discussing the same subject from an industry point of view.

Dr. Johnson is the Director of the Division of Nutrition, Office of Science, Bureau of Food and Drug Administration. Ogden was educated at the University of Illinois, received his Ph.D. Degree in Food Science in 1956. Prior to his present position with the Government, he was employed as Assistant Secretary of Council on Food and Nutrition, American Medical Association. Before that, he was on the nutritional program of the Mental Health Administration. Dr. Johnson is a member of numerous professional societies, including the Institute of Food Technology, American Institute of Nutrition, American Public Health Association and many others. So please welcome Dr. Ogden Johnson.