

Special attention was given to comparing the phytotoxicity of a straight-chain olefin with one wherein a methyl group was attached at the double bond, and cis and trans forms of a given material. For this purpose comparisons were made between 1-hexene and 2-methyl-1-hexene and between *cis*- and *trans*-3-hexene. The addition of a methyl group at the double bond markedly reduced plant damage and it is suggested that the methyl group interfered in some way with phytotoxicant formation. Changing from *cis* to *trans* forms did not alter level of phytotoxicity.

Oxidant damage was produced with about 2 p.p.m. of acrolein alone. While damage was not severe, it was definite and indistinguishable from the underside bronzing typical of oxidant damage. Haagen-Smit *et al.* (4) had shown that acrolein produced atypical damage on some of the test plants used.

**Eye Irritation.** No eye irritation was obtained with any ozone-hydrocarbon mixture that was appreciably

higher than that reported for clean air. Neither was irritation reported for ozone alone, even at high concentrations. For comparison of acrolein (a known lachrymator) with carbon-filtered air, the frequencies for none, medium, and severe irritation resembled a Poisson distribution; one of the appropriate transformations is  $y = \sqrt{x+1/2}$  (7).

#### Acknowledgment

Thanks are extended to Kenneth Greenlee, The Ohio State University, for preparing and supplying certain hydrocarbons, and to C. H. Hine and Frank Ivanhoe, University of California Medical School, San Francisco, for supplying the method of acrolein analysis.

#### Literature Cited

- (1) Arnold, W. N., *Intern. J. Air Pollution* **2**, 167-74 (1959).
- (2) Darley, E. F., Stephens, E. R., Middleton, J. T., Hanst, P. R., *Proc. Am. Petrol. Inst.* **38** (III), 313-23

(1958); *Intern. J. Air Pollution* **1**, 155-62 (1959).

- (3) Faith, W. L., Renzetti, N. A., Rogers, L. H., "Eye Irritation from Irradiated Auto Exhausts," Rept. 22, Fourth Technical Progress Report, Air Pollution Foundation, San Marino, Calif., 1958.
- (4) Haagen-Smit, A. J., Darley, E. F., Zaitlin, Milton, Hull, Herbert, Noble, W. M., *Plant Physiol.* **27**, 18-34 (1952).
- (5) Middleton, J. T., Darley, E. F., Brewer, R. F., *Proc. Am. Petrol. Inst.* **37** (III), 184-91 (1957); *J. Air Pollution Control Assoc.* **8**, 9-15 (1958).
- (6) Scott, W. E., Stephens, E. R., Hanst, P. L., Doerr, R. C., *Proc. Am. Petrol. Inst.* **37** (III), 171-83 (1957).
- (7) Snedecor, G. W., "Statistical Methods," 5th ed., pp. 315-16, Iowa State College Press, Ames, Iowa, 1956.

Received for review February 5, 1960. Accepted April 8, 1960. Division of Petroleum Chemistry, 136th Meeting, ACS, Atlantic City, N. J., September 1959. Research sponsored by the Project Advisory Committee IX, Smoke and Fumes Committee, American Petroleum Institute.

## MINERAL SUPPLEMENTS

### Copper, Nickel, and Cobalt Content of Oyster Shells

R. S. YOUNG

2012 Paul's Terrace, Victoria, B. C., Canada

The copper, nickel, and cobalt content in oyster shells has been found to be 33, 30, and 1 p.p.m., respectively. These quantities of copper and cobalt are too low to be significant in correcting soil and plant deficiencies by the application of oyster shells as liming material, but the cobalt present in oyster shells might be beneficial in poultry nutrition. Unlike copper, nickel is present in higher quantity in the shell of oysters than in the edible portion. The concentration of copper and cobalt from sea water to oyster shells is about the same, whereas nickel appears to be accumulated to a greater degree.

OYSTER SHELLS play a minor role in agriculture as a mineral supplement in poultry nutrition and occasionally as a soil additive in the form of lime or limestone. There does not appear to be a recorded observation on the cobalt or nickel content of oyster shells, and only a single reference to the quantity of copper in this material has been found.

A composite 10-pound sample of shells of the common oyster, *Ostrea virginica*, gathered on several occasions from different locations off the coast of Prince Edward Island, Canada, was thoroughly freed from organic matter by scrubbing with a stiff-bristle brush in fresh water. The shells were rinsed ten times in distilled water, dried overnight at 105° C., crushed, and pulverized to approximately 65 mesh. From this bulk sample a final 250-gram analytical sample was

obtained by riffing, and this was pulverized to -100 mesh. The washing and drying procedures used on these shells left such small quantities of organic matter and moisture that their presence had no effect on the concentration of metals in the parts per million range.

Standard colorimetric procedures were employed to determine nickel as nickel dimethylglyoxime in alkaline solution, and cobalt by the Nitroso-R salt procedure. After the addition of dimethylglyoxime and removal of nickel and cobalt by extraction with carbon tetrachloride, copper was determined with sodium diethyldithiocarbamate (5). The following results, in parts per million, were obtained: copper, 33; nickel, 30; and cobalt, 1.

The quantities of these metals found in sea water by different investigators show

considerable variation. This is not surprising when one considers the influence of land drainage, shore rock composition, currents, and other factors affecting the composition of sea water, as well as the difficulties in sampling and analysis for trace elements. The range of values published in recent years for copper, nickel, and cobalt in sea water is illustrated in Table I.

The figures reported by Young, Smith, and Langille (7) for copper and cobalt are of particular significance, as their samples were likewise taken from the sea around the Atlantic provinces of Canada. The average content of copper found by Young, Smith, and Langille agrees well with the mean compiled by Vinogradov (10), and their average cobalt value of 0.5  $\gamma$  per liter shows good agreement with the averages listed by

**Table I. Copper, Nickel, and Cobalt Reported in Sea Water**

| Reference | Micrograms per Liter |         |           |
|-----------|----------------------|---------|-----------|
|           | Cu                   | Ni      | Co        |
| (2)       | <3000                | 1.5-6   | <0.3      |
| (4)       | 4-10                 | 0.1-0.5 | 0.1       |
| (6)       |                      | 0.7-0.8 | 0.38-0.67 |
| (7)       |                      | 2       |           |
| (8)       |                      |         | 0.23-0.32 |
| (10)      | 20                   | 3       | 1         |
| (11)      | 13-22                |         | 0.33-0.67 |

other investigators. If the respective values of 18, 2, and 0.5  $\gamma$  per liter are accepted for copper, nickel, and cobalt in sea water, results of the present work indicate that copper and cobalt are concentrated in the oyster shell to nearly the same extent,  $1.8 \times 10^3$  and  $2 \times 10^3$ , respectively. The concentration of nickel, however, from sea water to shell is appreciably higher at  $1.5 \times 10^4$ .

Copper in the edible portion of oysters has been reported by various workers to vary over the wide range of about 5 to 2000 p.p.m. in the living matter. A fair average of the compiled data of Vinogradov for *Crassostrea virginica* would be approximately 1100 p.p.m. of copper. As the edible portion of oysters contains about 87% water, it is evident that on the dry basis this part, even for the lower ranges of copper recorded in the literature, contains more of this element than found in the shell in the present study. The single reference to copper in oyster shells which was noted (9) gave a value of 25 p.p.m., which is close to the findings reported here.

Figures quoted by Vinogradov on nickel in gills, mantle, and hepatopancreas of *Crassostrea angulata* are 1.3, 0.9, and 1.0 p.p.m., respectively, and a value for nickel in the edible portion of

*Crassostrea sp.* is given as 1.7 p.p.m. No reported reference to nickel in the shell was found. The author's value of 30 p.p.m. indicates that, unlike copper, there is considerably more nickel in the shell of oysters than in the edible portion.

Cobalt does not appear to have been determined in either edible portion or shell of oysters, though the element has been recorded in living matter and various organs of a few genera of Mollusca (10). The limited data indicate that, in general, cobalt is present in smaller quantity than nickel and that the nickel-to-cobalt ratio is about the same as that found in sea water.

Although the cobalt content of oyster shells is higher than that of most plant products, it is far too low to serve as a source of this element in cobalt-deficient areas by the application of lime derived from oyster shells. In regions of the world where maintenance of health in ruminants requires the addition of a cobalt supplement to fertilizer or lime, 2 pounds of the commercial sulfate per acre will last for 3 to 5 years. In other words, about 0.1 pound of cobalt per year will be needed, whereas the application of even 5 tons per acre of oyster shells would furnish only 0.01 pound of cobalt.

The copper content of oyster shells is probably too low for it to exert any significant effect in correcting a deficiency of this element in plants and soils. In various soils throughout the world where copper deficiencies for livestock, citrus fruits, or other crops occur, an application of commercial sulfate varying from 5 to 50 pounds per acre has been employed, and the effect usually lasts at least 3 years. A minimum of about 1 pound of copper per acre per year is thus indicated, whereas even 5 tons per acre of oyster shells would furnish only about 0.3 pound of copper.

There have been several reports on the beneficial effect from cobalt additions to poultry rations (7, 3). From 1 to 12 p.p.m. of cobalt in the feed have resulted in increased growth. It is possible, therefore, that the minute quantities of cobalt in oyster shells may have significance in poultry nutrition.

#### Literature Cited

- (1) Berzins, J., Rozenbachs, J., *Latvijas PSR Zinātņu Akad. Vēstis* 1953, No. 9, 39-46.
- (2) Black, W. A. P., Mitchell, R. L., *J. Marine Biol. Assoc. United Kingdom* 30, 575-84 (1952).
- (3) Burns, M. J., Salmon, W. D., *J. Agr. Food Chem.* 4, 257-9 (1956).
- (4) Harvey, H. W., "Recent Advances in the Chemistry and Biology of Sea Water," Cambridge Univ. Press, Cambridge, Eng., 1945.
- (5) Hopkin and Williams, Ltd, Chadwell Heath, Eng., "Organic Reagents for Metals," 5th ed., p. 175, 1955.
- (6) Ishibashi, M., *Records Oceanog. Works Japan [N.S.]* 1, No. 1, 88-92 (1953).
- (7) Laevastu, T., Thompson, T. G., *J. conseil. Conseil permanent intern. exploration mer* 21, 125-43 (1956).
- (8) Thompson, T. G., Laevastu, T., "Determination and Occurrence of Cobalt in Sea Water," Alaskan Science Conference, 1956.
- (9) Tressler, D. K., "Marine Products of Commerce," p. 529, Chem. Catalog Co., New York, 1923.
- (10) Vinogradov, A. P., "The Elementary Chemical Composition of Marine Organisms," pp. 341, 352, 364, Sears Foundation for Marine Research, New Haven, 1953.
- (11) Young, E. G., Smith, D. G., Langille, W. M., *J. Fisheries Research Board Can.* 16, 7-12 (1959).

Received for review March 14, 1960. Accepted August 7, 1960.

## DIETARY PROTEIN VALUES

### Complete vs. Total Protein in the Evaluation of Diets

THE PROTEIN CONTENT OF FOODS is customarily determined by multiplying the nitrogen content of the food by one of several factors which range in magnitude from 5.7 to 6.66. The misleading impression that such factors may impart concerning the real protein content of mixed foods has been discussed (5). However, even if the protein content is determined by more

adequate means, the result may or may not describe the value of this protein for nutritive purposes. The concept of "complete protein" was introduced (7) to define more clearly the quantity of protein which would be available to the organism for repair and synthesis of tissue. Complete protein is, by this definition, that fraction of the total dietary protein which is completely utilized

HARTLEY W. HOWARD and CLIFFORD D. BAUER

Borden Special Products Co., Division of the Borden Co., New York, N. Y.

RICHARD J. BLOCK

Boyce Thompson Institute for Plant Research, Inc., Yonkers, N. Y.

for anabolic purposes. As the pattern of essential amino acids required by an animal appears to depend on its physiological state (growth, tissue repair, pregnancy, lactation, adult maintenance), the amino acid composition of the complete protein will vary with the animal's requirements. In this study, complete protein is represented by the pattern of essential amino acids needed